8th International Soil Science Congress on "Land Degradation and Challenges in Sustainable Soil Management"

May 15 - 17, 2012 Çeşme - İzmir / TURKEY

PROCEEDINGS

Volume-II

Policies for Soil Health Management in Agriculture
Soil and Water Pollution

EDITORS
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This book of proocedings has been prepared from different articles sent to the congress secretary only by making some changes in the format. Scientific committee regret for any language and/or aim-scope

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PREFACE I

The 8th International Congress on Soil Science was held at the Altınyunus Hotel in Çeşme, Izmir, Turkey, from May 15th to 17th, 2012. The theme for this year was “Land Degradation and Challenges in Sustainable Soil Management”. The congress was organized by Ege University’s Department of Soil Science and Plant Nutrition, (Agricultural Faculty), and The Soil Science Society of Turkey (SSST). The congress also hosted the 6th International Conference on Land Degradation (ICLD).

The organization of International Soil Science Congresses is a long established custom for the SSST.
The 8 congresses held so far are listed below:

1998, Izmir:	1st International Congress on Soil Science
2000, Konya:	2nd International Congress on Soil Science
2002, Çanakkale:	3rd International Congress on Soil Science, organised by Onsekiz Mart University
2004, Erzurum:	4th International Congress on Soil Science, organised by Atatürk University
2006, Şanlıurfa:	5th International Congress on Soil Science, organised by Harran University
2008, Aydın:	6th International Congress on Soil Science, organised by Adnan Menderes University
2010, Samsun:	7th International Congress on Soil Science, organised by Ondokuz Mayis University
2012, Izmir:	8th International Congress on Soil Science, organised by Ege University

For this 8th International Congress we received more than one thousand abstracts from 54 countries worldwide. After a rigorous evaluation process, 655 of these were chosen for presentation either as seminars or posters during the congress.

There were two plenary lecturers: Prof. Richard Dick from Ohio State University, spoke on soil microbiology and Prof. Dr. Sergei Shoba from the Faculty of Soil Science, Lomonosov Moscow State University, talked about the challenges of soil degradation in arid areas.

The papers have been organized into five volumes according to topics for the Congress Proceedings Book The on-line version of these volumes is accessible at: http://www.soilcongress.ege.edu.tr.

We would like to take this opportunity to express our thanks to all the authors for their efforts in the preparation of these excellent contributions.

Yusuf KURUCU, Ph.D in Soil Science
Chair, The 8th ISSC 2012

PREFACE II

Today the world community has recognized the importance of sustainable use of soil, which is one of the key life-supporting components on the earth. As suggested in UN Conference on Environment and Development (UNCED) in 1992, soil degradation caused by over exploitation of fragile resources and misuse of marginal areas, degrease of potential agricultural areas by sealing, uneven distribution of potentially cultivable areas, declining trends in per capita food production, lack of adaptation of improved technologies by subsistence farmers, non-availability of essential off-farm input to resource poor farmers, and problems such as soil-mining should be considered in soil resources management plans.

Action programs are needed to protect and improve soil health by developing thematic strategies toward protecting soils from numerous of threads such as erosion, decline of organic matter content and biodiversity, sealing, soil salinizaiton, alkalinizitaion, flooding, and many others. Function of soils in environment in relation to human activates should be understood well to manage the soils without declining their quality. Unique role of a specific soil type for environment and human activities should be considered in managing soils to secure soil health, water quality, and food and fiber production for future generations. The evidence that we all depend on the thin layer of earth should be articulated to the people with no knowledge of soil and its importance. In addition, high quality technical information should be available and ready for growers, decision makers, government agencies, and so on for an effective use of science and technology in soil management.

This congress was organized to discuss issues in "land degradation and challenges in soil management". Interactions among soils, land degradation, and desertification were discussed and importance of soils for a better environmental quality and food security was stressed in the three-day congress. Poster and oral presentations covered a large spectrum of subject areas; including computer modeling, digital mapping, and new techniques and technologies used in data mining, decision making, and other related areas.

I trust that this proceeding will make a vigorous contribution to theoretical and practical soil science, and generate a prolific interest for appreciation of soil's importance to public well-being. I thank Organizing Committee and all worked and appreciate them for this high quality work.

Sabit ERŞAHIN, Ph.D in Soil Physics
President of Soil Sicence Society of Turkey
President of The Eurasian Federation of Soil Science Societies

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VOLUME II

Policies for Soil Health Management in Agriculture
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Plenary Lectures

Native Shrubs *Piliostigma reticulatum* and *Guiera senegalensis* as Companion Plants: Rhizosphere Hydrology and Microbiology in Relation to Crop Productivity in the Sahel

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The Sahel is experiencing landscape and soil degradation that reduces food and economic security of rural, underprivileged communities that depend on ecosystem services. The Parkland system of randomly distributed trees is an approach to address these challenges, but trees are slow growing and can compete with crops for water and nutrients. Conversely, two native shrubs, *Piliostigma reticulatum* and *Guiera senegalensis,* coexist in farmers' fields throughout the Sahel and until recently have largely been overlooked. It is well established that organic matter input to the soil is critical for improving soil quality and optimizing nutrient and water efficiencies, and ultimately crop productivity in the Sahel. Various non-indigenous vegetative systems have been proposed for the Sahel, but with limited adoption in cropped fields. Consequently, these two shrubs being indigenous and already found in farmers' fields to varying degrees, hold potential to meet these challenges. Unfortunately, the current management of spring coppicing and burning prior to cropping, is not utilizing this organic matter effectively. There has been very little research on how to ecologically or agronomically manage these shrubs. *Therefore, the global objective was to determine the unrecognized ecological function of these shrubs in agroecosystems of Senegal that are representative the Sahel*. To test these hypotheses, our team over the last 5 years, has conducted extensive field based investigations in the Peanut Basin of Senegal that included: ground surveys and remote sensing to determine the landscape levels of shrub C and biomass; hydrology and water relations between shrubs and crops; rhizosphere microbiology; residue decomposition; N and P cycling in relation to crops; and crop productivity. The project, funded by US National Science Foundation, graduated 4 PhD students and 3 post docs. The major findings in Senegal are that:

- shrubs are by far the largest source of organic matter on the landscape in cropped fields
- shrubs increase soil quality
- decomposition rates are rapid enough to allow non-thermal residue management
- shrub roots perform hydraulic lift by moving water from wet sub- to dry surface-soils that appears to drive microbial processes year around and assist crops through drought periods

- shrub roots recharge groundwater in the rainy season, reducing runoff and conserving water
- shrub rhizospheres promote microbial diversity and may harbour beneficial microbes
- intercropped shrubs do not compete with crops and actually stimulate yield by >50%
- repeated application of these low quality residues (in absence of live shrubs) begins increasing yields after 2 years.

Keywords: *Piliostigma reticulatum, Guiera senegalensis***,** soil remediation, Sahel, crop productivity

Soil Degradation Challenge in Arid Areas: The Role of Soil Databases and Soil Information Tools

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Abstract
Soil degradation is the biggest challenge to food security in the new millennium. Climatic change and increasing human pressure on ecosystems results in the extensive degradation of landscapes, especially in dry areas. Desertification is the major problem for agricultural development and for providing food for population in many regions of the world, including Eurasia. Both the south of Russia and neighboring suffer desertification, which consists of various complex processes, including the processes of soil degradation, such as salinization, compaction, organic matter loss etc.
In the context of soil conservation and protection we should develop scientific tools for effective prognosis of short-term and long-term soil degradation under scenarios of different land use and different paths of climatic changes. A reliable prediction is possible only on the basis of the most complete information on actual soil resources. The first component of soil information is the presence of soil databases, which allow spatial interpolation, data mining, and spatial and temporal modeling. The necessary feature of these databases should be a user-friendly format of data storage that allows multiple data management.
Our actual activities are aimed at the development of soil data storage system, the tools for effective soil data management, and the collection of legacy soil data. The main issues to be solved are poor compatibility of data in different regions, imprecise coordinates of soil pits in previous surveys, and low response from regional administrative bodies. However, with increasing desertification the attention to soil data should increase.
Keywords: land resources, sustainable development, risk evaluation, Russia, Central Asia.

Introduction
Despite of the impressive progress in technology, modern civilization is still completely based on agricultural production. The entire population of the Earth depends on the products obtained from soil either as crops or as forage. The development in agricultural technology is impressive, and now we can produce much more yields than few decades ago from the same area (Shoba, 2009). However, the population is growing rapidly, and we do not know, if the increase in productivity can compensate the increasing need in food and a loss of productive lands. The loss of productive arable lands is a widespread process that takes place all over the world due to various reasons, such as urbanization, soil erosion, desertification and many others. Some of these reasons are universal, and some of them are landscape-specific. One of the most vulnerable zones is the arid belt of Eurasia that is strongly affected by a complex of degradation processes associated with desertification.
Desertification is the major challenge for land management in many Eurasian countries. In the most severe cases we have to make difficult decisions on land use change and even change the whole strategy of the development of national economy. Soil degradation leads to food insecurity, poverty, and thus social instability. Even if a state has stable economy based on industrial production or mining industry, agriculture is a life-spring for millions of people who inhabit dry areas. For their sustainable living we should maintain a certain level of soil productivity, even taking into account progressing climatic changes.
For maintaining soil productivity and for successful planning of soil improvement and management we should have a complete set of data that characterize soil and water quality. In this short review we tried to show the state-of the art with soil resources information in Russia and neighboring Central Asian countries, and to outline the perspectives for future development.

Soil information in Russian drylands

Though Russia is traditionally considered to be a cold and humid country, its vast territory also possesses some areas with dry climates, where soils run the peril of drought (Fig. 1). These areas are localized mainly along the southern border of Russia, both in the European and Asian parts. The soils in these dry areas are affected by the degradation processes, which are common for most arid regions, such as salinization, alkalinization, wind erosion, and desertification (that is understood as a complex combined process). A comparison of the maps of drought probability and the distribution of the processes of soil degradation (Fig. 2) shows that there is a good agreement between dry soil regime and the distribution of the specific degradation processes mentioned above. Speaking more specifically, such process as salinization is widespread mostly in the southern part of European Russia, in Dagestan and Kalmyk Republics and in Volgograd region. In Asia most of saline soils are found in Novosibirsk, Omsk, and Altay regions. Alkaline soils are common in Kalmyk Republic and Volgograd region in European part of the country, and in Novosibirsk and Omsk regions in Siberia. Though desertification is not very common in Russia, being active only at 7% of the national territory, it strongly affects agricultural production. This negative process is especially dangerous in Kalmyk and Dagestan Republics, Astrakhan, Volgograd and Rostov regions (with lesser extent in Orenburg and Saratov regions) in European Russia, and in Altay and Omsk regions and Tuva, Khakassia, and Buryatia Republics in Siberia.

The processes of wind erosion are very active in dry areas. Totally 8.4% of the agricultural lands in Russia are affected by deflation; these are localized mainly in Stavropol and Krasnodar regions and in Kalmyk and Dagestan Republics (European part of Russia) and in Altay region and Khakassia Republic (Siberia).

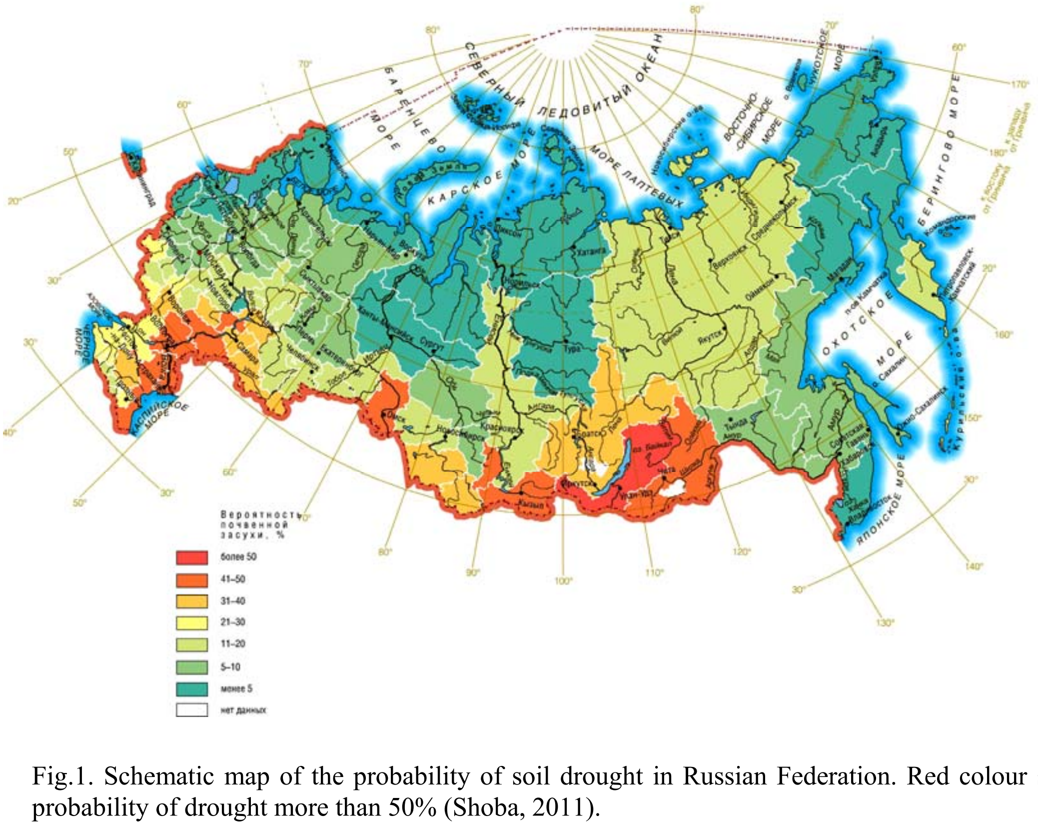

Fig.1. Schematic map of the probability of soil drought in Russian Federation. Red colour shows the probability of drought more than 50% (Shoba, 2011).

The other negative process usually associated with desertification is the loss of soil organic carbon. In the arid regions of Russia the balance of soil organic carbon is strongly negative that shows that the organic matter decomposes quickly in these soils (Fig. 3). Though arid soils usually have a positive balance of carbon from the point of view of global carbon cycling due to accumulation of carbonates, the loss of organic matter is a negative process resulting in the degradation of soil physical and chemical properties and in the decline of soil fertility.

In the context of soil conservation and protection we should develop scientific tools for effective prognosis of short-term and long-term soil degradation under scenarios of different land use and different paths of climatic changes. A reliable prediction is possible only on the basis of the most complete information on actual soil resources.

During the second part of the 20th century the soils of Russian drylands have been extensively studied, that allowed the development of soil and land evaluation maps of various scales, starting from the most detailed (1:10,000) to the most general (Shoba et al., 2010). These maps are valuable sources of information, but they have certain disadvantages. First, particular soil profiles seldom have exact coordinates. Second, some data are already outdated after several decades of anthropogenic transformation of soils. Third, the major part of this information existed only in paper form, and some important blocks of information are already lost.

Actually for successful soil data management, the first component of soil information is the presence of soil databases, which allow spatial interpolation, data mining, and spatial and temporal modeling (Panagos et al., 2012). Leaving apart an urgent need for updating soil data, the closest and the most reliable task is the development of soil database for Russia and possibly for neighboring countries. The necessary feature of this database should be a user-friendly format of data storage that allows multiple data management.

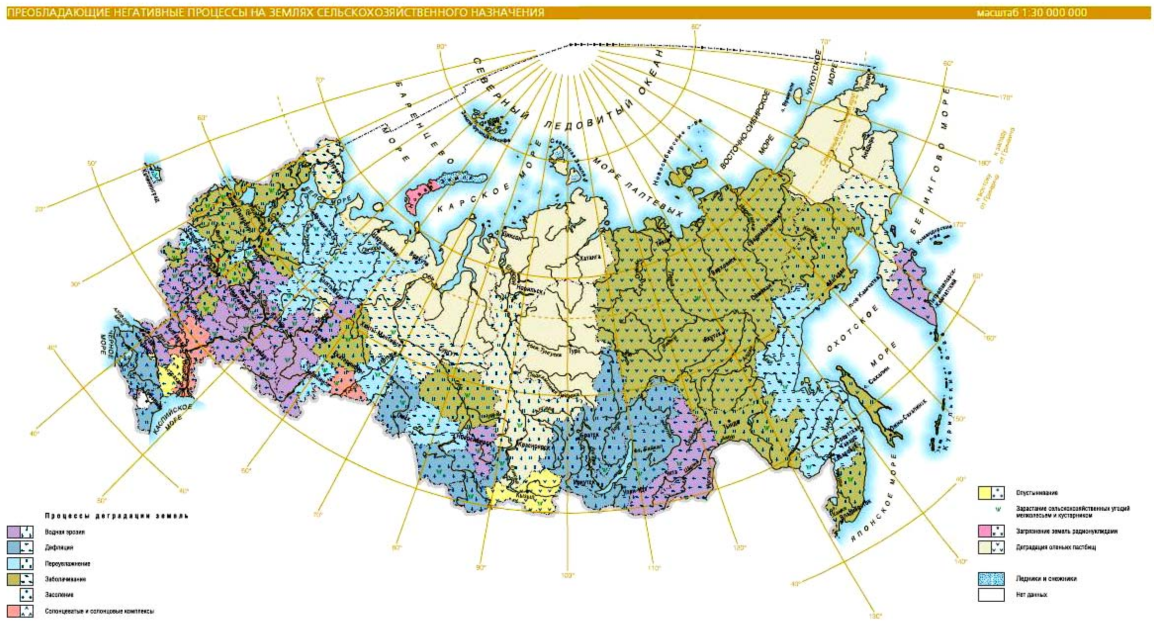

Fig.2. A schematic map of the processes of soil degradation in Russian Federation. Dark blue colour is for wind erosion, and yellow – for desertification (Shoba, 2011).

The development of such a soil geographical database started at the Faculty of Soil Science of Lomonosov Moscow State University several years ago (Shoba et al., 2008). The main blocks of the soil geographic database are the geographic database and the specialized attributive database (Rojkov et al., 2010). Relational Database Management (RDMS) System is used for data storage and processing. The Geographic Information Soil Database (GISD) forms the cartographic basis of the State Soil-Geographic Database of Russia. It consists of two digital coverages in MapInfo Professional. COVERAGE 1 is a digital map uniting the Soil Map of the RSFSR on a scale of 1: 2.5 M edited by Fridland (1988), and the digital map of the soil-ecological zoning of Russia. COVERAGE 2 is the digital map of the administrative division of Russia at a scale of 1:1 M. The soil profile (attributive) database of Russia is based on the concept of representative soil profiles (Kolesnikova et al., 2010). The database has a hierarchical structure ensuring soil description at several levels: SOIL–PIT–PROFILE–HORIZON–SAMPLE. The main object of the database is a specific soil profile with a set of soil horizons characterized by attributive data. The representative profiles should have an exact geographical location and be provided with a morphological description and a complete set of analytical data. The necessary conditions for the selection of representative profiles are: strict gridding of each soil profile, the most complete description of soil morphology, and the most extensive list of analytical soil characteristics (Shoba et al., 2011). The method of analysis, the units of measurement and the range of variation of each soil characteristic included in the database should be indispensably mentioned. For facilitating that, we developed uniform standards for soil information. The Program Soil-DB allows a provider of information to login in the site, to create and fill the soil description card, and to send it through Internet to the central server of the system (Anonimous, 2008). All necessary information on the properties and composition of soils is acquired by selecting representative soil profiles characterizing the main soil types in a generalized legend to the soil map on a scale of 1:2.5 M. Recently we started a joint project with neighboring countries for harmonizing the Russian, Ukrainian, and Belorussian soil databases.

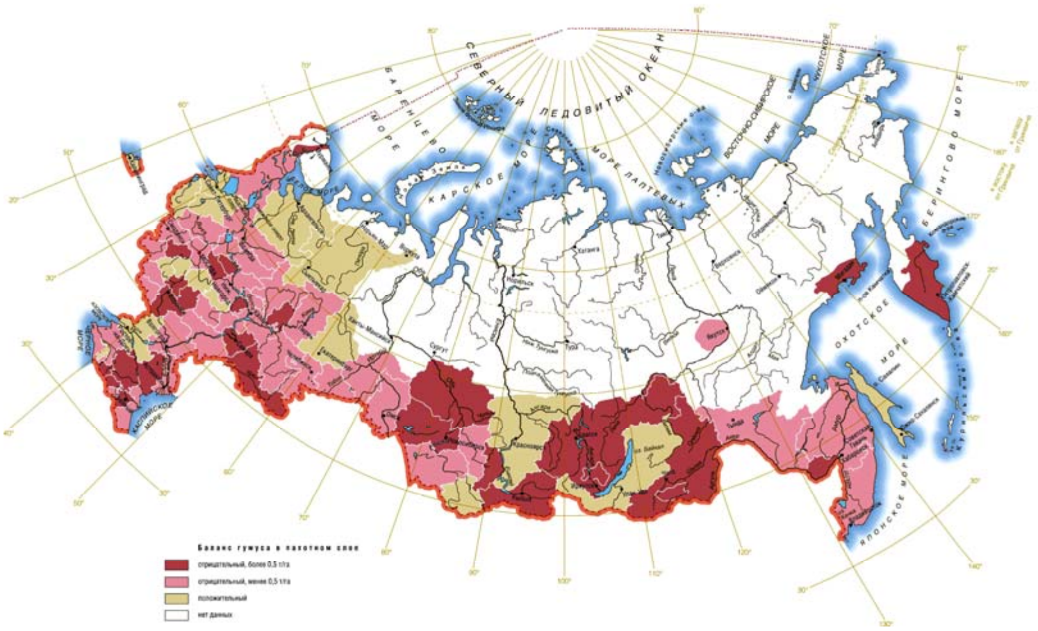

Fig.3. A schematic map of the balance of soil organic carbon in Russian Federation. Dark brown colour indicates strongly negative balance (Shoba, 2011).

Soil information in Central Asia

In the USSR the Central Asian region was always one of the most important centers of agriculture, because the climate allowed mellow-growing and especially extensive cotton production, which was impossible in other parts of the country. This epoch was both a time of great achievements in agriculture and a period of major negative impact on soils, especially related to secondary salinization. Several big state institutions and companies worked on the development of maps and explanatory notes on the land and water resources of Central Asia, with an emphasis on land improvement (irrigation, irrigation with drainage, chemical amelioration etc.). These data are important sources of information. Actually the Eurasian Center for Food Security of Lomonosov Moscow State University works on digitizing and publishing these materials on the web. Apart from this effort, we are discussing the possibility to include the information available on soil profiles of Central Asian countries in the joint Soil Database of Russia, Ukraine and Belorussia. We suggest using Soil DB tools for uploading the available soil information in Central Asia. The advantage of Russian Soil Database is that it can be easily converted to other soil database. Thus, the information would be integrated in the international data storage.

In Central Asia soil information obtained during the second part of the 20th century is of particular importance, because it can be used as a reference for soil monitoring, especially for irrigated areas suffering strong anthropogenic impact that have lead to soil salinization. The use of time series of remote sensing data may be also a good option for soil monitoring.

Perspectives of the use of soil databases

The perspectives for the future use and management of soil databases may be grouped in two main lines of research. Firstly, extensive soil information allows spatial modeling, and, secondly, it allows the prediction of the dynamics of soils on the basis of data mining, pedotransfer functions and dynamic models of the processes of soil degradation. These two lines are commonly interlinked, if we need to perform a spatial prediction of modeled variables.

The development of pedotransfer functions is of major importance for successful development of the guidelines for soil monitoring, land management planning and soil improvement. Usually the final user of soil information needs data in a format completely different from the primary soil data. The decision-makers do not care about pH values or electrical conductivity, they have to know, if the productivity of crops will be high enough to cover the investments to soil amelioration and management. This information may be produced only on the basis of pedotransfer functions, which, in their turn, can be obtained by data mining. For successful data mining we need thousands of soil profiler, otherwise any prediction and modeling would be baseless. Thus, both the development of information tools and filling the databases with reliable data are important components for information support of agriculture in dry areas.

Our task is to bring essential soil information to the decision-makers. Unfortunately, until now the response from regional administrative bodies is very low. However, now the situation changes slowly, because the governing bodies start to understand that soil degradation limits agricultural productivity, and, thus, the income of the population. Since increasing desertification leads to a drastic decrease in available soil resources and provides instability in agricultural production and food insecurity, the attention to soil data should increase.

Acknowledgements
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CONTENTS

POLICIES FOR SOIL HEALTH MANAGEMENT IN AGRICULTURE

ORAL PRESENTATIONS

Correlation between Aggregate Stability and Microbiological Activity in Tow Russian Soil Types

Mustafa I. Umer[1], Shayma M. Rajab[2]

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Abstract
Two Russian soil type, soddy-podzolic soil from Vladimerskaya region and dark-gray forest soil from Korskya region were taken .some microbiological parameters were assyed as basal respiration, substrate induced respiration, microbial biomass, microbial metabolic coefficient and correlated with soil aggregate stability concerning soil organic matter ,soil texture and soil bulk density . The result shown a positive correlation between all microbiological parameters with soil aggregate stability at this rank, microbial metabolic coefficient> microbial biomass= substrate induced respiration>basal respiration. Microbiological parameters and soil aggregate stability in dark-gray forest soil are greater than soddy-podzolic soil except basal respiration as a result of high organic content in this soil as will as the biomass as a percent of soil total organic matter. aggregate disintegration coefficient of dark-gray forest soil is 0.0028 with R^2 0.927 and need 85 rain drop (equivalent to an energy of 83385 J Kg^{-1}) greater than soddy-podzolic which had disintegration coefficient 0.0039 with R^2 0.849 and needed only 40 rain drop (equivalent to an energy of 39240 J Kg^{-1}) .
Keywords: Substrate induced Respiration, Basal Respiration, Biomass, Aggregate Stability, SOM

INTRODUCTION
Microorganisms are the primary agents of aggregate stabilization. Both fungi and bacteria contribute to stabilization of soil aggregates through deposition of extracellular polysaccharides and formation of degraded, aromatic humic materials that form clay–polyvalent metal–organic matter complexes. Though not as persistent, fungi also contribute to aggregate stabilization through hyphal anchoring of particles. The influence of fungi and bacteria on aggregate stabilization varies widely among species and depends considerably on the nature of the available substrates (Aspiras et al. 1971) and on the products of rhizode positions (Reid and Goss 1981). Furthermore, the type of land-use management can influence both the composition of microbial communities and their contribution to aggregate stabilization (Beare et al. 1994).

Soil microbial biomass is one of the most important soil biological properties. It regulates many critical processes in ecosystems, such as the biophysical integration of organic matter with soil solid, aqueous and gaseous phases. It also becomes vital in regulating the quantity and quality of components in the hydrologic cycle and in greenhouse gas emissions. The measurement of microbial biomass is useful for describing biomass turnover in different ecosystems.

Aggregation is a product of interactions of the soil microbial community, mineral and organic components, the composition of the above-ground plant community, and what has happened to the ecosystem in the past (ŽKemper and Koch, 1966; Tisdall and Oades,1982; Goldberg et al., 1988.). In addition to an indirect role in aggregation and soil structure via their contribution to humification, soil microorganisms also act directly. Bacteria and fungi exude colloidal polysaccharides that can glue soil particles. Soil fungi, for example, produce glomalin, which has been demonstrated to represent a high proportion of soil organic matter promoting aggregate formation (Rillig et al. 2002). The mechanical role of microorganisms is also considerable, given their biomass of 40–200gm−2 and their hyphal structure (Dighton and Kooistra 1993; Thorn 1997) that contributes to anchoring soil components to each other. The fungal mycelium has been described as a ‘sticky string bag’ because it entangles particles within the hyphae network and cements particles together through extracellular polysaccharide production (Oades and Waters, 1991).

Decomposition rates of organic matter (OM) in soil aggregates are reduced compared with OM not associated with aggregates (Besnard et al., 1996; Angers et al., 1997). This has been attributed to factors such as a reduced oxygen diffusion rate within aggregates (Sexstone et al., 1985) and the physical separation of SOM from microflora and fauna (Hattori, 1988).

Soil aggregate stability is the result of complex interactions among biological, chemical, and physical processes in the soil (Tisdall and Oades, 1982). Factors affecting aggregate stability can be grouped as abiotic (clay minerals, sesquioxides, exchangeable cations), biotic (soil organic matter, activities of plant roots, soil fauna and microorganisms), and environmental (soil temperature and moisture) (Chen et al., 1998). The aim of recent scientific work is to determine the most effective microbiological parameters correlated with soil aggregate stability and to link with the soil organic matter SOM.

Material and Method

Bulk soil samples were taken from two Russian soil type soddy-podzolic soil (дерново-подзолистой) from Vladimerskaya region (Владимирская область) and dark-gray forest soil from Korskya region (Курская область), soil aggregate in 10 -20 mm diameter are taken for microbiological and physical analysis from previous soils. Substrate-induced respiration (SIR) method are used to determine microbial biomass C in soils, the theory behind this method is that the initial rate of microbial CO2 production in response to a soluble energy-yielding substrate would be proportional to the mass of organisms. This method was initially developed to distinguish bacterial and fungal biomass, 100 g (oven-dry weight) are weighted and placed in to bags and treated with 55-60% water holding capacity then incubated at 22 C° for 7 days,1 g soil from incubated soil putted in vials by five replicates and 0.1 ml of 10% glucose solution were added to each replicates and caped with a septum. Time recorded and incubated at 22 C° for 3 hours, sample the headspace of the vial with a syringe and inject the 1 ml of gas into the Gas Chromatography to determine the ISR, where 1 g soil putted in vials by 5 replicates are incubated for 24 hours at 22 C° without adding glucose solution and measured by same way for Basic Respiration, five vials are incubated without adding soil and glucose as control (BS) (Anderson, Domsch,1978 ; Domsch et al.,1979)

Basal Respiration and Substrate Induced Respiration of soil are calculated:

SIR (µg C-CO2 /g soil · h) = (% volume CO2 sample - % volume CO2 air)·V vial (ml) · 12 µg /mole · 60 · 10 / 22 mmole/µl · m dry weight · Δt (minute)

Soil microbial biomass calculated from SIR velocity by this function:

C microbial (µg/ g soil) = (µl C-CO2 / g soil. h) · 40,04 + 0,37

Microbial Metabolic Coefficient calculated from the relation between basal and substrate induced respiration QR = BR/SIR

The rate of soil aggregate disintegration were determined by two ways A/ wet sieving technique were used to estimate the rate of soil aggregate disintegration using air dried samples of 4-8 mm in diameter which obtained by sieving, 20 g of oven dried soil aggregate transferred to a sieve having 0.25 mm diameter apertures. The water level was adjusted so that the aggregate on the sieve were just submerged at the highest point of oscillation. The oscillation rates were 40 cycles in minute, the amplitude of the sieving action was 40 mm and the period of sieving ranged from 2- 90 minutes. The weight of soil aggregate retained was monitored after different shaking times (0, 5, 10, 20, 30, 40, 50,60, 70,80 and 90 minutes) according to (Yoder, 1936) B/ measuring the energy required for aggregate disintegration using raindrop simulator which consists of 50-ml burette installed at 1-m height to form raindrops of 0.1 ml in volume .the number of simulated raindrops required to disintegrate an individual aggregate 0.5-0.52 g in weight and to pass through the 2.8 mm sieve was recorded. This test is known as counting the number of drop impacts (CND) (Imeson and Vis, 1984) .The particle size distribution analysis was performed by sieving and pipette method and bulk density was determined by clod method as outlined by (Klute,1986). The organic matter was determined by modified Walkley- Black method (Allison, 1965)

Statistical analyses were done by Microsoft Excel Software (2003) to determine the correlation between selected response physical variables and microbiological activity parameters in the soil.

Result and Discussion

The microbiological activity parameters which shown in table (1) are compared between each other's to determine which of them is most correlated with aggregate stability (rate of soil aggregate disintegration) as shown in table (2).

Table 1: Some of microbiological activity parameters (substrate induced respiration, basal respiration, microbial biomass, and microbial metabolic coefficient) in studied soils.

Soil type	SIR µg C-CO2/ g soil h	S.D	BR µg C-CO2 / g soil h	S.D	C.mic µg /g soil	S.D	QR	S.D
Dark-gray forest	19.091	±2.207	1.066	±0.107	1413	±163	0.056	±0.0034
Soddy-podzolic	18.057	±1.711	1.784	±0.150	1169	±111	0.099	±0.0136

The results are declared that the all microbiological parameters have a positive correlation with aggregate stability however the microbial metabolic coefficient (QR) is more correlated with aggregate stability than the others soil microbiological parameters in both soils, the correlation coefficient is more closed to dark-gray forest soil than the soddy-podzolic soil as a result of high organic content of dark-gray forest soil than soddy-podzolic soil as shown in table (2) and (3). This result is similar to (Machulla 2003) who reported that the best indicator of the whole metabolic activity of soil microbial populations is soil respiration, a robust parameter that can be rapidly and reproducibly determined. It allows gross comparisons of soils and reflects soil management changes, or the impact of elevated atmospheric CO2 on soil microorganisms (Machulla 2003).

Table 2: Some of important soil properties related with aggregate stability.

Soil type	Bulk density gm/cm^3	Particle size distribution $g.Kg^{-1}$			Texture	O.M $g.Kg^{-1}$	Biomass as a percent of soil organic C	Kinetic Energy $J\ Kg^{-1}$	No.of rain drop	Rate of soil aggregate disintegration $g.min^{-1}$
		Clay	Silt	Sand						
Dark-gray forest	1.248	266	605	129	SıL	62.1	3.92	83385	85	0.003
Soddy-podzolic	1.255	251	645	104	SıL	43.8	4.60	39240	40	0.002

While the soil biomass (microbial carbon) C.mic µg/ g soil and Induced soil respiration SIR µg C-CO2 /g soil h are seemed to be in the second of rank and have approximately the same level of correlation with soil aggregate stability as shown in table (3) because the varying degrees of correlation between aggregation and microbial biomass or microbial products are related to: (1) the different scales (i.e. macro versus microaggregate scale) of influence of fungi versus bacteria; (2) soil texture; and (3) soil mineralogy. The link between microorganisms and aggregation is pertinent, microbial biomass and water-extractable carbohydrates have been correlated to varying degrees with aggregation (Degens, 1997)

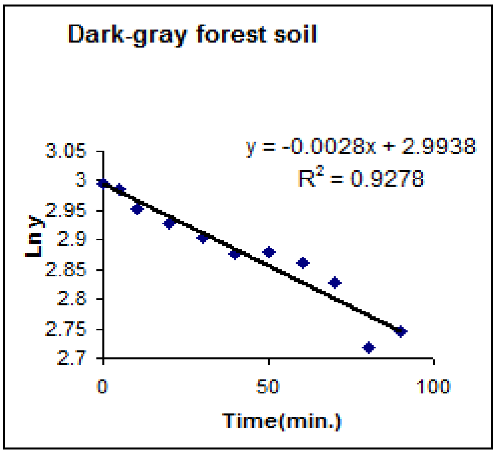
Table 3: Correlation coefficient between microbiological activity parameters and the rate of soil aggregate disintegration

Soil type	SIR µg C-CO2 /g soil · h	BR µg C-CO2 /g soil · h	C.mic µg/ g soil	QR
Dark-gray forest	0.25853	0.10268	0.258442	0.44525
Soddy-podzolic	0.102921	0.26096	0.10268	0.30636

The different organic fractions contribute to aggregate stabilization, including microbial biomass in particular fungi (Degens, 1997), microbial-derived polysaccharides (Haynes and Francis, 1993), humic substances (Piccolo and Mbagwu, 1999), and lipids (Dinel et al., 1991).
The basal respiration in soddy-podzolic soil has a greater positive correlation with aggregate stability than dark-gray forest soil which may be the result of that this soil contain high amount of microbial carbon (biomass) as a percent of total organic carbon in soil as shown in table (2) as reported that soil microbial biomass responds much more quickly than most other soil fractions to changing environmental conditions, such as variations in substrate input (Powlson et al. 1987)

Linked parameters (e.g., biomass-specific respiration or biomass as a percentage of soil organic C) are also useful because they possess "internal controls" (Barajas Aceves et al. 1999).

The two soils particularly have the same texture with little amount of clay and high amount of silt and have the same bulk density as shown in table (1) and (2) so the major role here for binding soil aggregate are the SOM and microbiological activity which explain the high correlation coefficient with microbiological parameters in dark-gray soil which contain high amount of SOM than soddy-podzolic , as reported that the contribution of soil microorganisms to aggregation is most apparent in soils of lower clay content and low shrink–swell capacities, where the abiotic effects of wet– dry and freeze–thaw cycles are reduced (Oades 1993),



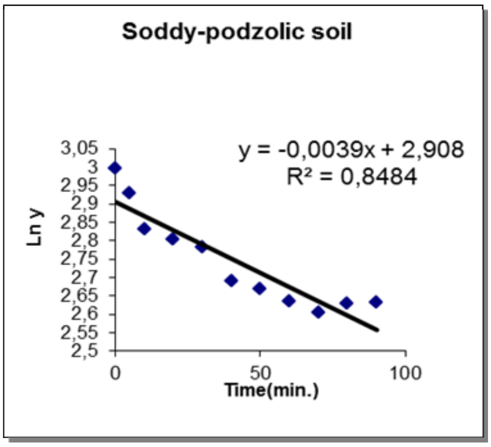


Figure 1. Change in soil mass of aggregate retained on 0.25mm sieve over time due to shaking during wet sieving

In fig (1) presents the mass of soil aggregates retained on 0.25 mm sieve as a function of shaking time in minutes for two soils from two locations . The soils from each location encompassed two different land use cropland and forestland. It can be noticed from these figure that there was a continuous decrease in mass of soil retained on the 0.25mm sieve over the entire range of shaking time which lasted 100 minutes. Further, it can be observed that the majority of disintegration occurred gradually during the whole time of sieving due to the resistance to break down upon wetting, however the dark-gray forest soil shown greater resistance against resistance as a result of high SOM content. Figure (1) also reveals that the slope of straight line (coefficient of determination) ranged from as low as 0.0028 for the dark-gray forest soil for Vladimerskaya region to as high as 0.0039 soddy-podzolic soil in Korskya region due to the same reason.

Water drop impact test as illustrated in table (2) exhibits the number of drop impacts required to disrupt the aggregate sufficiently for it to pass through the 2.8mm sieve and kinetic energy required to break up the aggregate. Dark-gray forest soil need 85 rain drop(equivalent to an energy of 83385 J Kg^{-1}) greater than soddy-podzolic more than twice which needed only 40 rain drop (equivalent to an energy of 39240 J Kg^{-1}), close examination of this data reveal that the resistance of aggregates to disruption by rain drop was mainly related to soil organic matter content. Canton et al. (2009) found a significant correlation between the number of drop impacts and soil organic matter content under wet conditions.

Conclusions

Soil aggregate stability is more governed by SOM than microbiological activities in the soils which have a proportional same texture and bulk density. All microbiological activity parameters have a positive relation with aggregate stability in both dark-gray forest and soddy-podzolic soil. Microbial metabolic coefficient which is a proportion between soil basal respiration and substrate induced soil respiration considered a good index to determine the soil aggregate stability which is directly proportional to the decrease in QR values. While the biomass-specific respiration or biomass as a percentage of soil organic C is inversely proportional with aggregate stability. Water drop impact test gives is more proper, simple and gives distinct clear result than the wet sieving technique to determine the aggregate stability in soils particularly have high SOM.

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Influence of arbuscular mycorrhizal fungi and micronutrients deficiency on tomato and sorghum dry weights in perlit culture medium

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Abstract
It has widely been accepted that arbuscular mycorrhizal fungi (AMF) can affect their host plants growth through nutrient uptake enhancement in soil growth medium, but scant studies were carried out for that effect on plants cultivated in the hydroponic and aeroponic systems. In this study, tomato (*Lycopersicon esculentum* Mill.) sorghum (*Sorghum bicolor* L.) plants were grown in sterile perlite and inoculated with either *Glomus etunicatum* (GE) or *G.intraradices* (GI), while the control set was left un-inoculated. Rorison's nutrient solution with three levels of zero, half and full strength (C_0, $C_{0.5}$ and C_1, respectively) of micronutrients was applied to the pots during 85 days of growth period. In tomato plants, the mycorrhizal symbiosis was not observed; also fungi treatments did not effect on root and shoot dry weights. In sorghum plants, Average root colonization with GE and GI fungi were 43% and 37% respectively. Mycorrhizal symbiosis decreased root and shoot dry weights. Roots dry weights have not affected by the micronutrient levels. But, the zero level of micronutrient in comparison with other two levels (half and full) decreased dry weights of shoots. It seems that, these results related to macroelements (especially P) concentration in Rorison's nutrient solution.
Keywords: mycorrhizal fungi, sorghum, tomato, perlit, dry weight

Introduction
Arbuscular mycorrhizal fungi are soil microorganisms that have establish mutual symbiosis with the majority of higher plants roots, such as sorghum and tomato plants that have high infection potential with AMF. The AMF provides a direct physical link between soil and plant root. It has widely been accepted that AMF have increasing affect their host plants growth in soil growth medium (Smith and Read, 2008) through nutrient uptake enhancement (Giridhar and Sudhakara, 2011), stimulating growth regulating substances, increasing photosynthesis, improving osmotic adjustment under drought stress, increasing resistance to pests and tolerance to environmental stresses such as drought and salinity, and improving soil properties (Bethlenfalvay et al., 1988; AL-Karaki, 2000). But scant studies were carried out for that effect on plants cultivated in the hydroponic and aeroponic systems. Nutrient availability can have major effect on arbuscular mycorrhizal infection. The effectiveness of symbiosis depends on the continued supply of nutrients, such as C, N, P and other essential nutrients, however, the majority of studies have focused on the significance of P (Valentine et al., 2001), and little attention has been paid to the effect of micronutrients. Results of some studies indicated that high concentrations of heavy metals in soil reduced root colonization. A negative correlation between Zn or Cu concentration and AMF root colonization was found for plants grown in soil after application of sewage sludge (Boyle and Paul, 1988). Also, some evidence exist that root colonization was increased with decreasing metals concentration in soil (Schreiner, 2007). However, little information is available about the effect of micronutrients deficiency on root colonization by AMF in the hydroponic systems. And is it benefit mycorrhizal symbiosis for plants in this condition? It seems that these cases need to be understand.

Materials and Methods
Mycorrhizal inoculum production
Two species of arbuscular mycorrhizal fungi, Glomus etunicatum (Becker & Gerdemann) (GE) and G.intraradices (Schenck & Smith) (GI) were propagated with sorghum plants in 7 liter pots containing sterile sandy loam soil (Aliasgharzad et al., 2001). Rorison's nutrient solution, 20 mM $Ca(NO_3)_2.4H_2O$; 10 mM $MgSO_4.7H_2O$; 10 mM $K_2HPO_4.3H_2O$; 0.5 mM FeNaEDTA; 0.1 mM $MnSO_4.4H_2O$; 0.5 mM H_3BO_3; 0.01 mM $(NH_4)_6 Mo_7O_{24}.4H_2O$; 0.02 mM $ZnSO_4.7H_2O$ and 0.015 mM $CuSO_4.5H_2O$ in deionized water (Merryweather and Fitter, 1991) with 1/2 strength of phosphorus was added to the pots twice a week to bring the soil moisture to field capacity. Pots were kept in growth room with 28/20 ±2°C day/night temperatures and 16h photoperiod. After four

months, top plants were cut off and pot materials containing soil, mycorrhizal roots, hyphae and spores were thoroughly mixed and used as fungal inoculum. Root colonization percentage (Giovanetti and Mosse, 1980) and number of spores per 10 g soil (Gerdemann and Nicolson, 1963) were assessed to determine inoculum potential. Both inocula had an average of 65% root colonization and ~150 spores per 10 g soil.

Plant culture

Tomato (*Lycopersicon esculentum* Miller) and Sorghum (*Sorghum bicolor* L.) seeds were surface sterilized with 0.5% sodium hypochlorite for 15 min, and 10 seeds were sown in pots containing 2.8 liter acid washed and sterilized perlite. Fungal inocula were rinsed three times with distilled water to minimize their micronutrients content. Each pot received 60 g mycorrhizal inoculum as a layer of 0.5 cm thickness, 5 cm below the seeds. Control pots (non-mycorrhizal) received 60 g autoclaved inoculum. Two weeks after sowing, tomato and sorghum plants were thinned to 1 and 3 plants per pot respectively. Rorison's nutrient solution with three levels of 0, half and full strength (C_0, $C_{0.5}$, C_1, respectively) of micronutrients was applied to the pots twice a week during total growth period of 85 days. Pots were kept in growth room with 28/20 ±2°C day/night temperatures and 16h photoperiod.

Plants dry weights and root colonization

Eighty five days after sowing, plants were harvested and whole root system was washed. plants were divided to parts (root and shoot) and then dried in oven. Whole of that the shoot and root dry weights were recorded. Fine feeding roots (0.5 g fresh weight) were sub-sampled, cleared in 10% KOH and stained with trypan blue. Root mycorrhizal colonization percentage was determined by gridline intersects method (Giovanetti and Mosse, 1980).

Statistical analysis

A factorial randomized in complete blocks design was used with two factors of (1) mycorrhizal fungi with three variations (G.etunicatum, G.intraradices and non-mycorrhizal), (2) nutrient solution with three concentrations of micronutrients (C_0, $C_{0.5}$ and C_1), with three replications per treatment. Analysis of variance and mean comparison by Duncan's Multiple Range Test were carried out using MSTATC software.

Results

Tomato root colonization with AMF

This experiment repeated with two varieties of tomato seeds in the same condition, but results were not different. In conclusion, the mycorrhizal symbiosis was not observed in tomato plants.

Tomato root and shoot dry weight

There were no significant differences between fungi treatments in each level of nutrient solution. Also, three levels of nutrient solution didn't have significant differences with together in each of the GE and GI fungi treatments (Table 1).

Table 1. Effect of mycorrhizal fungi on tomato dry weight (g) in variable regimes of micronutrients

Roots	C_0	$C_{0.5}$	C_1
NM	2.23b*	3.10a	2.77ab
GE	2.57ab	2.50ab	3.00ab
GI	2.23b	2.63ab	2.70ab

Shoots	C_0	$C_{0.5}$	C_1
NM	16.27abc	16.93a	14.83c
GE	15.33abc	16.50ab	15.97abc
GI	15.30abc	16.40abc	14.97bc

NM, non-mycorrhizal; GE, *Glomus etunicatum;* GI, *Glomus intraradices.* C_0, $C_{0.5}$ and C_1 respectively are Rorison nutrient solution in three levels of zero, half and complete concentration of micronutrients.*Means in each column followed by same letter are not significantly different (p<0.05).

Sorghum root colonization with AMF

There were no significant differences in root colonization between G.E and G.I fungi treatments with supplied each levels of nutrient solution. Also, root colonization in the non-mycorrhizal treatments did not appear. In each mycorrhizal treatment (G.E or G.I) between supplied three levels of nutrient solution didn't have significant differences (table 2).

Table 2. Effect of variable mycorrhizal fungi inoculums and regimes of micronutrients on sorghum root colonization(%).

	C_0	$C_{0.5}$	C_1
NM	0b*	0b	0b
GE	46a	41a	42a
GI	40a	35a	37a

NM, non-mycorrhizal; GE, Glomus etunicatum; GI, Glomus intraradices. C0, C0.5 and C1 respectively are Rorison nutrient solution in three levels of zero, half and complete concentration of micronutrients.*Means in each column followed by same letter are not significantly different ($p<0.05$).

Sorghum Root dry weight

In this experiment between mycorrhizal treatments (G.E and G.I fungi) were no significant differences, But in each levels of nutrient solution, mycorrhizal treatments in comparison with non mycorrhizal treatment had low root dry weight. In non-mycorrhizal treatment, C_0 level of nutrient solution in comparison with level of C_1, significantly reduced root dry weight (Table 3).

Sorghum Shoot dry weight

Between mycorrhizal treatments in each levels of nutrient solution were no significant differences. In supplied each three levels of nutrient solution, mycorrhizal treatments in comparison with non-mycorrhizal treatments reduced shoot dry weight. In the non-mycorrhizal treatment, C_0 level of nutrient solution in comparison with the levels of C_1 and $C_{0.5}$, also in the G.E fungus treatment supplying C_0 level of nutrient solution in comparison with level of C_1, significantly reduced shoot dry weight. But, in the G.I fungus treatment, between supplying three levels of nutrient solution were no significant differences (Table 3).

Table 3. Effect of mycorrhizal fungi on sorghum dry weight (g) in variable regimes of micronutrients

Roots	C_0	$C_{0.5}$	C_1
NM	1.44b*	2.11ab	3.05a
GE	0.13c	0.23c	0.29c
GI	0.22c	0.19c	0.19c

Shoots	C_0	$C_{0.5}$	C_1
NM	6.42b*	9.24a	10.34a
GE	0.69d	1.17cd	2.01c
GI	0.50d	1.00cd	1.26cd

NM, non-mycorrhizal; GE, *Glomus etunicatum;* GI, *Glomus intraradices.* C_0, $C_{0.5}$ and C_1 respectively are Rorison nutrient solution in three levels of zero, half and complete concentration of micronutrients.*Means in each column followed by same letter are not significantly different ($p<0.05$).

Discussion

The results in sorghum experiment shows that the three levels (zero, half and complete) of the micronutrient concentration didn't effect on root colonization by AM fungi. Even in tomto plants, the mycorrhizal symbiosis was not observed. With attention to results of some studies such as, AM fungi colonization and extraradical hyphae growth were suppressed when plants were grown with the high level of micronutrients but this level of elements was not toxic to the plants and don't suppressed growth (Liu et al., 2000a), or Colonization can be reduced in the presence of high levels of Zn, Cu, Ni, and Cd. Also AM fungi infection was found to depend on both P supply and the availability other nutrients, and Plant grown in low P and sufficient other nutrient elements had the highest AM fungi infection (Valentine et al., 2001; Schreiner, 2007). In this process appear a fact that heavy metals reduced AM fungi colonization when the availability of this elements are very high such as supply of soil and water polluted with these metals but moderate or lower concentration of these metal elements don't effect on root infection by AM fungi. Also, it seems that root colonization more affected by availability of P element in bed culture. The experiment results of Liu et al. (2000a) confirm this assumption. In present study P concentration supplied were equal in all of treatment compounds, it seems that this agent caused sorghum root colonization didn't have significant difference. Maybe, P concentration in nutrient solution treatments with to other conditions of present experiment caused the mycorrhizal symbiosis couldn't be organized with tomato plants.

In sorghum plants, the mycorrhizal (G.E and G.I fungi) treatments in comparison with non mycorrhizal treatments reduced root and shoot dry weight. Also, reduction growth of mycorrizal sorghum plants more affected by AM fungi symbiosis than deficiency of micronutrients. In fact the AM fungi is benefit to their host plant, principally by increasing uptake of nutrient elements, improved drought resistance, increased resistance to soil pathogens, increased tolerance of salinity and heavy metals (Smith and Read, 2008) But instead, the fungi receive carbon from the host plant. Though the AM association can offer multiple benefits to the host plant it may not be obviously mutualistic at all points in time, and it is possible under some condition that the AM fungi may cheat their host plant in to supplying carbon with no apparent benefit to the plant that in some cases, this can cause a decline in growth (Lerat et al., 2003). But often, positive effect of AMF in growth and nutrient uptake appeared in soils that have low nutrient availability (Schreiner, 2007). However, proving that AM fungi are actually cheating is difficult, because it is benefit to the host plant in the wide range, and their cheating occur at specific times or under certain environmental condition or stress. In some experiments, AM fungi symbiosis reduced weight of plants (Fidelibus et al., 2000; Valentine et al., 2001; Walling and Zabinski, 2006). In many cases reduction of growth in mycorrhizal plant were observed when the P availability of soil were high (Graham, et al., 1996; Valentine et al., 2001; Schreiner, 2007), also Fidelibus et al. (2000) suggested that under P limited condition might provide insight the management strategies to enhance AM fungal benefit. Furthermore, suggesting that suppression of growth may be is due to increasing metabolic activities of AM fungus and resulting in higher carbon costs to the plant (Peng et al., 1993; Graham, et al., 1996; Fidelibus et al., 2000; Walling and Zabinski, 2006). Notice to the latter reasons, it seems that existence full strength of P concentration in supplied nutrient solution caused reduction AM fungi benefits and increased carbon costs for plants and consequently reduced plant growth in present study. These results have apparent that micronutrient concentration in range of supplied didn't effect on sorghum root colonization with AMF. And probably, P concentration in supplied nutrient solution (that were in full strength) is main agent in reducing mycorrhizal sorghum dry weight and preventing of initiation of mycorrhizal symbiosis in tomato plants. It seems that, in hydroponic systems for growing mycorrhizal plants, P concentration in nutrient solution should be reducing.

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Effect of salt stress on photosynthetic pigments in pistachio leaves (*Pistacia vera* cv.Badami-Riz-zarand) inoculated with arbuscular mycorrhizae (*glomus mosseae*)

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Abstract
Drought stress is one of the most important abiotic stress factors which is generally accompanied by photosynthesis inhabitation due to the perturbation of the biochemical processes involving the photosynthetic pigments degradation. On the other, it has been proved that mycorrhizal symbiosis can ameliorate the adverse effects of salt stress in many plants. In this greenhouse experiment, mycorrhizal seedlings of pistachio (Pistacia vera cv.Badami-Riz-zarand)inoculated with Glomus mosseae were exposed to 4 levels of saltiness (0.5, 3, 6 and 9dSm-1)and harvested 21 and 42 days after the commencement of salt treatments. Increase in salt stress intensity caused a significant decrease in chl a while total chl and carteniods content did not affect by salt stress and chl b showed a significant increase at highest salt stress intensity. Leaves of micorrizal plants contained significantly more photosynthetic pigments compare with non-mycorrhizal plants. Degradation of photosynthetic pigments were increased in second harvesting date. We conclude that mycorrhizal symbiosis can protect photosynthesis rate of pistachio seedlings against the adverse affects of saltiness at least in partial by inhibiting of their degradation.
Keywords: Mycorrhizae, salt stress, pistachio, chlorophyll, cartenoids, photosynthesis

Effect of salt treatment on micorrhization extent and water relations of pistachio seedlings (*pistacia vera* cv.Badami-Riz-zarand)

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Abstract
In a greenhouse experiment, the effect of arbuscular mycorrhizae (*Glomus mosseae*)was investigated on water relations of pistachio seedlings(*Pistacia vera cv*.Badami-Riz-zarand)under salt treatments. Pistachio seeds were surface-sterilized and sown in 5kg plastic pots containing autoclaved soil and inoculated with 10 g of inoculom. Six-months-old plants were exposed to 4 levels of saltness (0.5, 3, 6 and 9dSm^{-1}) and harvested 21 and 42 days after the start of salt treatments. Salt solution was prepared using sodium chloride and calcium chloride in the ratio of 2:1 and applied every two days. The amount of salt solution used for each pot was 20% more than field capacity to avoid salt accumulation in the soil. Based on the results, mycorrhizal infection was increased with increase in salinity level up to EC of 6 dSm $^{-1}$and then decreased sharply in both harvesting date. Salt stress duration had no negative effect on mycorrhization percentage up to EC of 3 dSm $^{-1}$ and then decreased. Relative water content (RWC) of leaves was higher in non-mycorrhizal plants in compare with mycorrhizal plants. RWC of mycorrhizal plants was increased with increase in salt stress intensity while in non-mycorrhizal plants, the highest amount of RWC was recorded with control. Generally, in both mycorrhizae treatments and all salt stress levels, RWC were higher in second harvesting date. Water use efficiency (WUE) of pistachio plants was affected by mycorrhizal symbiosis positively while salt stress levels and stress duration had no significant effect on the parameters. It can be concluded that mycorrhizal symbiosis can improve water relations of pistachio seedlings at least under moderate salt stress
Keywords: Mycorrhizae, salt stress, pistachio, water relations

Soil Petroleum Hydrocarbon Contamination Cleanup by Bioremediation

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Abstract

In our country Iran, soil contamination with petroleum compounds had happened from the beginning of discovery and use of petroleum in the last century. The pollution changes the natural conditions of soil and water resources and decreases soil productivity. In the this decade, biological remediation systems has been used to extract many organic pollutants from environment because these systems both have the ability to compete with other methods (physical and chemical) by their lower cost and less environmental hazard. In this research, the application of barley (*Hordeum vulgare*), *Pseudomonas putida* Bacterium and effevescent tablet combined of some bacteria (which has been studied for the first time in Iran) in a greenhouse study with contaminated soil (with loam texture) was investigated. The intensity of Total Petroleum Hydrocarbons (TPHs) was measured with the standard method of the U.S. Environmental Protection Agency[1] simultaneously with three steps of plant growth. The results showed that with applying 2×10^8 CFU of *Pseudomonas putida* per kg of soil, intensity of the TPHs was remediated by 46.63%, and applying 2.2×10^6 CFU of *Bacillus* inocolant combined of five species (*B.licheniformis B.megaterium B.pumilus*, *B.laterosporus and Bacillus subtilis*) in each kg of soil remediated 46.99%. In statistical analysis of the results with SAS program, the initial and final intensity of TPHs showed a significant difference under these three treatments at the significance level of 0.05. Also, the intensity of contamination of samples showed a significant difference with Blank soil in all steps of measurment. Furthermore, intensity of TPHs under two bacterial treatments showed no significant difference between initial and final insensities, and, this showed that effervescent tablet had higher efficiency in pollution remediation, because by lower number use of cells, it had an equal efficiency in remediaton with *Pseudomonas putida* treatment, approximately. Both bacterial treatments had higher efficiency in remediation of petroleum hydrocarbon pollution than barley.

Keywords: Bacillus, Barley, Bioremediation, Petroleum, Pseudomonas putida

Introduction

Most problems of soil in large cities are due to modern industrial developments. Factories need to extract and process on organic and mineral sources, forest products and petroleum materials, so that these activities would lead to producing high amount of pollutants (Behrouz, 2005). Human being's concern about distribution of natural cycle of earth, life and destruction of sources has increased more than before, in 21st century.

High petroleum contamination in "Bagher Shahr" area (in Tehran province) near the petroleum refinery has caused environmental problems and dangers. Ignoring the results of use and remediation of petroleum in Iran for about one century had caused a much hazard to a large part of soil and water resources. So, taking a useful national strategy in order to solve this problem and achieving stable development is vital. Undoubtedly this strategy should be based on knowing the equipment and should have the minimum risk on the environment. Achieving this goal, in this research, we used "biological" remediation method which is the most convenient and the least dangerous remediation method. This study was conducted to lessen the damage on remediating the soil.

Materials and Methods

Polluted soil was taken from Tehran petroleum refinery surroundings. Soil sample passed from a 4-mm sieve and was used in soil analysis and bacterial treatments. The hydrocarbon contamination was mixed with soil uniformly. TPHs measurement performed by Environmental Protection Agency of USA method (EPA413.1) (Hutchinson et al, 2001). Experiments of soil analysis were done

[1]EPA413.1

according to the tolerance of both used bacterial treatments to hard condition, no particular operation was done to supply bacterium's need. In this stydy, 2×10^{8} cells of *pseudomonas putida* bacteria and effervescent tablet of 5 species of *Bacillus* (*B.licheniformis B.megaterium B.pumilus*, *B.laterosporus and Bacillus subtilis*) that have 2.2×10^{6} cells per kilogram of soil was used as it has tolerance to petroleum and is applied in petroleum remediation. In the barly cultivated treatment, 30 seeds of barley was planted. We also considered blank treatment in style of Randomized Complete Block Design (RCBD) and factorial experiment with 3 replicates. The rate of TPHs in different steps of plant growth was measured.

Results

Results from analysis of contaminated soil and TPHs measurement are given in Tables 1 and 2, respectively. In Table 2 TPHs1, TPHs2, TPHs3 and TPHs4 indicate the rate of initial soil contamination, rate of soil contamination after about 20, 85 and 95 days after initiation of treatments performance, which are initial soil contamination, rate of soil contamination after germination, growth and procreative steps, respectively.

Table 1. Results of soils analysis

$CaCo_3$ %	P (ppm)	Na (ml)	K (ppm)	Mg (meq/L)	Ca (meq/L)	N%	OC%	Texture	EC (dS/m)	pH
16	37	9.1	28	17.2	16.4	0.46	.85	loam	2	7

Table 2. The rate of contamination under treatments in different steps of plant growth (mg/kg of soil)

TPH4	TPH3	TPH2	TPH1	Treatment
11830	14500	15083	22167	*Pseudomonas*
11750	11916	13833	22167	*Bacillus* mix
13900	14250	17830	22167	*Hordeum vulgare) (* Barley
18500	18830	21083	22167	Blank

Table 3 shows the percentage of TPHs reduction by deferent treatments in 3 steps of plant growth. This table shows the efficiency of treatments, and Table 4 shows the Price of preparing the bacteria in Iran.

Table 3. Percentage of TPHs reduction by deferent treatments in 3 steps of plant growth

4	3	2	1	Treatment
46.63	34.59	31.96	0	*Pseudomonas*
46.99	46.24	37.6	0	*Bacillus* mix
37.29	35.71	19.56	0	*Hordeum vulgare) (* Barley
16.54	15.05	4.89	0	Blank

Table 4. Price of preparing bacteria treatments in Iran

Bacterium	Price for 10^{8} cells ($)
Pseudomonas putida	2.50
Effervescent tablet of *Bacillus* mix	10

Figure 1, 2 and 3 show the trend of TPHs reduction, percentage of TPHs reduction and significant differences in TPHs reduction by different treatments, respectively.

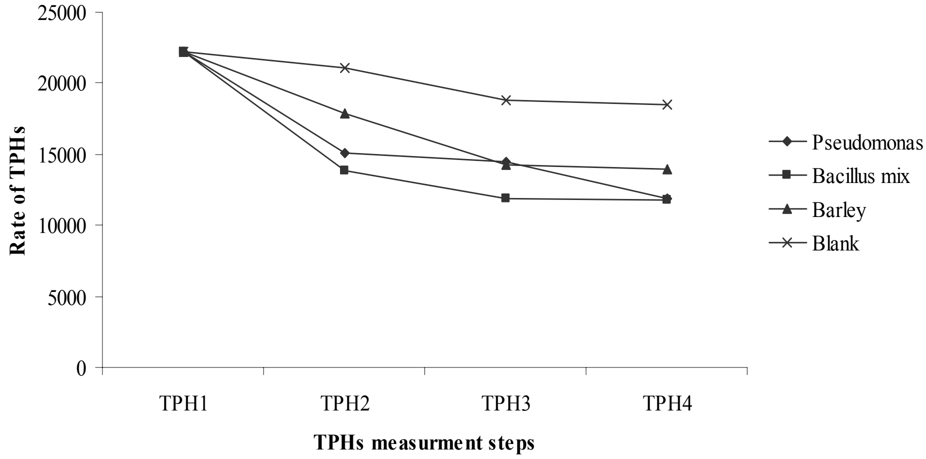

Fig 1. Trend of TPHs reduction by different treatments

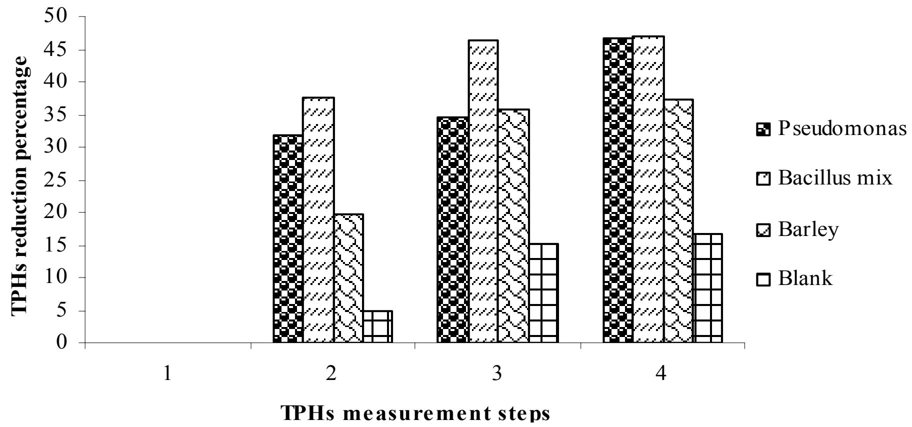

Fig 2. Percentage of TPHs reduction by different treatments in 3 steps of plant growth

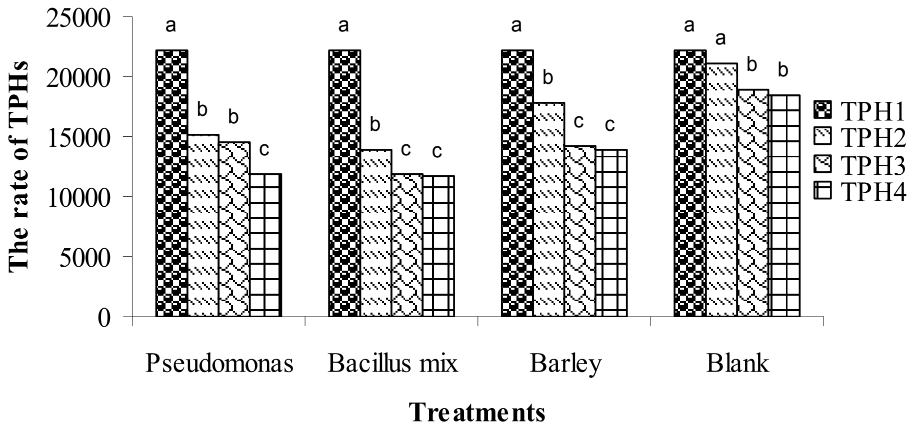

Fig 3. TPHs reduction by different treatments in 4 steps of pollution measurement and significant differences

Discussion

The maximum TPHs reduction rate was found for the " *Bacillus* mix" treatment (Fig.1, and Table 2). Maximum TPHs reduction percentage was found for the " *Bacillus* mix" treatment that reduced the contamination up to 46.99%, and in the second level"*Pseudomonas putida* " reduced the contamination up to and 46.63% (Fig. 2 and table 3) . In comparison with blank soil, this reduction was more than blank soil that shows the effect of root in petroleum hydrocarbons decomposition. It is obvious from results that using *Bacillus* inoculant combined of five species with less cells per 1 kg of soil had better results in comparison with *Pseudomonas* treatment. Preparing *Pseudomonas putida* had lower cost than *Bacillus* mix effervescent tablet (Table 4). Others also confirmed the ability of petroleum compounds decomposition by *Pseudomonas* and *Bacillus* genus and Barley plant (Frick et al. 1999 ; Mohammadi 2007; Wackett et al 1989). *Pseudomonas* ability in decomposition of TPHs have been proved in some other researches (Kim and Hao 1999; Lee and Gibson 1996; Lee and Gibson 1995; Shields et al 1991). Boossert, and Bartha in 1984 introduced the *Pseudomonas ،Arthrobacter ،Alkaligenes، Corynebacterium، Flavobacterium، Achromobacter ، Micrococcus ، Mycobacterium* and *Nocardia* as the most active bacteria in decomposing hydrocarbon compounds. Schwab and Banks showed some results on adsorption of organic pollutants by roots and decomposing them in their reports (Schwab and Banks 1998). The results of present study shows a good agreement with their results, the TPHs rate in rhizosphere reduced gradually (Table 2 and Fig.1) . There is no significant difference between TPH3 and TPH4 in "*Bacillus* mix" and barley treatments (Figure3). So performance of these treatments up to TPH3 step was reasonable.

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Effect of Land Use Change and Biological Restoration on Carbon Sequestration in Arid Area of Iran

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Introduction

In terrestrial ecosystems the amount of carbon in soil is usually greater than the amount in living vegetation. The amount of organic carbon stored in soil results from the net balance between the rate of soil organic carbon inputs and rate of mineralization in each of the organic carbon pools described (Walter 2002**.**
Study on variation of SOC in rangeland under plantation practices by woody plant in arid zone show the remarkable increasing in SOC (Nosetto 2006).
Because of variation in plant production and SOC have a strong correlation (Fang 2007).
This study focused on quantifies the influence of biological restoration on SOC in arid rangelands of Semnan province that restored by *Haloxylom aphyllum* plantation and change of rangeland to *pistachia Vera* cultivation.

Material and method

This research was conducted in Semnan and Ivanaky regions located in Semnan province, Iran. In order to study, 2 sites were selected. To estimate C storage and their distribution in the different treatment, biomass (above ground and belowground), soil (0-100 cm) and litter were sampled and analyzed.
Soil was sampled by collecting 5 cores in every treatment and its control area from depth of 100cm in a completely randomized design. Soil samples were air-dried, Gravel and roots were separated and samples were passed through the 2-mm sieve. Gravel was weighed and roots were taken to weigh after drying. To determine soil organic C the Walkley–Black method was used. Soil bulk density (Blake and Hartge, 1986) was determined for converting soil C concentrations (gr/kg) to C aggregate (ton/ha) in the soil.
Statistical analyses were conducted by T-test. Soft wares EXCEL 2003 and SPSS 15 was used to analyze data.

Results

Result show that both land use change to *Haloxylon ammodendron* and *pistachia Vera* plantations have increased 89% and 185% in carbon of biomass respectively.
In the soil, although *H. ammodendron* plantations have increased SOC but Pistachio has decreased SOC storages of soil. Generally haloxylon plantation has considerably increased sequestrated carbon of ecosystem. The similar results were not recorded for land use change into *pistachio* plantations (Table 1).

Table 1. Variation of C storage in rangelands and planted treatment in study areas

	Treatments			
Sources	*Rangeland*	*H.aphyllum*	*Rangeland*	*P.*vera
Aboveground Biomass	2.27±1.6 a	7.23±2.5 b	2.11±0.6 a	0.61±0.1 b
Belowgroun Biomass	1.72±0.2 a	4.40±1.1 b	1.58±0.6 a	0.91±0.1 b
Total Biomass	4.00±1.7 a	11.42±1.5 b	3.70±0.6 a	7.10±1.5 b
Soil	23.40±3.1 a	49.50±4.8 b	27.50±3.6 a	23.20±3.1 b
Total	27.40±5.0 b	61.20±7.4 b	61.20±9.4 a	30.20±3.5 b

Discussion

Plantation by *H.aphyllum* increase the amount of organic carbon in soil and biomass and it lead to increase carbon storage of ecosystem.

This matter explains the remarkable effect of plantation to increase amount of organic carbon in arid ecosystems (Nosetto et al, 2006).
Plantation by *P.vera* can not affect the carbon storage of ecosystem; *p.vera* plantation in this area increase the aboveground biomass but it case that the belowground biomass decreased with 41%.
The reasons of this result can be related to more extension of root biomass in *haloxylon* in compare with Pistachio and effect of agriculture practices on decreasing C of the soil (Kollia et al., 2009).
Then we find that plantation with species with huge proportion of belowground biomass can reboots organic carbon storage of the arid ecosystems.

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Co-inoculation effects of P- solubilizer (PSM) and plant growth promoting rhizobacteria (PGPR) on root and shoot growth in corn (*Zea mays* L.)

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Abstract: Plant-microbe interactions can positively influence plant growth through a variety of mechanisms, including fixation of atmospheric nitrogen, increased biotic and abiotic stress tolerance and direct and indirect advantages imparted by plant growth- promoting rhizobacteria. Therefore, a field experiment was conducted at research farm of Sari Agricultural Sciences and Natural Resources University during 2009. The experiment was arranged as split plot based on randomized complete block design with three replications for each treatment. Three levels of manures as main plots and eight levels of biofertilizers (consisting of 1-NPK or conventional fertilizer application; 2-NPK+PSM+PGPR; 3-$NP_{50\%}K$+PSM+PGPR; 4-$N_{50\%}PK$+PSM+PGPR; 5-$N_{50\%}P_{50\%}K$+ PSM+ PGPR; 6-PK+ PGPR; 7-NK+PSM and 8-PSM+PGPR) as sub plots were treatments. The results showed highly significant differences between treatments. Application of farmyard manure markedly increased shoot and root weight as compared to control. Direct inoculation of seeds with the plant growth promoting rhizobacteria (PGPR) significantly influenced growth parameters; distribution and average root length, root surface and the number of tips. Generally, according to our findings application of PSM and PGPR together could reduce P application by 50% without any significant reduction of growth parameters of corn.
Keywords: Inoculation, PSM, PGPR, Root, Corn

Introduction

Uses of bio-fertilizers containing beneficial microorganisms instead of synthetic chemicals are known to improve plant growth through supply of plant nutrients (Egamberdiyeva, 2007) and may help to sustain environmental health and soil productivity (Nikolay et al, 2006; Chaiharn and Lumyong, 2011). Plant growth-promoting rhizobacteria (PGPR) are a group of free-living bacteria that colonize the rhizosphere and benefit the root growth (Afzal and Asghari, 2008). The use of PGPR in agriculture for promoting the circulation of plant nutrition and reducing the need of chemical fertilizers is well recognized (Karnataka, 2007). Plant growth-promoting rhizobacteria, particularly those from the genus *Azospirillum* spp may affect root functions such as growth, nutrient and water uptake, which in turn may affect shoot growth (Lerner et al, 2006; Kapulnik et al, 2007). The use of PGPR offers an attractive way to replace chemical fertilizer, pesticides, and supplements; most of the isolates result in a significant increase in plant height, root length, and dry matter production of shoot and root of plants (Nikolay et al, 2006; Anjum et al, 2007).

Inoculation of forest trees, vegetables, and agricultural crops with PGPR may result in multiple effects on early-season plant growth, as seen in the enhancement of seedling germination, stand health, plant vigor, plant height, shoot weight, nutrient content of shoot tissues, early bloom, chlorophyll content, and increased nodulation in legume (Cakmaci et al, 2005; Shaharoona et al, 2006; Afzal and Asghari, 2008; Saharan and Nehra, 2011). Application of biological fertilizers such as biological phosphate fertilizers improves soil fertility. Several studies have conclusively shown that PSM solubilizes the fixed soil P and applied phosphates, resulting in higher crop yields (Roesti et al., 2006; Esitken et al., 2006; Violante and Portugal, 2007). Bacteria are usually effective on phosphate solubility due to different mechanisms such as production and secretion of organic acids (Turan et al, 2006; Saharan et al, 2011). Several studies have conclusively shown that PSM solubilizes the fixed soil P and applied phosphates, resulting in higher crop growths (Roesti et al., 2006; Violante and Portugal, 2007; Esitken et al., 2006). Bacteria of diverse genera were identified as PGPR of which *Bacillus* and *Pseudomonas* spp are predominant. Root colonization, influenced by a number of biotic and abiotic components, is a limiting factor for the success of PGPR. Diverse reporter genes and nucleic acid-based methods were developed to track the introduced PGPR in the rhizosphere, and also to determine their metabolic status, and their effect on the native rhizosphere microbial communities (Zhuang et al, 2007; Oliveira et al, 2008).

Nitrogen and phosphorus are essential nutrients for plant growth and development in corn (Wu et al., 2005). Nitrogen is one of the most important nutrients for maize production as it affects dry matter production by influencing leaf area development and maintenance as well as photosynthetic efficiency. N can be easily lost by leaching, denitrification or volatilization. Furthermore, Phosphorus is second only to nitrogen in mineral nutrients most commonly limiting the growth of terrestrial plants. Ironically, soils may have large reserves of total P, but the amounts available to plants is usually a tiny proportion of this total (Delvasto et al., 2008).
Corn (*Zea mays* L.) is one of the major cereal crops and grown under both tropical and temperate conditions. In this crop, root development plays a vital role in the growth of the corn plant and determining yield potential. The root system can be divided into two basic parts, the seedling root system and the nodal root system. Important developmental processes, such as root-hair formation, primary root growth and lateral root formation, are particularly sensitive to changes in the internal and external concentration of nutrients (lopez et al, 2003). Factors participating in the partitioning of dry matter between the shoots and the roots are highly significant to the well being of the plant (Lopez et al, 2003; Klaring and Kyuchukova, 2007). It is generally accepted that the dry matter distribution between shoots and roots follows the principle of functional equilibrium, although the underlying mechanism of this is still not understood. However, there is very little information regarding co-inoculation of PSM and PGPR effects in corn systems in northern Iran. The objective of this study was to determine co-inoculation effects of P- solubilizer (PSM) and plant growth promoting rhizobacteria (PGPR) on root and shoot growth in corn (cv. SC 604) under northern Iran conditions.

Materials and Methods
The experiment was conducted at research farm of Sari Agricultural Sciences and Natural Resources University (Latitude 42. 36 N, longitude 13. 53 E and 16 m above mean sea level), Iran during 2007. Experiment laid out as split plot based on randomized complete block design with three replications. Three levels of manures (consisted of 20 Mg ha^{-1} farmyard manure, 15 Mg ha^{-1} green manure of barley and control or without any manures) as main plots and eight levels of biofertilizers (consisted of 1- NPK or conventional fertilizer application; 2- NPK+PSM+ PGPR; 3- $NP_{50\%}K$+PSM+PGPR; 4- $N_{50\%}PK$+PSM+PGPR; 5- $N_{50\%}P_{50\%}K$+PSM+PGPR; 6- PK+PGPR; 7- NK+PSM and 8- PSM+PGPR) as sub plots were treatments. To chemical analyze, soils and farmyard manure were sampled before the experiment. Results of the soil and farmyard manure analysis are shown in Table 1 and 2. After soil testing, the NPK fertilizers at rate of 300, 120 and 100 kg ha^{-1} were applied in the form of urea, diammonium phosphate and muriate of potash, respectively. All of PK and half of N (starter fertilizer) were mixed with the soil at the time of sowing, while remaining N was applied in solution form at tasseling stage. The bacteria used in this study were phosphates solubilization microorganisms (*Pseudomonas putida, Bacillus lentus*) and plant growth promoting rhizobacteria (*Azotobacter coroococum, Azospirili brasilense*).
Corn (cv. SC 604) seeds were surface sterilized with 70% sodium hypochlorite for 1 min at room temperature. Then the seeds were washed repeatedly with sterile distilled water. Bacterial were suspended in suspension of sugar in water. This slurry was used to introduce the bacteria as corn seed coatings. The plants were removed 8 weeks after sowing, and roots were washed using slow running water to remove soil particles and organic debris. The number of secondary roots per sample was counted. Furthermore, for a more detailed study of the effects of PGPR and PSM bacteria, root samples were divided into three parts of ten centimeters. Then, the dry weight was measured separately for each section. After 4, 8 and 10 weeks shoot height and dry weights were determined. Shoots height was taken from the base of the stem to the apex. Each time four plant of corn from each treatment were randomly harvested. The shoots and the roots were dried in an oven at 70 ^{0}C until constant weight. They were then weighed separately and the weights recorded. Data were subjected to ANOVA using the SAS statistical software package using a GLM procedure (SAS Institute, 2000) and Duncan's multiple range tests was performed to compare the treatment means. The level of statistical significant was accepted as $P<0.05$.

Table 1. Soil chemical properties, and soil particle distribution of the top soil layer (0-30 cm)

Type	pH	OM (%)	N (mg 100 g^{-1})	P (mg 100 g^{-1})	K (mg 100 g^{-1})	Soil particle size (mm)		
						2. 0–0. 2	0. 2–0. 02	<0. 02
Silty loam	7.5	3.48	193	12.3	367.3	47.3	42.1	10.6

Table 2. Chemical properties of farmyard manure

pH	EC (dS/m^2)	N (%)	OM (%)	P	K	Cu	Zn	Mn	Fe
				(ppm)					
8.18	3.39	2.038	7.35	76	68	3.78	36.93	93.45	45.39

Results and Discussion

The results showed that application of farmyard manure significantly increased shoot and root weight. These results may be due to the higher levels of organic matter and nutrients (Table 2) in farmyard manure (Evanylo et al, 2008). Therefore, nutrient-rich organic amendments by farmyard manure can improve soil physical and chemical properties, increasing water holding capacity and nutrient availability, and promoting seedling establishment (Cherr et al, 2006). In addition, optimizing root system architecture can support yield limitations in crop plants caused by water or nutrient deficiency. Meanwhile, plant growth promoting rhizohacteria (PGPR) and phosphate solubilization (PSM) inoculation significantly increased root weight of corn (Fig 1).

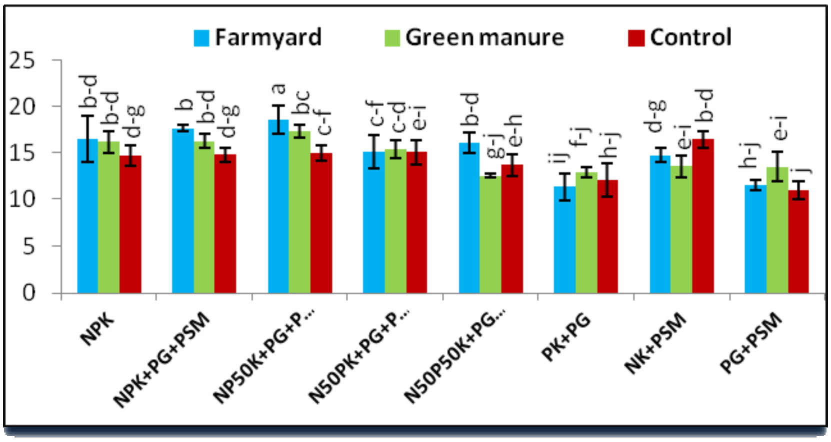

Fig 1. Co-inoculation effects of P- solubilizer (PSM) and plant growth promoting rhizobacteria (PGPR) on root weight (10-20 cm) in corn

Direct inoculation of seeds with the plant growth promoting rhizobacteria (PGPR) significantly influenced growth parameters; distribution and average root length, root surface and the number of tips (Table 3). Furthermore, using of PSM and PGPR in addition to conventional fertilizer applications (NPK) and $NP_{50}K$+PGPR+PSM treatment, could improve number of tips of root in corn (Fig 2). Similarly, Molla et al (2001) reported that total root length, root number, specific root length, root dry matter, root hair development and shoot dry matter were significantly increased by *Azospirillum* alone and its co-inoculums. Plant roots perform many essential functions including water and nutrient uptake, storage of reserves, synthesis of specific compounds, anchorage to the soil, and the establishment of biotic interactions in the rhizosphere (Lopez et al., 2003). It is concluded that single and dual inoculation along with P fertilizer is 18-22% better than only P fertilizer for improving total dry matter of corn. However, dual inoculation without inorganic fertilizer (P) decreased grain yield as compared to P application. The increase in root biomass of inoculated plants coincided with larger surface areas and greater cumulative lengths of roots than values observed for control plants which significantly different after 8 weeks. Distribution of roots and root length increased in a $NP_{50\%}K$+PGPR+PSM treatment (Table 3). It seems that, PGPR are more effective in promoting plant growth under limited supply of nutrients (50% of phosphate in this treatment). PGPR and PSM, help in increasing nitrogen fixation, increase supply of nutrients, such as phosphorus, sulphur, iron and copper and produce plant hormones.

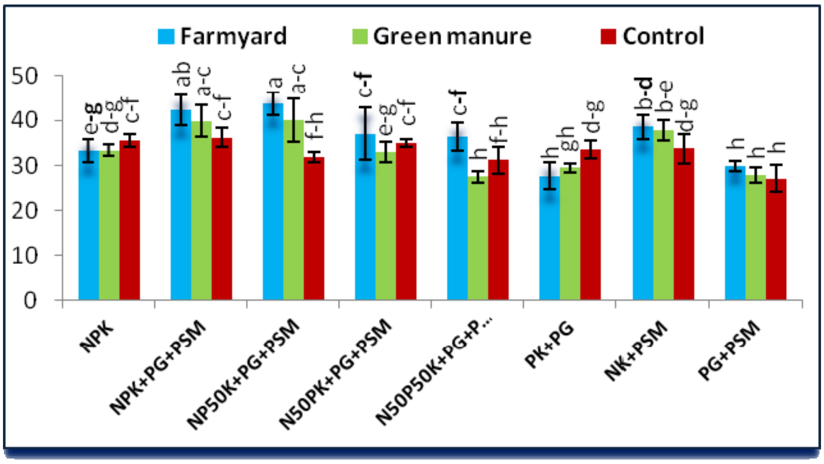

Fig 2. Co-inoculation effects of P- solubilizer (PSM) and plant growth promoting rhizobacteria (PGPR) on number of tips of root in corn

Table 3. Co-inoculation effects of PSM and PGPR on root and shoot growth of corn under different treatments

	Treatments	Root weight (g)	Shoot weight (g)	Total dry matter root (g)	Ratio of root: shoot	Total dry matter of plant (g)
Farmyard manure	NPK	0.93^{ab}	53.15^{a-c}	21.50^{bc}	0.41^{c-e}	310.40^{a-d}
	NPK+PGPR+PSM	1.10^{ab}	66.15^{ab}	23.41^{b}	0.36^{de}	351.61^{ab}
	$NP_{50}K$+PGPR+PSM	1.09^{ab}	68.10^{a}	28.97^{a}	0.43^{c-e}	353.90^{a}
	$N_{50}PK$+PGPR+PSM	0.78^{b-f}	40.40^{c-i}	19.75^{b-d}	0.50^{a-e}	298.93^{a-e}
	$N_{50}P_{50}K$+PGPR+PSM	0.79^{b-f}	47.75^{c-e}	21.65^{bc}	0.45^{a-e}	325.24^{a-c}
	PK+PGPR	0.45^{fg}	42.85^{c-h}	14.32^{f-h}	0.33^{de}	237.42^{d-g}
	NK+PSM	0.79^{b-f}	52.01^{b-d}	20.14^{b-d}	0.40^{de}	310.57^{a-d}
	PGPR+PSM	0.27^{g}	42.57^{c-h}	13.74^{gh}	0.32^{e}	215.34^{e-g}
Green manure	NPK	1.22^{a}	42.17^{c-h}	20.56^{b-d}	0.49^{a-e}	250.19^{c-g}
	NPK+PGPR+PSM	1.14^{ab}	46.11^{c-f}	21.82^{bc}	0.47^{a-e}	304.79^{a-d}
	$NP_{50}K$+PGPR+PSM	0.95^{a-d}	38.02^{d-j}	22.57^{bc}	0.60^{a-c}	354.32^{a}
	$N_{50}PK$+PGPR+PSM	0.77^{b-f}	37.08^{d-j}	19.14^{b-e}	0.52^{a-e}	311.23^{a-d}
	$N_{50}P_{50}K$+PGPR+PSM	0.86^{a-e}	36.53^{e-j}	15.88^{d-h}	0.44^{c-e}	290.83^{c-f}
	PK+PGPR	0.66^{c-g}	24.92^{j}	15.90^{d-h}	0.64^{ab}	253.16^{c-g}
	NK+PSM	1.04^{a-c}	28.08^{h-j}	18.44^{c-g}	0.66^{a}	327.91^{a-c}
	PGPR+PSM	0.51^{e-g}	31.02^{g-j}	15.93^{d-h}	0.52^{a-e}	225.51^{d-g}
Control	NPK	0.89^{a-e}	41.12^{c-i}	17.91^{c-h}	0.46^{a-e}	269.14^{a-g}
	NPK+PGPR+PSM	0.79^{b-f}	49.99^{d-j}	18.82^{b-f}	0.38^{de}	274.30^{a-g}
	$NP_{50}K$+PGPR+PSM	0.80^{b-f}	43.48^{c-g}	19.29^{b-d}	0.45^{b-e}	259.81^{c-g}
	$N_{50}PK$+PGPR+PSM	0.33^{g}	39.97^{c-j}	16.33^{d-h}	0.41^{c-e}	268.07^{a-g}
	$N_{50}P_{50}K$+PGPR+PSM	0.57^{d-g}	31.76^{f-j}	16.42^{d-h}	0.53^{a-d}	271.09^{a-g}
	PK+PGPR	0.27^{g}	28.39^{h-j}	14.41^{e-h}	0.51^{a-e}	211.47^{fg}
	NK+PSM	0.46^{fg}	31.83^{f-j}	19.37^{b-d}	0.61^{a-c}	265.45^{b-g}
	PGPR+PSM	0.28^{g}	26.48^{ij}	13.35^{h}	0.51^{a-e}	198.20^{g}

In each column means followed by the same letter (s) are not significantly different at $P \leq 0.05$.

The root-shoot ratio (R/S) of non-inoculated and inoculated plants was calculated. Treatments with PGPR and PSM significantly affected in terms of the R/S ratio (Table 3). Reduction in the root-shoot ratio in the NK+PSM and PK+PGPR treatment indicated that the root is probably limited. Reduction in the root-shoot ratio is owing to response to more favorable growing conditions. An increase in the root-shoot ratio, on the other hand, revealed that a plant was probably growing under less favorable conditions (Klaring and Kyuchukova, 2007). Shoot-to-root ratio in plants is affected by the balance between shoot-specific (carbon supply) and root-specific activities. When root growth is limited by a factor to be absorbed by the root system, then root growth is relatively favored; conversely, when the limiting factor has to be absorbed by the shoot, its growth is relatively favored. This observation concurs with several earlier studies that have reported positive interactions between PSM and a wide range of PGPR, including phosphate-dissolving bacteria (Molla et al, 2001; Zhuang et al, 2007; Saharan and Nehra, 2011). PGPR exert a direct effect on plant growth by production of phytohormones, solubilization of inorganic phosphates, increased iron nutrition through iron-chelating siderophores and volatile compounds that affect the plant signaling pathways. Additionally, by antibiosis, competition for space and nutrients, and induction of systemic resistance in plants against a broad-spectrum of root and foliar pathogens, PGPR reduce the populations of root pathogens and other deleterious microorganisms in the rhizosphere, thus benefiting the plant growth. Generally, according to our findings application of PSM and PGPR together could reduce P application by 50% without any significant reduction of growth parameters of corn.

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Factors influencing local community participation in sustainable management of land and water resources programs in Iran

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Abstract: During the last years, land and water resources in Iran have suffered severe degradation. In this regard governments of Iran have established several policies to sustainable management of natural resources. Most of these efforts have taken top down strategy and often unsustainable. In recent year's government has developed participatory approach in sustainable management of land and water resources in several areas. Hable-Rud program is one of these participatory programs to sustainable management of land and water resources which is initiated as a joint program of UNDP and Islamic Republic of Iran in 1997. The degree of popular participation in such programs is a major determinant of success or failure, but the factors which make participation efforts successful still remained a mystery. This study was designed to discover this mystery and determine the factors that influence local community participation in sustainable management of land and water resources. The data for this study were gathered from 200 respondents through personal interviews. The findings of study showed that level of participation in program was moderate to low; however people preferred more involvement in social rather than economical and environmental activities of program. Correlation analysis showed that five factors have positive and significant relationship with level of participation. Regression analysis discovered that these factors provided the best prediction and explained 45 % of the variation in level of participation. These five factors were; satisfaction of prior programs, attitude toward program, knowledge of program, monthly income from alternative occupation, and met expectations of program.
Keywords: Community, Participation, Sustainable Management, Hable-Rud, Iran

Introduction

About 50% of the world population lives in rural area, and majority of them are poor with no leverage to change their conditions. In order to bring change to these people, they need to be involved in program formulation and implementation. At the World Conference on Agrarian Reform and Rural Development (WCARRD) in 1979, the international community had linked the reason for the failure of rural development initiatives to the lack of active participation of the people in the development programs which were designed to assist them (FAO, 1992). There are 1.2 billion poor people in the world and 75 % live in rural areas and for the most part, they depend on agriculture, and related activities for survival. Rural areas in many of developing countries closely linked with agriculture, consequently to bring progress in the rural area requires changes and improvements in the agricultural sector. In the other side agriculture is closely linked with land and water resources, without land and water, agriculture are not possible. Moreover, because of rural dependency to agriculture, without developing agriculture the rural development goals for a sustainable livelihood cannot be met. The term participation has gained a lot of popularity during the last few years, particularly in reference to sustainable agricultural and rural development projects. The public participation today is demanding a greater role in decision making processes about the management of natural resource (Gwena, 2007). There are several studies which highlighted the importance of participation (Platt, 2006; Irvin & Stansbury, 2004; Ayee, 2001; Clayton, et al, 1998; Wainwright & Wehrmeyer, 1998). In point view of Platt, lack of participation in the society is one aspect of poverty. Irvin and Stansbury believed that citizen participation will produce more public preference decision making. In point view of Wainwright & Wehrmeyer participation by citizens and users presents an important concept and strategy for planners, designers, community organizers, and government officials. After land reform in Iran, in early 1960s, the natural resources came to control and property of state and from that time government is owner of all the natural resources. But in fact, the users of the natural resources are people of the regions in which resources located and government has not been able to control and posses the natural resources in practice. This issue is one of the chronic problems in natural resources management in Iran that exacerbated natural resources degradation and non-sustainability (Haji Rahimi & Hamed. 2008).

During the last years, land and water resources in Iran have suffered severe degradation. According to United Nation Development Program (UNDP); Iran is facing serious environmental challenges; environmental and natural resources have been substantially degraded (UNDP, 2006). In this regard governments of Iran have established several policies to sustainable management of natural resources during the last years. Most of these efforts have taken top down strategy and often unsustainable. In recent year's government has developed participatory approach in natural resources management in several rural areas. Hable-Rud Watershed Management program is one of these participatory programs to sustainable management of land and water resources in Iran which is initiated as a joint program of UNDP and Islamic Republic of Iran in 1997. The degree of popular participation in such programs is a major determinant of success or failure, but many opportunities for participation are met with little cooperation by citizens and the factors which make participation efforts successful still remain a mystery especially in Iran participation is quite a challenge for country with a long tradition of top-down management. Many studies have developed numerous and sometimes different views concerning to the dimensions of participation. Many studies on participation have been descriptive in nature and focused on distal factors; such as demographic and socio economic variables; on who joins and participate in development programs (Masiah, 2006). Internationally there are several researches that have utilized theoretical models to predict the determinants of participation (Gilles, 1995; Wilson, 1997; Mahon, 1998; Dolisca, et al, 2006; Keough, 2006) but there is a limited research that has been conducted on the context of participation with using theoretical model in Iran. In this study researcher attempts to analyze people participation in WMP based on the previous literature and social exchange theory, to examine the relationship of several factors with the level of participation to provide a better understanding of the role of each variable in the level of people participation in WMP. The framework of this study is based on social exchange theory. Central idea of social exchange theory is that the exchange of social and material resources is a fundamental form of human interaction (Ingoldsby & Smith, 1995). This theory basically asserts that people develop attitudes toward other people and things in the context of anticipated personal benefits and costs to be derived from contact with them. Activities that generate net benefits will tend to be perceived positively, while those activities that generate net losses will tend to be perceived negatively (Napier & Napier, 1991). Social exchange theory poses that all human relations are formed by the use of a subjective cost-benefit analysis and the comparison of alternatives. For social exchange theorists, when the costs and benefits are equal in a relationship, then that relationship is defined as equitable. The notion of equity is a core part of social exchange theory. In point view of Homans (1958) the initiator of theory, social behavior is an exchange of goods, material goods but also non-material ones, such as the symbols of approval or prestige. It also noted that individuals will enter into and maintain a relationship as long as they can satisfy their self-interests and at the same time ensure that the benefits outweigh the costs (Blau, 1964). According to Blau, in satisfactions human beings, experience in their social associations, depend on the expectations they bring to them as well as on the actual benefits they receive in them. The man who expects much from his associates is more easily disappointed in them than the one who expects little, and the same degree of friendliness might attract the first man to other people and discourage the second from associating with them. Social exchange theory also is tied to rational choice theory and on the other hand to structuralism, and features many of their main assumptions. Blau outlined three types of expectations of social rewards: general expectation, particular expectation and comparative expectation. Spencer & Steers (1980) have mentioned, met expectations in organizations are the extent to which once expectations concerning to organizational life have been met on the job. By meeting or exceeding the expectations of employees, organizations can decrease an individual's level of intent to quit (Turnley & Feldman, 2000). Organ (1998) proposed that supervisor fairness leads to employee citizenship because a social exchange relationship develops between employees and their supervisors, when supervisors treat employees fairly, social exchange and the norm of reciprocity. Gouldner (1960) dictate that employees reciprocate. Organ suggested that organizational citizenship behavior is one likely avenue for employee reciprocation. Expectations contribute to a "norm of reciprocity" because individuals have certain beliefs about what a program should provide to them as a participant in exchange for their efforts. To a certain degree, expectations are formed from societal norms. By meeting or exceeding these expectations, program managers can establish a baseline of perceived support which could serve as a buffer keeping a participant from leaving the program. Satisfaction depends on expectation, which is shaped by prior experience, especially memorizing events of the recent past. Indeed this factor closely related to exchange theory to understanding people behavior in relation to a subject. In terms of continuing relationships, Blau believed that individuals will try to maintain those exchanges which have proven to be rewarding in the past, to break off those which proved to be more costly than rewarding, and to establish new relations which have a good chance of being more rewarding than costly. Indeed fulfillment of a motivational desire, after need satisfaction has occurred, there is no further motivation for gratifying that need. Many studies have shown that prior experience affect on decision of people to participation in the current projects (Effati, 1992; Hibbard & Tang, 2004).

Participation in WMP may differ among people according to their socio demographic characterizes. Thus, participation in watershed management involves a combination of individual characteristics and as well as subjective evaluations. This individual characteristics influence decision-making regarding household behavior whether or not participate in watershed management programs. Several studies have shown that participation may depend on individual characteristic such as age, gender, marital status, education, household size, income, etc (Hosseini, et al, 2006; Zainuddin, 1977; Ilbery, 1978; McDowell, C, & Sparks, R, 1989). The differences in emphasis on watershed perceptions, allow men and women to maximize their individual well being. Knowledge, awareness, attitudes, and behavior are four interrelated components of an individual's action (Strauss, 1982). Education and knowledge are also important determinants of how benefits from watershed management programs are perceived. People cannot be expected to exhibit positive attitudes toward watersheds if they are unaware of the benefits and cost associated with their participation. Education and knowledge about watershed management issues make people more positive in their views. Prior to an individual's attitude and behavior, one has to have knowledge or awareness of the object, even, or problem under consideration. therefore, the cognitive mechanism of participation, in certain activities include communication, utilization of information, and a better understanding by individuals of activities in which their participation is sought (Hamid, 1996). Some studies revealed that education and knowledge are important determinants of how benefits from development programs are perceived. People cannot be expected to exhibit positive attitudes toward forest if they are unaware of the benefits and costs associated with their participation. Education and knowledge about forest conservation issues make people more positive in their views (Infield, 1988; Heinen, 1993; Mkanda & Munthali, 1994). Better informed and educated people should be more aware of potential benefits to be derived from the forest than individuals who are ignorant and illiterate. An attitude is a hypothetical construct that represents an individual's degree of like or dislike for an item.

Attitudes are generally positive or negative views of a person, place, thing, or event- this is often referred to as the attitude object. Attitudes are generally viewed as one's relatively enduring affective, cognitive and behavioral dispositions toward various aspects of the world including persons, events and subjects. It has been generally believed that attitude change is necessary before other behavioral modifications can be effected. Abu Samah found that" there is positive relationship between farmer's attitude toward program and intensity of farmer's participation in agricultural development program. Kraft et al (1996) found that' farmers with a negative attitude toward governmental involvement with wetland regulations were less likely to want to participate in the water quality incentives program. Rishi (2003) outlined that understanding of attitudes is one of the central concerns in social life and is vital for bringing desired change in the behavior. Social actions of people or program personnel are directed by their attitudes. By knowing the attitudes, it may be possible to do something about the prediction and control of their behavior, which may be ultimately useful for the more successful implementation of programme. Shahroudi and Chizari (2008) in their study in Iran found that there is significant and positive relationship between farmer's attitude and their participation in irrigation networks management. Vicente and Reis (2008) expressed, positive attitudes toward recycling and information are important factors in explaining recycling participation.

Material and methods

Hable-Rud basin located in north part of Iran, between altitudes 51.39' and 53.08' north and longitudes 34 26' and 35 57' east. This area has 57000 household distributed in 704 villages and includes the region that is characterized by high population density, land and water resource degradation and declines in agricultural productivity; posing significant challenges to rural peoples to provide for the growing population, while maintaining the productivity of natural resources. Based on literature review, a set of factors were adopted as independent variables to examine their relationship with level of people to participate in WMP. Data in this study mainly drown from the survey questionnaire. Development of the survey instruments was formed based on literature review and interviews with program staffs and also with participants in three villages in study area prior to actual data collection. Based on above procedure, specific questions addressing some of the measurements of the independent and dependent variables were extracted to test their relationship with level of participation. Socio demographic characteristics were measured depending on its appropriateness. Knowledge instrument were employed two point scales for measurement, attitude instrument were employed five point likert scales for measurement, and satisfaction of prior programs instruments were employed three point scales and level of people participation were measured with five point scales. Once the instruments were developed, it was reviewed by a panel of experts in UPM, SCWMRI, and FRWMO, in Iran to ensure the content validity of the instruments. A pilot test was carried out among thirty respondents during the August 2008. The Cronbach Alpha that is greater than 0.70 was used to measure the reliability of the instruments. Results of the computed reliability coefficients were; .812, .806, .937, .816 and .903 for knowledge, attitude toward WMP, satisfaction, expectations and participation in WMP, respectively. The selection of a sample for this study was multi stage random sampling procedures. In the first level, the name of villages and number of their population was obtained from project office and Statistics Center of Iran (SCI), and based on Israel G. D, (1992) adequate sample size determined for gathering data for this study. Based on the sample size formula and with regard to population of study area, a total of 200 respondents were determined as sample size of this study. In the second level three villages selected randomly to get determined sample size. In the last level, name of all participants' households in each selected villages were listed. Finally respondents randomly were selected from each selected villages. Data for this study were collected through personal interviews by using a questionnaire during the August and September 2008. Descriptive analysis, Parsons Product moment correlation, and regression analysis was used to analyze the data in this study.

Results

Findings of study showed that educational level in study area was relatively high (18 % diploma and high) and mean of respondents age was 46 years. Study also showed that main occupation of the majority was farming (55 %) and average of their total monthly income was 3.5 Million Rial per month. Findings of study also showed that average land ownership of the respondents was 3.12 hectares and 8.5 % of the respondents were landless. Study also showed that 58% of respondents were member at least in one local group and 47.5 % of respondents were joined to program with their self interest. Descriptive analysis of data showed that level of respondent's knowledge of WMP was low, however they were indicated positive and relatively high level attitude toward WMP. Findings of study also showed that respondent's expectations of WMP and their satisfaction of prior programs were moderate to high. Study showed that level of overall participation in WMP was moderate, however respondent's were preferred more involvement in social activities rather than economic and environmental. Table 1 shows the summary of respondent's knowledge of WMP, attitude toward WMP, satisfaction of prior programs, expectation of WMP and participation in WMP.

Table 1: level of knowledge, attitude, expectation, satisfaction and participation in WMP

Level	Knowledge of WMP(%)	Attitude Toward WMP(%)	Satisfaction of WMP(%)	Expectation of WMP(%)	Participation In WMP(%)
Low	39.0	7.00	27.0	14.5	37.5
Moderate	50.5	87.5	49.5	48.0	57.0
High	10.5	3.50	23.5	37.5	5.50
Total	100	100	100	100	100

Pearson's product moment correlation analysis showed that, income, satisfaction of prior programs, attitude toward WMP, expectation of WMP and knowledge of WMP all have significant and positive relationship with level of participation. Table 2 shows the summary of the correlation analysis between independent variables and level of people participation in WMP. Based on this table satisfaction of prior WMP have highest relationship (r =.518, p=.000) with level of participation in WMP and followed by attitude toward WMP (r =.489, p=.000), Knowledge of WMP (r =.435, p=.000), met expectations of WMP (r=.411, p=.000), total monthly income(r=.177, p=.012) and alternative monthly income (r =.158, p=.025).

Table 2: Correlation between independent variables and level of participation

Independent variables	**(r)**	**(p)**
Age	-.065	.358
Household size	-.050	.480
Main income	.013	.589
Alternative income	.158	.025
Total income	.177	.012
Land ownership	.073	.307
Knowledge of WMP	.435	.000
Attitude toward WMP	.489	.000
Expectation of WMP	.411	.000
Satisfaction of prior WMP	.518	.000

The stepwise method of regression was employed to determine the significant contributions among the predictor variables in explaining participation. A summary of the stepwise multiple regression procedure was presented in Table 3. This procedure was set at 0.05 significant levels. New independent variables were to be included in the analysis when they contributed significantly (p=0.05) to the multiple regression coefficient (R). ANOVA Table showed that the data fit to the model (F=31.759, sig-t =.000, Durbin Watson, 1.92). First step of regression model was started with satisfaction variable based on zero order correlation that had the highest correlation with participation (r=.518). The other variables were chosen to enter the model based on the highest partial correlation. Finally, with the combination of the five variables, the model 5 with highest R square (R= 45 %) was the best model for this study. Satisfaction of prior projects was the first independent variable to enter the analysis. This step yielded R-value of 0.518, reflecting the most powerful variable. The satisfaction score explained about 26.8 % of variance in the dependent variable. Additional variables included in the analysis were based on how well they improved the prediction from the predicting variable. Attitude toward WMP represented the second variable included in the analysis. This variable explained another 9.5 % of the variance in the dependent variable. The improvement accrued from the knowledge, alternative income, and expectation of WMP were; 4.9 %, 2.4 %, and, 1.4 % respectively. Study showed that these five independent variables explained 45 % of variation in the dependent variable. The proposed prediction equation of this study for overall factors was:

$$Y=0.32X1+0.18X2+ 0.25X3+.0.16X4+0.15X5$$

Where:
Y= Level of participation in WMP
X1= Satisfaction of prior WMP
X2 = Attitude toward WMP
X3= knowledge of WMP
X4= Alternative income
X5= Expectations of WMP

Table 3: Multiple regression for overall factors on level of participation

Variables	Standardized Beta	r	R	R^2	R^2 change
Satisfaction of prior program	.322	.518	.518	.268	-
Attitude toward WMP	.183	.489	.603	.363	.095
Knowledge of WMP	.248	.435	.642	.412	.049
Alternative income	.159	.158	.661	.436	.024
Expectations of WMP	.148	.411	.671	.450	.014

Conclusion

Based on the findings of this study it can be concluded that, a person who was more satisfied with previous programs, more likely to participate in watershed management programs activities, this is consistent with social exchange theory and with findings of Effati, (1992) and Hosseini, et al (2006) which found positive correlation between participation and satisfaction of prior programs. Study also showed that those who are more knowledgeable about program, more likely to participate in WMP, this result also congruent with findings of, Infield, (1998), Abu Samah, (1992), Heinen, (1993), Mkanda, (1994)], Fiallo and Jacobson, (1995), which found positive relationship between knowledge and level of participation. In addition study indicated that people who has positive attitude toward program more likely to participate in program. These findings also are consistent with results of Abu Samah, (1992), Kraft, et al., (1996), Rishi, (2003), Shahroudi and Chizari, (2008), Vicente (2008) and Asadi, et al., (2009) in their research that found significant relationship between attitude and level of participation. Findings of study showed that those who are met more his or her expectations of program; more likely participate in program, this finding also consistent with social exchange theory and with findings of Turnley & Feldman, (2000) and Organ, (1988). Finally study showed that a person who have high income more likely to participate in program activities, this finding also congruent with findings of McDowell and Sparks,(1989), Abu Samah, (1992) which found significant relationship between level of respondent participation in developing programs with their level of income. Results of this study showed that there are a large number of young potential work forces in the study area. This work force should be mobilized by program managers to generate additional projects by developing the program activities and alternative occupation along this project. Study also showed that the level of income in the study area is low, and poverty is spread among the rural people. Based on the above recommendation and availability of work force, creating new jobs can help and reduce the incidence of poverty among people in the study area. Study also showed that local organizations such as rural council have an acceptable place in study area and can help to develop efforts in WMP. Thus, program managers should continuous to provide material, technical, and advisory support to further mobilize these organizations to enhance people's participation in WMP. Study discovered that meeting the expectations of WMP has a positive relationship with the level of participation. However, people meeting the expectations were relatively low. Thus, more effort is needed for WMP management to consider people expectations in order to promote people participation in WMP. Study showed that the respondent's knowledge of WMP has a positive relationship with the level of participation. Nonetheless, the overall people knowledge of WMP was relatively low. Thus, more effort is needed for WMP management to deliver information on WMP to promote people participation in WMP. Study also showed that the respondent's attitude toward WMP has a positive relationship with the level of participation. This is a potential for the managers to develop such programs. Thus, more effort is needed for WMP management to develop WMP activities. Study also recommends that similar research should be conducted in other WMPs to validate the findings of this study and a more in depth study should be done by incorporating other variables such as; people trust to government, people trust to project staff and attitude toward government and project staffs, to further enhance the identification of factors that affect people, s participation in WMP to improve the prediction of the level of people's participation.

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POLICIES FOR SOIL HEALTH MANAGEMENT IN AGRICULTURE

POSTER PRESENTATIONS

Effect of Soil Contamination with Azadirachtin on Dehydrogenase and Catalase Activity of Soil

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Abstract
Insecticides are used in modern agriculture in large quantities to control pests and increase crop yield. Their use, however, has resulted in the disruption of ecosystems because of the effects on non-target soil microorganisms, some environmental problems, and decreasing soil fertility. These negative effects of synthetic pesticides on the environment have led to the search for alternative means of pest control. One such alternative is use of natural plant products such as azadirachtin that have pesticidal activity.
The aim of this experiment was to study the effect of soil contamination by azadirachtin ($C_{35}H_{44}O_{16}$) on dehydrogenase (DHA) and catalase activity (CA) of soil under field conditions in Perm, Russia. The tests were conducted on loamy soil (pH_{H2O} 6.7, EC_{H2O} 0.213 dSm^{-1}, organic carbon 0.99%), to which the following quantities of azadirachtin were added: 0, 15, 30 and 60 mL da^{-1} of soil. Experimental design was randomized plot design with three replications. The DHA and CA analyses were performed 7, 14 and 21 days after the field experiment was established.
The results of field experiment showed that azadirachtin had a positive influence on the DHA and CA at different soil sampling times. The increased doses of azadirachtin applied resulted in the higher level of DHA and CA in soil. The soil DHA and CA showed the highest activity on the 21th day after 60 mL azadirachtin da^{-1} application doses.
Keywords: Azadirachtin, soil, enzyme, dehydrogenase, catalase

Introduction
One of the side effects of the application of pesticides is that these chemicals accumulate in soil. Pesticides are biologically highly active, therefore they will have an effect not only on the organisms subjected to their activity but also on a number of other organisms present in this environment, many of which are useful in nature. Such side effects are compounded by long degradation times of some pesticides in soil (Wyszkowska, 2002). Persistence of these xenobiotics in soil depends on several factors determining the rate of their disappearance from the environment, of which the chemical structure of the active substance of a preparation, its chemical properties, formation of bonds with other compounds and biodegradability are most significant (Chapman et al., 1981; Demoute, 1989). An important role is also played by environmental and agricultural factors, including temperature, pH, moisture, soil type, organic matter content, fertilisation and count and activity of soil microorganisms (Pedziwilk, 1995; Kızılkaya, 1997; Wyszkowska, 2002).
Degradation of pesticides is catalysed by enzymes excreted by microorganisms, producing in effect some intermediate metabolites, which may have a selective influence on soil microflora. Accurate determination of such modifications occurring during microbiological and biochemical processes is essential for sustaining and regenerating the fertility of soil (Wyszkowska, 2002). An assay of the enzymatic activity of soil, especially the activity of enzymes involved in the conversion of C, N and P, can be regarded as a good indicator of the effect of pesticides on soil metabolism, and also soil enzyme activities are very sensitive to both natural and anthropogenic disturbances, and show a quick response to the

induced changes (Dick, 1997). Therefore, enzyme activities can be considered effective indicators of soil quality changes resulting from environmental stress or management practices.

The aim of the studies was to evaluate the effects of azadirachtin on dehydrogenase and catalase enzyme activity in soil. Specific objectives were as follows: (i) to determine the effects of different azadirachtin application doses on dehydrogenase (DHA) and catalase (CA) activity, and (ii) to determine changes in the DHA and CA were determined in soil samples taken in 7, 14 and 21 days after the field experiment.

Materials and Methods

Experimental field and climate: The field experiment was conducted at the Experimental Station of Perm State Agricultural Academy, Perm, Russia (57^{0}56′00″ N, 56^{0}14′59″ E) at an altitude of 127 m above mean sea level (Figure 1). The data on climatic parameters such as precipitation and temperature are shown in Figure 2.

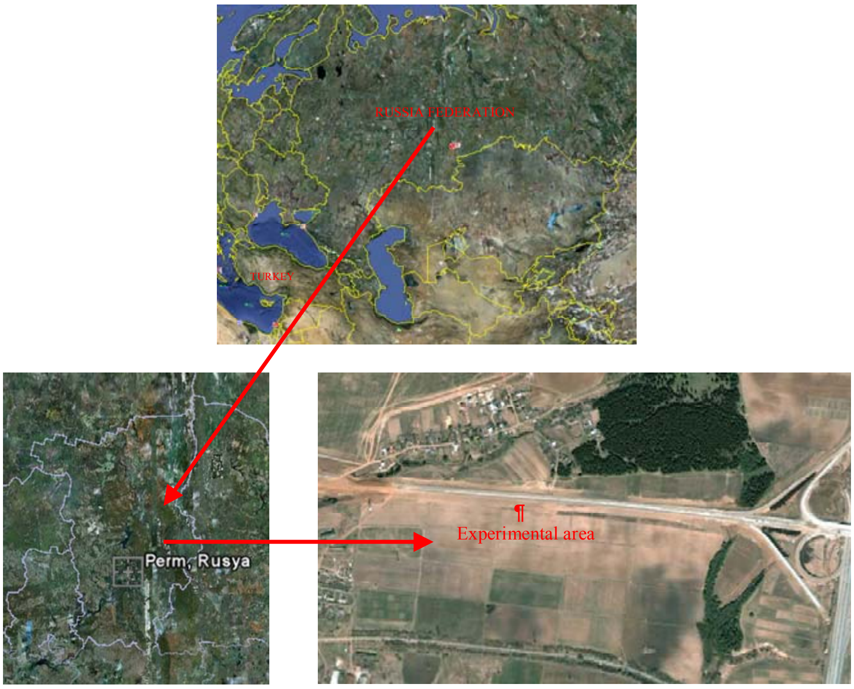


Figure 1. Location of the experimental field in Perm, Russia

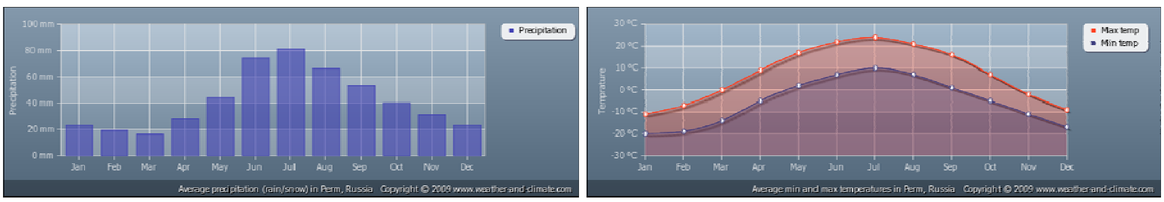


Figure 2. Climatic data in Perm, Russia

Soil: The soil of the experimental site is loam (31.4% sand, 45% silt, 23.6% clay). A composite surface soil sample from 0-20 cm depth was collected from the experimental site before initiating the experiment and was analyzed for physicochemical properties according to Rowell (1996) and Jones (2001). Soil samples were air dried at room temperature; sieved with <2 mm screen. The basic physico-chemical characteristics of the soil are as follows: pH (1:1, soil:water): 6.70, electrical conductivity (1:1, soil:water): 0.81 dSm^{-1}; $CaCO_3$ content: 0.04%; total organic carbon: 0.99%; total nitrogen: 0.086%;

available phosphorus (0.5M $NaHCO_3$ extractable P): 13.34 $mg.kg^{-1}$, and exchangeable potassium (1N NH_4OAc extractable K): 1.382 $cmol(+).kg^{-1}$. The soils had no history of receiving any pesticide treatment six months prior to this study. Experimental soil was classified as "Albic Luvisol" according to the FAO (2006).

Azadirachtin ($C_{35}H_{44}O_{16}$) : The azadirachtin (NeemAzal®–T/S) was imported by VIT, Turkey. This insecticide (10g azadirachtin L^{-1}) was used as technical and added to soil.

Experimental design: This experiment was conducted to determine the effects of azadirachtin on soil enzyme activities under field conditions. Experimental design was a randomized plot design with three replications, and was established June 26, 2011. Each plot was an area of 1 x 1 m. The treatments were: (1) control: 0 mL azadirachtin da^{-1}, (2) low application doses: 15 mL azadirachtin da^{-1}, (3) recommended application doses: 30 mL azadirachtin da^{-1}, (4) high application doses: 60 mL azadirachtin da^{-1}, respectively. In order to homogenoeus azadirachtin application in soil, azadirachtin were applied in 2.5 L water per m^2. Changes in the dehydrogenase and catalase activities were determined in soil samples taken in 7, 14 and 21 days after the field experiment was conducted.

Soil sample preparation: Field moist soils were collected and brought to the laboratory in properly labeled and sealed polythene bags. The sieved soil samples (<2 mm) were homogenized and kept in polyethylene boxes, and also stored at 4 ^{0}C until the analyses were carried out. The acclimatized soil samples were used for the enzyme analyses.

Enzyme analyses: To assess the enzyme analyses to the adjustments in the soil variables, with and without pesticides, enzyme analyses were estimated by following methods.
Dehydrogenase activity (DHA) was determined according to Pepper et al (1995). To 6 g of sample 30 mg glucose, 1 ml of 3% TTC (2,3,5-triphenyltetrazoliumchlorid) solution and 2.5 ml pure water were added and the samples were incubated for 24 h at 37^{0}C. The formation of TPF (1,3,5 triphenylformazan) was determined spectrophotometrically at 485 nm and results were expressed as µg TPF g^{-1} dry soil.
Catalase activity (CA) was measured by the method of Beck (1971). Ten ml of phosphate buffer (pH, 7) and 5 ml of a 3% H_2O_2 substrate solution were added to 5 g of soil. The volume (ml) of O_2 released within 3 minutes at 20^{0}C was determined. Three replicates of each soil were tested and controls were tested in the same way, but with the addition of 2 ml of 6.5% (w/v) NaN_3. Results were expressed as ml O_2 g^{-1} dry soil.

Statistical analysis: All data were analyzed using SPSS 11.0 statistical software (SPSS Inc.). Analysis of variance (ANOVA) was carried out using one-factor randomized complete plot design; where significant F-values were obtained, differences between individual means were tested using the LSD (Least Significant Difference) test, with a significance level of $P<0.01$. All figures and tables presented include standard deviations of the data and F-values. The asterisks, * and ** indicate significance at $P<0.05$, and 0.001, respectively.

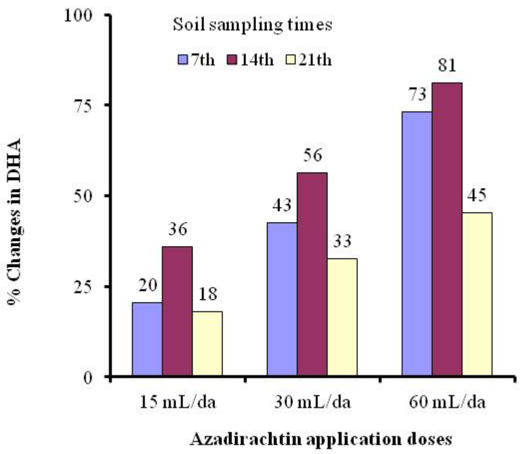
Results and Discussion

Dehydrogenase (DHA) and catalase (CA) activity varied significantly in response to azadirachtin application doses over time (Table 1). The DHA and CA in different doses of azadirachtin applied soils during the experiment are shown in figure 3.

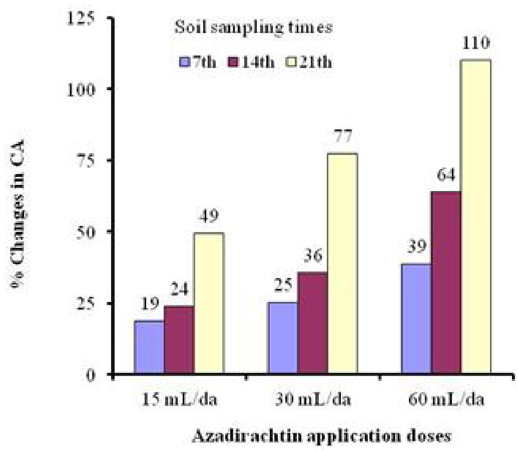
Table 1. Azadirachtin impacts on dehydrogenase and catalase activity in soil at different sampling times

Enzyme activity	Soil sampling times	Azadirachtin application doses			
		Control	15 mL da^{-1}	30 mL da^{-1}	60 mL da^{-1}
DHA	7 days	101,01 (4,02)	121,59 (3,68)	144,08 (6,37)	174,95 (6,37)
	14 days	91,48 (4,57)	124,26 (2,88)	142,93 (7,50)	165,80 (5,72)
	21 days	99,10 (6,99)	117,01 (3,49)	131,50 (5,72)	144,08 (4,12)
CA	7 days	40,14 (2,04)	47,71 (2,06)	50,29 (3,28)	55,72 (2,88)
	14 days	40,50 (2,72)	50,15 (3,55)	54,94 (3,10)	66,38 (5,53)
	21 days	40,91 (2,40)	61,14 (1,60)	72,59 (2,07)	85,97 (2,28)

Notes: Standard deviation are shown in parentheses.
DHA = Dehydrogenase activity (µg TPF g^{-1} dry soil), CA = Catalase activity (ml O_2 g^{-1} dry soil)



(a)



(b)

Figure 3. The changes of DHA and CA in different doses of azadirachtin applied soils
a) Dehydrogenase activity (DHA) b) Catalase activity (CA)

Considerable variations in DHA and CA were found for the different doses of azadirachtin at different sampling times. Statistically significant variations were found in DHA and CA at various azadirachtin application doses. The DHA and CA were also affected by incubation period. The analysis of variance of the results obtained in our experiment on the periodic sampling times with azadirachtin showed that all factors (azadirachtin doses and soil sampling times) significantly influenced DHA and CA (Table 2). After azadirachtin application a rapid and significant increase in DHA and CA were observed in soils. At the end of the sampling times, the DHA and CA measured in azadirachtin applied soils were statistically different from those measured in the control soils.

Biological oxidation of organic compounds is a dehydrogenation process (Tabatabai 1982); mediated by many different intracellular and specific dehydrogenases. Therefore, dehydrogenase activity (DHA) of soil is supposed to reflect microbial activity (Skujins 1976; Kumar and Tarafdar, 2003). The activity of dehydrogenase is considered an indicator of the oxidative metabolism in soils and thus of the microbiological activity (Skujins 1973), because, being exclusively intracellular, it is linked to viable cells (Quilchano and Maranon, 2002; Kızılkaya, 2008). At all sampling times, significant positive effects were observed for all azadirachtin application doses. Dehydrogenase activity was found to be max. stimulated by doses of 600 mL azadirachtin da^{-1}. The maximum increase value was 81% (600 mL azadirachtin da^{-1}, 21 days after application) (Figure 3a). The experimental data are consistent with results reported by other authors on the effect of different pesticides on this enzyme. Dinelli et al. (1998) and Accinelli et al. (2002) reported that sulfonylurea herbicides at a rate up to 20 mg kg-1 inhibited dehydrogenase activity. The results Radivojević et al. (2008) showed a decreased activity of dehydrogenase under all atrazine concentrations (8.0, 40.0 and 80.0 mg kg^{-1}) from the 1st to the 30th day after atrazine application. The decrease ranged: 12.5-18.2% for 8.0 mg concentration, 4.8-24.8% for 40.0 mg, and 6.6-39.6% for 80.0 mg.

Table 2. Results of ANOVA

Variables	DHA		CA	
	F-value	$LSD_{\alpha=0.01}$	*F*-value	$LSD_{\alpha=0.01}$
AAD	243.440***	6.994	145.020***	3.996
SST	17.426***	6.057	98.740***	3.461
AAD x SST	6.419***	12.113	14.037***	6.922

AD = Azadirachtin application doses, Soil sampling times
^{ns} not significant * P<0.05 , ** P<0.01, *** P<0.01

Biological oxidation of organic compounds is a dehydrogenation process (Tabatabai 1982); mediated by many different intracellular and specific dehydrogenases. Therefore, dehydrogenase activity (DHA) of soil is supposed to reflect microbial activity (Skujins 1976; Kumar and Tarafdar, 2003). The activity of dehydrogenase is considered an indicator of the oxidative metabolism in soils and thus of the microbiological activity (Skujins 1973), because, being exclusively intracellular, it is linked to viable cells (Quilchano and Maranon, 2002; Kızılkaya, 2008). At all sampling times, significant positive effects were observed for all azadirachtin application doses. Dehydrogenase activity was found to be max. stimulated by doses of 600 mL azadirachtin da^{-1}. The maximum increase value was 81% (600 mL azadirachtin da^{-1}, 21 days after application) (Figure 3a). The experimental data are consistent with results reported by other authors on the effect of different pesticides on this enzyme. Dinelli et al. (1998) and Accinelli et al. (2002) reported that sulfonylurea herbicides at a rate up to 20 mg kg-1 inhibited dehydrogenase activity. The results Radivojević et al. (2008) showed a decreased activity of dehydrogenase under all atrazine concentrations (8.0, 40.0 and 80.0 mg kg^{-1}) from the 1st to the 30th day after atrazine application. The decrease ranged: 12.5-18.2% for 8.0 mg concentration, 4.8-24.8% for 40.0 mg, and 6.6-39.6% for 80.0 mg.

The CA is sensitive to both natural and O_2 level, and shows a quick response to induced changes. Also it may be affected by cast formation by earthworm in anaerobic condition. The CA is based on the rates of oxygen release from the added hydrogen peroxide, and may be related to the metabolic activity of aerobic organisms (Glinsky et al., 1986; Kızılkaya et al., 2004; Kızılkaya and Hepşen, 2007). The changes in CA as influenced by the application of azadirachtin are presented in Table 1 and Figure 3b. All application doses where an azadirachtin was applied were found to increase the CA significantly in comparison with control. The maximum increase value was 110% (600 mL azadirachtin da^{-1}, 21 days after application).

Conclusion

In this investigation, azadirachtin was applied at different concentrations on the soil for 21 days. Short-term changes or stimulation were observed in the activities of the enzymes studied. However, at the end of the experimental period, these activities were significantly stimulated. The present findings mean that the azadirachtin is only relative safe pesticides which could not cause environmental risk and would not cause an ecological problem from the microbial point of view.

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Evaluation of physiological growth stage of faba bean to drought

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Abstract: This study was carried out during fall season of (2003 - 2004) at Danadan Research Station , Mosul University , Mosul city center. The experiment were included in this investigation to evaluate the response of faba bean cultivars namely (French, Syrian, Tacka357, Towaytha and Babylon) and therefore the experiment was conducted to study the designed to detect which plant physiological stage is the most sensitive to water stress and thus complementary watering was ceased either during (Vegetative, Flowering or pod growth and seed filling stages). The results showed that Pod development and filling stage appeared to be the most water stress sensitive stage a sit showed a reduction of (97.91 cm) in plant height and (21.08 cm) in pod length and (4.78) in number of seed / pod and (559.76 g-m^{-2}) in dry pods weight and (434.39 g-m^{-2}) in dry seed yield and (144.53 g) in 100 seed weight as compared to check. So we can concluded that we have to irrigate the plants very well till ripening of pods and seed to get a good quality. French and Syrian cultivars might be classified as semi conservative cultivars since they have the ability to keep partial stomata close for CO_2 exchange to match with normal photosynthesis accompanied by significant transpiration reduction, while Towaytha and Babylon cultivars appeared to be conservative cultivars. The determinate Tacka 375 cultivar could be categorized as drought escapable cultivar since it matures nearly more than two weeks before the other corresponding cultivars.

Introduction

Faba bean *Vicia faba* L. crop is Mediterranean basin origin (Summer field and Roberts, 1985). However, faba bean production in this region facing serious problems owing to the synchronization of rapid temperature increases accompanied by low rainfall incidences with fruit setting , pod development and seed filling at the end of the growing season (Loss and Siddique, 1996 and Abdel, 1997).China is the highest faba bean producer and consumer as well where it produces (2.7 ton.year^{-1}) which constitute (65%) of the overall global faba bean production , followed by Ethiopia (9%) and Egypt in the third order as it produces (262 ton.year^{-1}). However, in Iraq, faba bean is a secondary crop and it consumed as boiled green mature pods or boiled seed (Abdel, 1993). Recently, (consumptions are increased since it possesses a relatively high protein content 23-42% (Stan, 1997), and considered the highest protein producing legume crops owing its higher yield 5 t.ha^{-1} (Schulz et al., 1999).

In area where supplementary irrigation is required such as Northern Iraqi provinces where the water demand at varying physiological phases is of high significance in order to aid faba bean plants during the critical stage to ovoid yield reductions (Abdel, 1997). Subsequently Faba bean growth was categorized to vegetative, flowering, fruit setting, pod swelling and seed filling phases; it was found that pod swelling and seed filling were the most critical drought susceptible stages (El-Nadi et al, 1969). However , vegetative stage was found to be less effective and manifested the recovering capability (El-Nadi, 1970) , followed by flowering stage which was found to be less sensitive than on coming stage owing to the ability of Faba bean plants to produce profuse number of flower where setting of (10%) of these flower may be adequate to ensure sufficient pod number. Therefore, pod swelling and seed fillings are the most drought susceptible stages where watering should be applied if which otherwise yield would be greatiy reduced (Doss et al., 1974 and Milburn, 1974).

Sustaining adequate watering during pod swelling and seed filling stages resulted in maximum faba bean yield (French et al., 1976; Sprent et al., 1977; Nairizi and Rydzewski, 1977). Moreover Stock (1977) obtained the highest yield with adequate watering from flowering to harvesting. Faba bean production risk maintained throughout the growing season unless the seeds are completely matured (El-Sarrag et al., 1988; Abdel, 1993). Yield reducation was reported when plants were exposed water scarcity during flowering stage (Sparrow et al., 1995; Sanders, 1997). The attributed their results to the influence of drought on nitrogenase activity. Mwanamwenge et al. (1999) confirmed the significance of irrigating faba bean plants during pod swelling and seed fillings.

The objective of this investigation was to detect the yield reduction that resulted from irrigation casement during varying growth phases of five faba bean cultivars in order to improve water consumptive use and water use efficiency.

Materials and methods

This experiment was carried out during fall season of (2002-2003) at Danadan Field of Research, Mosul University, to evaluate the response of five faba bean cultivars namely (Aquadulce, Local Syrian, Taka 357, Towatha and Babylon). Seed were obtained from Atomic Energy Organization, Baghdad.

Split Plot within Factorial Randomized Complete Block Design (split F-RCBD) was chosen where supplementary irrigation represented the main plot (A): (a_1 RWW) supplementary irrigation was ceased during the vegetative stage (from germination till the appearance of first flower) , (a_2 WRW) supplementary irrigation was ceased during the flowering stage (the appearance of first flower to the commencement of pod setting) , (a_3 WWR) no irrigation during the pod swelling and seed fillings stage and (a_4 WWW) continuous irrigation was applied throughout the growing sesson where plants were irrigated whenever (25%) of available water capacity of soil was depleted to a depth of (30 cm). The Sub main plot (B) was dedicated to the above mentioned five faba bean cultivars. Supplementary, (20 treatments) were included in this experiment, each treatment was replicated three times and a replicate was represented by a furrow of (0.75 m) width and (4 m) length planted on both sides with (0.2 m) plant intra space.

Filed soil was plowed twice horizontally and once more vertically then dissected to fit the experimental design. Gypsum block were positioned at each replicate to (25 and 50 cm) soil depth (Heerman and Juma, 1993; Ruggiero et al., 1999), in order to truck soil moisture fluctuation throughout the growing season. Finally, main plots were separated by (4 m) spaces to avoid water seepage among them.

Seed were sown on(November 5^{th} 2003) then plants were thinned to two plants per hill. Weeds were manually eradicated throughout the growing season, thereafter; plants were fertilized twice by (NP 27:27) at rate of (150 $kg.ha^{-1}$) on December (20^{th} 2003) and repeated on (February 10^{th} 2004) (Cochran and Schlentner, 1995). Benomyl systematic fungicide was applied as protective spray at rate of (1 $ml.l^{-1}$) (McEwen and Yeoman, 1979). Malathion at rate of (2 $ml.l^{-1}$) was sprayed to control Black Aphid. Parameters were taken from (10) selected plants from each replicated. Therefore , Plant height (cm) , Branches number per plant , Leaf area index , Flowering nodes number on main stem , Flower number per inflorescence , Setting percentage , Pod number on main stem , Pod length (cm) , Seed number per pod , Fresh weight of vegetative parts and mature pods ($g.m^{-2}$) , weight of mature pods ($g.m^{-2}$) , Pod dry weight ($g.m^{-2}$) , Yield of dry seed ($g.m^{-2}$) and weight of 100 seeds (g) were recorded.

Results and discussion

The influence of ceasing supplementary irrigation during varying physiological phases of growth on vegetative growth of faba bean.

The obtained results (Table, 1) revealed that sustaining adequate irrigation during the entire growth (WWW) Substantially increased plant height (11.1%) in relation to (WRW) , branches number (12.2%) as compared to (RWW). Results also confirmed that exposing faba bean plants to water scarcity during flowering (WRW) and pod swelling and seed fillings (WWR) stages highly reduced all detected parameters. These results can be attributed to the plant capability to recover the adverse of drought that occurs at early vegetative growth since they're going to be enough time for curing and substituting the lost growth (Abdel, 1982; Sparrow et al., 1995).

Towatha cultivar gave the highest plant height (114.89 cm) , as compared to other cultivar. Syrian cultivar significantly exceeded Taka 357 in leaf area index (28.64). The lowest vegetative growth was accompanied to the determinate Taka 357 cultivar. It gave the lowest plant height (50.68 cm) , leaf area index (10.71). However it manifested the highest branches number per plant (13.41) and thus this cultivar showed lower completion ability as compared to other cultivar, despite its highest branching Taka 357 still cannot complete other cultivar growths.

Dual interaction results exhibited that the Towatha (RWW) was the best treatment in term of branches number per plant (6), whereas under (WWW) Aquadulce cultivar was the best in term plant height (123.10 cm), and under (WWW) Syrian cultivar was the best in term leaf area index (33.65). The worst results were found in Taka 357 (RWW) treatment in term of plant height (49.66 cm) , and leaf area index (9.91) with ((WWR). It is worthy to recall that this cultivar gave the highest branches number per plant (14.33) under (WWW).

From this study it could be inferred that drought possesses direct influences on vegetative growth traits. However the influences during early growth stage was slight and being more severe through out the growing season particularly in lattes growth of plants were fruit setting , pod swelling and seed fillings. These finding were in agreement is with those reported by (El-Beltagy and Hall, 1974; Moursi et al., 1978; Obroucheva, 1992; Link et al., 1999). They found slight effects of drought when faba bean plants were exposed to drought and they attributed their results to the substantial influences of drought on cell expansion sand exiguously affects on cell divisions. Therefore stunting was coincided to droughted plants which showed closer internodes as compared to well irrigated plants.

Table 1. The influence of ceasing supplementary irrigation during varying physiological phases of growth on vegetative growth of five varieties of beans during the growing season (2003-2004).

1- Plant height (cm)

cultivars	WWW	RWW	WRW	WWR	Mean
Aquadulce	123.10 a	117.53 a-c	108.96 b-d	107.46 b-e	114.26 a
Local Syrian	119.13 ab	113.26 a-d	104.66 de	111.83 a-d	112.22 ab
Taka 357	52.23 f	49.66 f	50.16 f	50.66 f	50.68 c
Towatha	119.80 ab	112.86 a-d	113.53 a-d	113.36 a-d	114.89 a
Babylon	118.93 a-c	109.30 b-d	96.43 e	106.23 c-e	107.72 b
Mean	106.64 a	100.52 b	94.75 c	97.91 bc	99.95

2-Branches number per plant

cultivars	WWW	RWW	WRW	WWR	Mean
Aquadulce	7.33 b-d	7.00 b-d	9.33 b	6.66 cd	7.58 b
Local Syrian	9.33 b	8.00 b-d	7.00 b-d	9.00 bc	8.33 b
Taka 357	14.33 a	12.33 a	13.00 a	14.00 a	13.41 a
Towatha	7.66 b-d	6.00 d	7.66 b-d	8.00 b-d	7.33 b
Babylon	7.66 b-d	7.33 b-d	6.66 cd	7.33 b-d	7.25 b
Mean	9.26 a	8.13 b	8.73 ab	9.00 ab	8.78

3-Leaf area index

cultivars	WWW	RWW	WRW	WWR	Mean
Aquadulce	27.60 a-e	22.08 de	31.02 a-c	23.07 c-e	25.94 ab
Local Syrian	33.65 a	28.30 a-d	20.44 de	32.16 ab	28.64 a
Taka 357	11.39 f	9.92 f	11.62 f	9.91 f	10.71 d
Towatha	26.47 a-e	22.08 de	24.47 b-e	23.25 c-e	24.07 bc
Babylon	19.74 de	23.58 c-e	20.52 de	19.42 e	20.81 c
Mean	23.77 a	21.19 a	21.61 a	21.56 a	22.03

* Means followed by the same letter within a column do not differ significantly from each other using Duncan's Multiple Range Test at 5% level.

The influence of ceasing supplementary irrigation on flowering of faba bean.

The obtained results (table, 2) confirmed no variation in detected flowering traits in adequate irrigation treatments. These results might be referred to adequate rainfall incidences during flowering stage which synchronized with February to the commencement of March. On other hand significant differences in cultivar responses were observed in flowering traits. These differences might be due to cultivar growth habits genome variation and cultivar capabilities in cold resistance were plants exposed to three days period chilling (14.8 C^o) in February.

These chilling period may adversely influenced on some cultivars since there were no significant differences were observed among Aquadulce , Syrian , Towatha and Babylon despite their superiority over Taka 357 in term of flowering nodes number on main stem by (70.9 , 69.8 , 71.5 , and 71.1%) respectively and in flower number per inflorescence by (44.5 , 46.7 , 43.3 and 42.4%)

respectively. However Taka 357 exceeded other cultivars in setting percentage. These may be due to lower number of flowering nodes number on main stem (4.41) and to lower flower number per inflorescence (2.23) as compared these produced on other cultivars. Since Taka 357 is a determinate cultivar. Similar results were reported in previous studies (Chapman and Peat, 1978; Pilbeam et al., 1990).

Ceasing supplementary irrigation and adequately irrigated Towatha cultivar combined with (WWW and WRW) manifested the highest number of flowering nodes number on main stem (16.66), Syrian cultivar combined with (WWR) showed the highest flower number per inflorescence (4.32). However Taka 357 exhibeted the lowest values number of flowering nodes number on main stem (4) under (RWW and WRW) ; lowest flower number per inflorescence (2.09) under (RWW). However it revealed the highest setting percentage (68.52%) under (RWW). Additionally it significant exceeded Babylon (WRW) in setting percentage (88.2%). Very close results were (Abdel, 1997) who found setting reduction in Aquadulce particularly during low tempeture owing to the low pollination and fertilizing capability resulting from poor pollen tube germination and poor reception of these tubes by stigma besides low insect and bee populations.

Table 2. The influence of ceasing supplementary irrigation during varying physiological phases of growth on flowering growth of five varieties of beans during the growing season (2003-2004).

1-Flowering nodes number on main stem

cultivars	WWW	RWW	WRW	WWR	Mean
Aquadulce	14.00 a	15.00 a	16.33 a	15.33 a	15.16 a
Local Syrian	16.00 a	14.00 a	14.33 a	14.00 a	14.58 a
Taka 357	4.33 b	4.00 b	4.00 b	5.33 b	4.41 b
Towatha	16.66 a	14.33 a	16.66 a	14.33 a	15.50 a
Babylon	14.66 a	16.00 a	14.66 a	15.66 a	15.25 a
Mean	13.13 a	12.66 a	13.20 a	12.93 a	12.98

2- Flower number per inflorescence

cultivars	WWW	RWW	WRW	WWR	Mean
Aquadulce	4.15 ab	4.00 ab	4.28 a	3.67 ab	4.02 a
Local Syrian	4.01 ab	4.16 ab	4.25 a	4.32 a	4.18 a
Taka 357	2.39 c	2.09 c	2.25 c	3.18 c	2.23 b
Towatha	3.74 ab	3.70 ab	4.17 ab	4.11 b	3.93 a
Babylon	3.61 ab	4.26 a	4.14 ab	3.47 b	3.87 a
Mean	3.58 a	3.64 a	3.82 a	3.55 a	3.65

1- Setting percentage

cultivars	WWW	RWW	WRW	WWR	Mean
Aquadulce	19.43 c	10.44 c	8.09 c	19.26 c	14.30 b
Local Syrian	11.98 c	10.36 c	8.96 c	13.46 c	11.19 b
Taka 357	52.40 b	68.52 a	60.19 ab	46.77 b	56.97 a
Towatha	13.17 c	13.31 c	13.82 c	16.97 c	14.32 b
Babylon	13.86 c	9.73 c	8.07 c	14.54 c	11.55 b
Mean	22.17 a	22.47 a	19.82 a	22.20 a	21.67

* Means followed by the same letter within a column do not differ significantly from each other using Duncan's Multiple Range Test at 5% level.

Effect of ceasing supplementary irrigation at varying phases on pod development traits of faba bean.

Tabulated results (table, 3) revealed that continuous watering (WWW) gave the highest pod length (23.26 cm), white (RWW) showed the highest seed number per pod (5.36). However the worst results were confined to (WWR) in pod length (21.08 cm) and seed number per pod (4.78).

Significant differences were not detected among Aquadulce, Syrian, Towatha and Babylon cultivars. Furthermore these cultivars were significantly exceeded Taka 357, where Towatha exceeded the later cultivar in pod number on main stem (36.9%) and Syrian in term of pod length (17.5%) and seed number per pod (35.3%).

The highest pod number on main stem (11) was accompanied by Aquadulce adequately irrigated (WWW) , pod length (26.93 cm) was confined to Syrian (WWW) and Syrian (RWW) revealed the highest seed number per pod (6.40). The lowest pod length (20.16 cm) and seed number per pod (3.60) was found with Taka 357 (RWW).

Results is (table, 3) confirmed the adverse effects of ceasing watering out pod swelling and seed filings on detected parameters. These results were in agreements with those obtained by Sparrow et al., (1995).

They confirmed the susceptibility of flowering and pod swelling stagesto drought since exposing faba bean plants to water stress at these stages apparently reduce the Nitrogenase activity which negatively reflected on plant performance. Reductions were also attributed to the low photosynthesis and assimilate production under drought at these stages (Robert et al., 1990; Ravi et al., 1996).

Table 3. The influence of ceasing supplementary irrigation during varying physiological phases of growth on pod development traits of five varieties of beans during the growing season (2003-2004).

1-Pod number on main stem

cultivars	WWW	RWW	WRW	WWR	Mean
Aquadulce	11.00 a	6.33 cd	5.66 d	10.33 ab	8.33 ab
Local Syrian	7.66 a-d	6.00 cd	5.33 d	7.66 a-d	6.66 bc
Taka 357	5.33 d	5.66 d	5.33 d	5.33 d	5.41 c
Towatha	8.66 a-d	7.00 b-d	9.00 a-d	9.66 a-c	8.58 a
Babylon	7.33 a-d	6.33 cd	5.00 d	8.00 a-d	6.66 bc
Mean	8.00 a	6.26 b	6.06 b	8.20 a	7.13

2- Pod length (cm)

cultivars	WWW	RWW	WRW	WWR	Mean
Aquadulce	22.23 d-h	22.36 d-h	22.76 c-f	20.40 hi	21.94 b
Local Syrian	26.93 a	26.13 ab	24.63 bc	22.53 d-g	25.05 a
Taka 357	20.96 e-h	20.16 i	21.03 e-i	20.53 g-i	20.67 c
Towatha	23.00 c-e	23.16 cd	20.80 f-i	20.96 e-i	21.98 b
Babylon	23.16 cd	22.43 d-h	22.33 d-h	20.96 e-i	22.22 b
Mean	23.26 a	22.85 ab	22.31 b	21.08 c	22.37

3- Seed number per pod

cultivars	WWW	RWW	WRW	WWR	Mean
Aquadulce	5.00 c-g	5.76 a-d	5.20 c-f	4.53 f-h	5.12 b
Local Syrian	6.06 ab	6.40 a	6.06 ab	5.73 a-d	6.06 a
Taka 357	3.73 i	3.60 i	4.23 g-i	4.13 hi	3.92 c
Towatha	5.43 b-e	5.23 c-f	4.93 d-h	4.93 d-h	5.13 b
Babylon	5.06 c-f	5.83 a-c	5.33 b-f	4.60 e-h	5.20 b
Mean	5.06 ab	5.36 a	5.15 a	4.78 b	5.09

Means followed by the same letter within a column do not differ significantly from each other using Duncan's Multiple Range Test at 5% level.

Effect of ceasing supplementary irrigation at varying physiological stages on yield components of faba bean

The results (table, 4) confirmed that exposing faba bean plants to drought during pod swelling and seed filling stage (WWR) resulted in negative effected on yield components, where droughted plants gave the lowest fresh weight of vegetative parts and mature pods (4796.84 $g.m^{-2}$), weight of mature pods (2388.58 $g.m^{-2}$), pod dry weight (559.76 $g.m^{-2}$) and weight of 100 seeds (144.53 g). On the other hand continuous irrigation (WWW) supplementary exceeded that of (WWR) in all detected parameters accept weight of 100 seeds which revealed the highest values in fresh weight of vegetative parts and mature pods (7356.97 $g.m^{-2}$), weight of mature pods (4068.77 $g.m^{-2}$), pod dry weight (838.01 $g.m^{-2}$) and yield of dry seed (637.10 $g.m^{-2}$), whereas weight of 100 seeds was highest in (WRW) (165.26 g). However, significant differences were not found between (WWW) and (RWW) treatments.

Aquadulce and Syrian cultivars manifested the highest responses to all water stressed stages since there were in significant differences were found in all detected characteristics. Aquadulce exhibited the highest pod dry weight (923.39 $g.m^{-2}$), yield of dry seed (714.97 $g.m^{-2}$) and weight of 100 seeds (157.88 g). Syrian cultivar also revealed the highest fresh weight of vegetative parts and mature pods (7752.66 $g.m^{-2}$), weight of mature pods (4266.94 $g.m^{-2}$). Towatha and Babylon showed very close moderate results. However, Taka 357 was the worst since it manifested the lowest values accept weight of 100 seeds which was superior over Aquadulce by (6.5%).

Continuous watering (WWW) of Syrian cultivar showed superiority over other treatments in all investigated parameters accept that of 100 seeds weight which was confined with Towatha (WRW). However this superiority was not significant in relation to Syrian cultivar. Syrian (WWW) treatment which showed the highest fresh weight of vegetative parts and mature pods (10639.68 $g.m^{-2}$) , weight of mature pods (5978.39 $g.m^{-2}$) , pod dry weight (1172.03 $g.m^{-2}$) and yield of dry seed (877.97 $g.m^{-2}$). The lowest values were confined with Taka 357 (RWW) in term of fresh weight of vegetative parts and mature pods (2629.03 $g.m^{-2}$) , weight of mature pods (1136.45 $g.m^{-2}$) , pod dry weight (161.29 $g.m^{-2}$) , yield of dry seed (100 $g.m^{-2}$) and weight of 100 seeds (136.21g).

The most susceptible physiological stages was pod swelling and seed filling stage (WWR). This sensitivity resulted from ceasing supplementary irrigation which synchronized with low rainfall in cidances. In this stage faba bean plant growth, pod formation, pod swelling and seed filling were combined with each others (Xia, 1997; Stan, 1997; Mwanamwenge et al., 1999). These results were in agreement with those confirmed by Doss et al. (1974), Milburn (1974) and Sprent et al. (1977). They stated that seed filling and pod swelling were found to be the most sensitive stage to drought. Exposing faba bean plants to water stress at this stage resulted in significant pod shedding and yield reduction. Mahoney (1991), McDonald and Paulsen (1997), Chmielewski and Kohn (1999) attributed faba bean yield reduction that caused by water stress at pod swelling and seed filling stage to the direct influence of drought on flowering and setting of pods. Moreover water stress also adversely influences nitrogenase activity and its capability in nitrogen fixation which negatively reelected on yield reduction (Sparrow et al., 1995) and to the combination of water stress and high temperature during this stage (Loss and Siddique, 1996; Chmielewski and Kohn, 1999).

Table (4). The influence of ceasing supplementary irrigation during varying physiological phases of growth on yield components of five varieties of beans during the growing season (2003-2004).

1- Fresh weight of vegetative parts and mature pods ($g.m^{-2}$)

cultivars	WWW	RWW	WRW	WWR	Mean
Aquadulce	7381.61 b-e	8784.84 ab	7037.74 b-f	5220.32 d-i	7106.13 ab
Local Syrian	10639.68 a	7387.10 b-e	6548.39 b-f	6435.48 b-f	7752.66 a
Taka 357	3091.29 hi	2629.03 i	3636.45 g-i	2852.90 i	3052.42 c
Towatha	7940.97 bc	5903.23 c-g	5570.00 c-h	4889.35 e-i	6075.89 b
Babylon	7731.29 b-d	8215.16 a-c	5070.00 d-i	4586.13 f-i	6400.65 b
Mean	7356.97 a	6583.87 ab	5572.52 bc	4796.84 c	6077.55

2- weight of mature pods ($g.m^{-2}$)

cultivars	WWW	RWW	WRW	WWR	Mean
Aquadulce	4397.74 b-d	5333.23 ab	3891.29 b-g	2796.77 e-i	4104.76 ab
Local Syrian	5978.39 a	4231.29 b-e	3718.39 c-h	3139.68 c-i	4266.94 a
Taka 357	1263.55 j	1136.45 j	1639.68 ij	1191.29 j	1307.74 d
Towatha	4112.90 b-f	3320.97 c-h	2925.81 d-i	2551.61 g-j	3227.82 c
Babylon	4591.29 a-c	4679.68 a-c	2630.00 f-j	2263.55 h-j	3541.13 bc
Mean	4068.77 a	3740.32 a	2961.03 b	2388.58 b	3289.68

3- Pod dry weight ($g.m^{-2}$)

cultivars	WWW	RWW	WRW	WWR	Mean
Aquadulce	973.13 ab	968.81 ab	941.94 a-c	809.68 b-d	923.39 a
Local Syrian	1172.03 a	942.48 a-c	867.74 bc	662.58 cd	911.21 a
Taka 357	201.84 f	161.29 f	322.68 ef	191.06 f	219.22 c
Towatha	869.90 bc	672.58 cd	724.74 b-d	558.06 de	706.32 b
Babylon	973.13 ab	969.90 ab	769.35 b-d	577.42 de	822.45 ab
Mean	838.01 a	743.01 a	725.29 a	559.76 b	716.52

4- Yield of dry seed ($g.m^{-2}$)

cultivars	WWW	RWW	WRW	WWR	Mean
Aquadulce	741.94 a-c	748.39 a-c	730.10 a-c	639.45 b-e	714.97 a
Local Syrian	877.97 a	740.32 a-c	670.00 a-d	506.77 de	698.77 a
Taka 357	137.65 f	100.00 f	227.97 f	133.97 f	149.90 c
Towatha	680.10 a-d	530.10 c-e	569.90 b-e	445.71 e	556.45 b
Babylon	747.84 a-c	761.29 ab	597.84 b-e	446.03 e	638.25 ab
Mean	637.10 a	576.02 a	559.16 a	434.39 b	551.67

5- weight of 100 seeds (g)

cultivars	WWW	RWW	WRW	WWR	Mean
Aquadulce	150.29 b-f	165.35 a-c	167.32 ab	148.59 b-f	157.88 a
Local Syrian	145.48 d-f	166.70 ab	163.53 a-d	142.26 ef	154.49 ab
Taka 357	146.69 c-f	136.21 f	165.72 a-c	142.11 ef	147.68 b
Towatha	142.48 ef	155.72 a-e	170.53 a	144.32 d-f	153.26 ab
Babylon	157.93 a-e	148.91 b-f	159.23 a-e	145.38 d-f	152.86 ab
Mean	148.57 bc	154.58 b	165.26 a	144.53 c	153.23

- Means followed by the same letter within a column do not differ significantly from each other using Duncan's Multiple Range Test at 5% level.
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Soil Exchangeable Cations: A Geostatistical Study from Russia

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Abstract
In present study, geostatistical techniques was applied to assess the spatial variability of exchangeable cations such as; calcium (Ex-Ca^{2+}), magnesium (Ex-Mg^{2+}), potassium (Ex-K^{+}) and sodium (Ex-Na^{+}) in the tillaged layer in a Perm State Agricultural Academy Farm site in Perm region, West Urals, Russia. A 250x100 m plot (approximately 2.35 ha) was divided into grids with 25x25 m spacing that included 51 sampling points from 0-0.2 m in depth. Soil reaction (pH) was the least variable property while the Ex-K was the most variable. The greatest range of influence (237.6 m) occurred for Ex-Ca and the least range (49.7 m) for Ex-Mg.
Keywords: exchangeable cations, spatial variability, site specific management

Introduction

Soil properties of terrestrial ecosystems are controlled by a variety of factors that operate at different spatial and temporal scales. Soil physical, chemical and biological properties are all likely to change markedly across small distances, within a few hectares of agricultural fields (Benayas et al., 2004; Cambardella et al., 1994; Chien et al., 1997). The small-scale variability may be difficult to measure and not apparent to the casual observer. Analysis of small-scale variability has practical uses in managing soil fertility for a chosen field (Brady and Weil, 2002). The five cations in soils are calcium (Ca^{2+}), magnesium (Mg^{2+}), potassium (K^{+}), sodium (Na^{+}) and, in strongly acid soils, aluminium (Al^{3+}). Exchangeable Al only becomes significant at pH levels less than 5.5 in water or about 4.7 in $CaCl_2$.The cations manganese (Mn^{2+}), iron (Fe^{2+}), copper (Cu^{2+}) and zinc (Zn^{2+}) are usually present in amounts that do not contribute significantly to the cation complement. Therefore, it is common practice to measure the concentrations of only the five most abundant cations. Cation exchange capacity (CEC) is the capacity of the soil to hold and exchange cations. It provides a buffering effect to changes in pH, available nutrients, calcium levels and soil structural changes. As such it is a major controlling agent of stability of soil structure, nutrient availability for plant growth, soil pH, and the soil's reaction to fertilizers and other ameliorants (Brady and Veil, 2002).

Soil properties can be evaluated as statistically due to application of geostatistical methods on soil science recently. Geostatistics, increasingly popular in soil science, are useful in predicting the spatial distribution of spatially dependent soil properties in the field with a number of samples (McBratney and Webster, 1983; Oliver, 1987; Kerry and Oliver, 2004, Aşkın and Kızılkaya, 2006; Aşkın, 2010). Semivariograms and autocorrelograms are typically used to study the spatial structure of soil properties. Soil exchangeable Ca, Mg, K and Na are the dominant exchangeable cations in soils and hence, to examine the spatial variability was expected to better understanding of the related soil chemical parameters for long-term observatory study within a field-scale (Jiang et al, 2008).

The objective of the present study was to assess the spatial variability of soil exchangeable cations in field-scale using geostatistical techniques.

Materials and Methods

Study site

The study area was located in a field on the Perm State Academy Farm in the Perm Region, West Urals, Russia in 2011. Perm is a city and the administrative center of Perm Krai, Russia, located on the banks of the Kama River, in the European part of Russia near the Ural Mountains. From 1940 to 1957 it was named Molotov (Figure 1). This area is characterized gently sloping with a well-drained medium textured soil.

Figure 1. Location map of the study area showing the sampling design. The coordinates are in meters in UTM datum.

Perm has a continental climate with warm summers and long, cold winters. The highest and the lowest the temperature was from -47.1 °C to 37.2 °C. The annual mean temperature was 7.1 °C and the annual mean precipitation was 657 mm based on a 50 year period.

The study site (250x100 m) was chosen for its apparent homogeneity. It was marked with regular rectangle grids (25x25 m each) and included 51sampling points.

Soil analyses

Soil samples were air-dried and ground to pass a 2 mm sieve for related chemical analysis. Soil samples were extracted with 1.0 M ammonium acetate at pH 7.0 and the extracts were then analyzed for exchangeable Ca, Mg, K and Na by flaming emission at the wavelength of 422.7, 285.2, 766.5 and 589.0 nm, respectively, using an atomic spectrophotometer (Page, 1982). Selected soil physicochemical properties were determined by the following methods: organic carbon content by the modified Walkley-Black method (Nelson and Sommers, 1982), particle size distribution by the hydrometer method (Gee and Bauder, 1979), soil pH and electrical conductivity (EC) in 1:1 (w/v) soil-water ratio using pH-meter and EC-meter (Peech, 1965).

Statistical analysis

Classical statistical parameters, i.e., mean, standard deviation, median, minimum, maximum and data normality, were calculated using SPSS 15.0 software. Isotropic semivariances of data were calculated using GS^+ geostatistical software (GS^+, 2006). Semivariance γ(h) is defined in the following equation:

$$\gamma(h) = \frac{1}{2N(h)} \Sigma \left[Z(x_i) - Z(x_i + h) \right]^2$$

where, N(h) is the number of sample pairs at each distance interval h and Z (X_i,) and Z (X_i + h) are the values of variable at any two places separated by distance h. The semivariogram is the plot of the semivariance against the distance. Its shape indicates whether the variable is spatially dependent. Experimental semivariograms were fitted by theoretical models that have well-known parameters nugget (C_0), sill (C_0 + C) and range (A_0) of spatial dependence (Cambardella et al., 1994).

GS^+ has several models that can be fitted to estimate semivariograms, but in this study, we used the isotropic exponential, spherical and Gaussian models:

$$\gamma(h) = Co + C \cdot \left[1{,}5 \cdot \left(\frac{h}{Ao} \right) - 0{,}5 \cdot \left(\frac{h}{Ao} \right)^3 \right] \qquad h \le Ao \qquad \text{Spherical model}$$

$$\gamma(h) = Co + C \qquad h > Ao$$

$$\gamma(h) = Co + C\left[1 - \exp\left(\frac{-h^2}{Ao^2}\right)\right]$$ Gaussian model

$$\gamma(h) = Co + C\left[1 - \exp\left(\frac{-h}{Ao}\right)\right]$$ Exponential model

Where; Co is the nugget variance ≥0, C is the structural variance ≥Co, (Co+C) is the sill variance, and Ao is the range of spatial correlation (GS^+, 2006).
In this study, point kriging was used before constructing of contour maps to provide enough estimated data. The contour maps of exchangeable Ca, Mg, K and Na were constructed using ArcGis software.

Results and Discussion

Soil properties and exchangeable cations

The soils of study are has 33.6% sand, 39.7 silt and 26.7% clay fraction and soil textural class was named as loamy. Also descriptive statistics of soil properties are given in Table 1.

Table 1. Summary statistics on the soil properties and exchangeable cations (*n = 51*)

Soil Properties	Mean	S_e	Min.	Max.	S_d	Skw	Kur
pH (1:1 soil: water suspension)	7.07	0.028	6.67	7.67	0.203	0.785	1.177
Electrical conductivity (*EC*), dS m^{-1}	0.15	0.007	0.05	0.29	0.053	0.797	-0.021
Organic carbon content (*OCC*), %	1.15	0.060	0.45	2.75	0.430	1.560	3.420
Exchangeable sodium (Ex-Na), cmol(+) kg^{-1}	0.22	0.006	0.15	0.35	0.041	0.839	1.227
Exchangeable potassium (Ex-K), cmol(+) kg^{-1}	1.08	0.083	0.48	4.32	0.591	3.526	17.695
Exchangeable calcium (Ex-Ca), cmol(+) kg^{-1}	10.29	0.251	7.00	14.70	1.790	0.430	-0.524
Exchangeable magnesium (Ex-Mg), cmol(+) kg^{-1}	1.37	0.051	0.68	2.24	0.361	0.565	0.132

S_d, standard deviation; S_e, standard error; Skw, skewness; Kur, kurtosis

The soils were mostly medium in texture, neutral in soil reaction, medium in organic matter content (average of 1,98%) and low in electrical conductivity (<0.98 dS m^{-1}) (Soil Survey Staff, 1993).

Spatial variability of exchangeable cations

Distances between Ex-Na, Ex-K, Ex-Ca and Ex-Mg pairs and semivariance values were calculated using the GS^+ package program. The exponential, spherical and Gaussian models with the smallest reduced sums of squares (RSS) values and the biggest R^2 values were selected for evaluating spatial variability of these exchangeable cations in the study area by the GS^+ package program (Table 2).

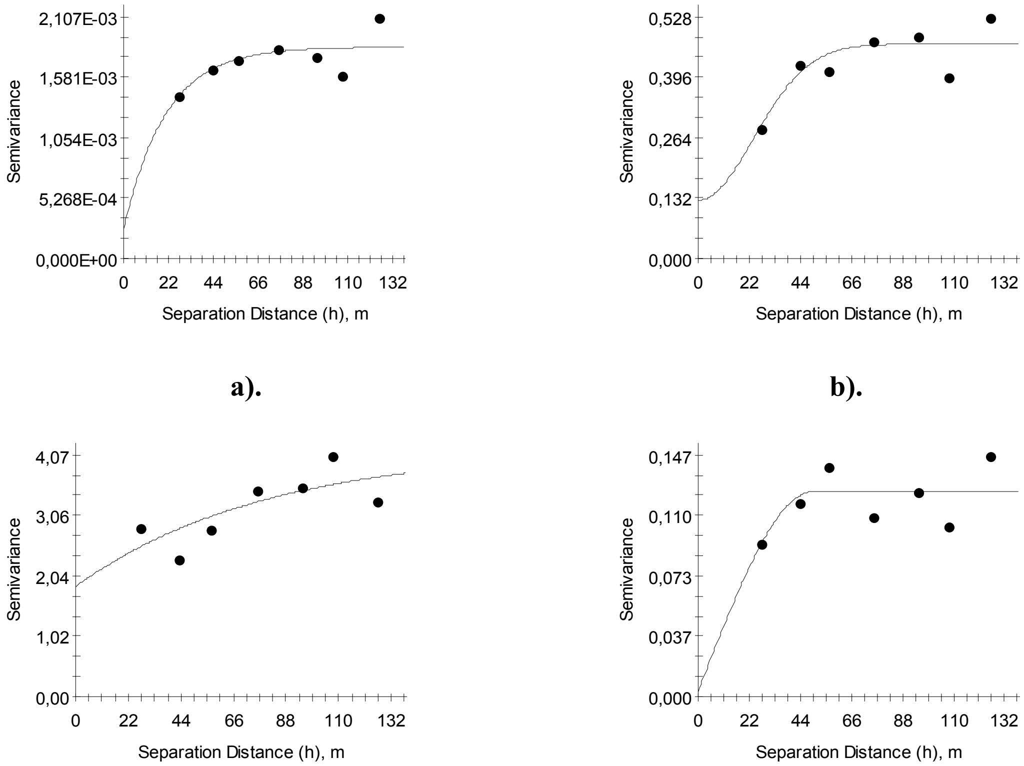
Table 2. Isotropic models fitted to variograms of exchangeable cations

Exchangeable cations	Nugget Co	Sill Co+C	Range (Ao), m	C/Co+C %	Co/Co+C %	R^2	Model	SD
Ex-Na	0.000262	0.001844	62.4	85.8	14.2	0.522	Exp	S
Ex-K	0.127	0.468	58.2	72.9	27.1	0.715	Gaus	M
Ex-Ca	1.857	4.171	237.6	55.5	44.5	0.847	Exp	M
Ex-Mg	0.0033	0.1246	49.7	97.4	2.6	0.995	Sph	S

Exp, exponential; Gaus, Gaussian; Sph, spherical; SD, spatial dependence; M, moderate; S, strong

The nugget effect, representing the undetectable experimental error and field variation within the minimum sampling space, was quite large relative to the sill, which represents total spatial variation. The ratio of nugget variance to sill expressed in percentages can be regarded as a criterion for classifying the spatial dependence of soil properties. If this ratio is less than 25%, then the variable has strong spatial dependence; if the ratio is between 25 and 75%, the variable has moderate spatial dependence; otherwise, the variable has weak spatial dependence (Chien et al., 1997). Jiang et al (2008) interpreted strong and moderate spatial variability as interactions among field-scale variability of soil exchangeable cations physical, chemical, and biological soil components.

The influence zones for Ex-Na, Ex-K, Ex-Ca and Ex-Mg were 62.4 m, 58.2 m, 237.6 m and 49.7m, respectively. The highest nugget effect occurred for Ex-Ca and the lowest for Ex-Mg. The isotropic models showed the best fitting value for the computed semivariance values for exchangeable cations. The isotropic exponential model for Ex-Na and Ex-Ca; the isotropic spherical model for Ex-Mg and the isotropic Gaussian model showed the best fitting value for the computed semivariance values for Ex-Na. The model parameters and the experimental variograms for Ex-Na, Ex-K, Ex-Ca and Ex-Mg are illustrated in Figure 2a,b,c, and 2d, respectively.



c). d).

Figure 2. Izotropic semivarograms for a). Na, b)K, c)Ca and d)Mg

Ex-Na, Ex-K, Ex-Ca and Ex-Mg were point-kriged based on the isotropic models by 7875 points using the ten nearest neighboring points. The descriptive statistics are presented in Table 4 for observed and point-kriged Ex-Na, Ex-K, Ex-Ca and Ex-Mg values.

Table 3. Descriptive statistics on the observed and kriged values of exchangeable cations

Descriptive statistics	Ex-Na cmol(+) kg^{-1}		Ex-K cmol(+) kg^{-1}		Ex-Ca cmol(+) kg^{-1}		Ex-Mg cmol(+) kg^{-1}	
	Obs.*	Predict.**	Obs.	Predict.	Obs.	Predict.	Obs.	Predict.
Number of samples (n)	51	7875	51	7875	51	7875	51	7875
Minimum	0.15	0.16	0.48	0.51	7.00	7.54	0.68	0.72
Maximum	0.35	0.34	4.32	3.85	14.70	13.85	2.24	2.18
Mean	0.22	0.22	1.08	1.07	10.29	10.28	1.37	1.39
Standard deviation	0.041	0.0005	0.591	0.154	0.361	0.061	0.361	0.076
Prediction Errors								
Mean		0.00085		0.01987		0.04989		0.00575
RMS		0.04093		0.5816		1.773		0.3331
Mean Standardized		0.01508		0.02502		0.02029		0.0124
RMS Standardized		0.973		1.095		0.9664		1.083

*Obs., observed; **Predict., predicted; RMS, Root-Mean-Square

As seen from Table 3, the mean reduced error was near zero and the squared differences between the predicted and the original values, the variance of the reduced error, was lowest for the fitted models. This means that the kriging estimates are accurate, and the spatial relationships derived from the studied part of the research site may be applicable to other areas with similar characteristics in this area (Trangmar et al., 1985; Öztaş, 1996;Ardahanlıoğlu et al., 2003; Başkan 2004; Aşkın, 2010; Aşkın et al., 2011).

The range of point-kriged Ex-Na values (0.16-0.34 cmol(+) kg^{-1} with a mean of 0.22 cmol(+) kg^{-1}) was somewhat narrower than the range of the measured Ex-Na (0.15–0.35 cmol(+) kg^{-1} with a mean of 0.22 cmol(+) kg^{-1}). The standard deviation of the kriged Ex-Na values was lower than on the measured selected model. Figure 3a shows a point-kriged map of Ex-Na illustrated using the same 7875 points used to krige Ex-Na.

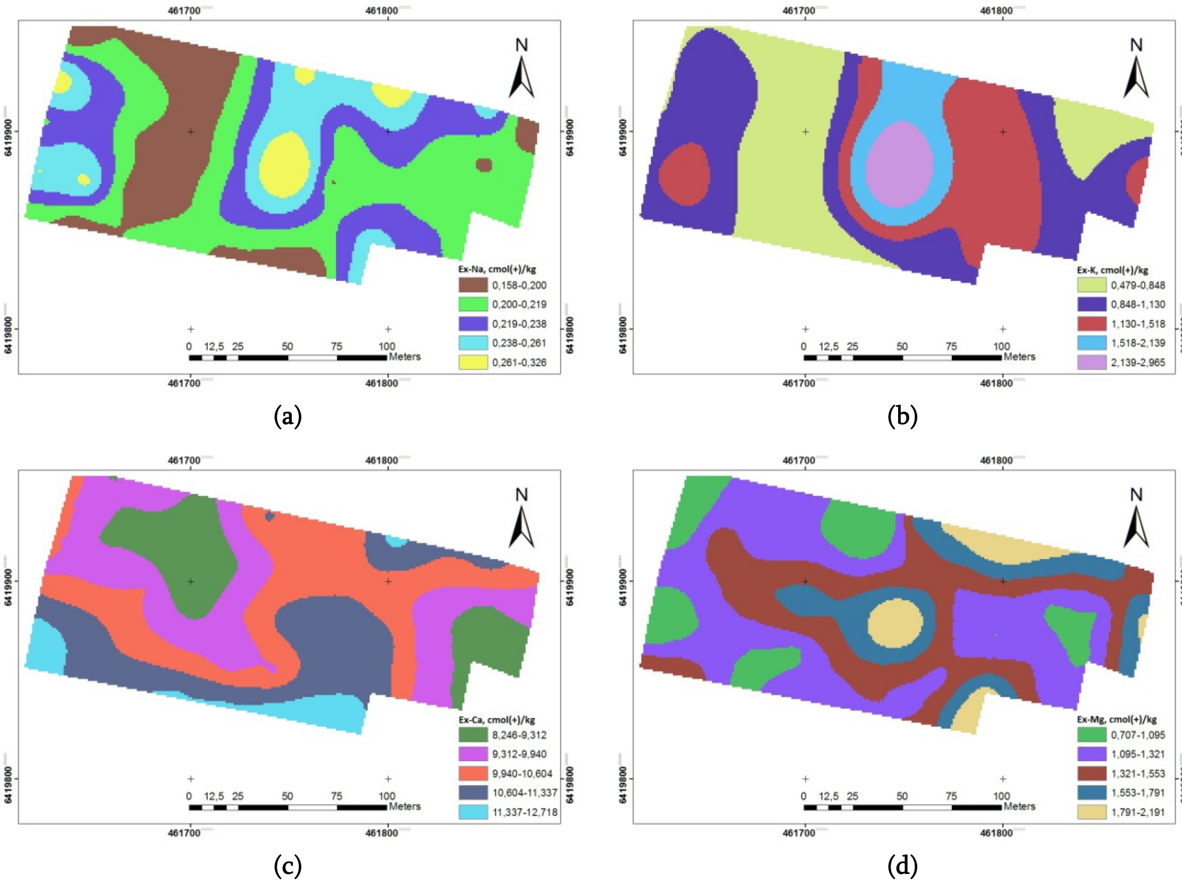


Figure 3. Point-kriged maps for a) Ex-Na; b) Ex-K; c) Ex-Ca; and d) Ex-Mg

The range of point-kriged Ex-K values (0.51-3.85 cmol(+) kg^{-1} with a mean of 1.07 cmol(+) kg^{-1}) was somewhat narrower than the range of the measured Ex-K (0.48–4.32 cmol(+) kg^{-1} with a mean of 1.08 cmol(+) kg^{-1}). The standard deviation of the kriged Ex-K values was lower than on the measured selected model. Figure 3b shows a point-kriged map of Ex-K illustrated using the same 7875 points used to krige Ex-K. The point-kriged Ex-Ca values ranging from 7.54 to 13.85 cmol(+) kg^{-1} with a mean of 10.28 cmol(+) kg^{-1} that was somewhat narrower than the range of the measured Ex-Ca (7.00–14.70 cmol(+) kg^{-1} with a mean of 10.29 cmol(+) kg^{-1}). The standard deviation of the kriged Ex-Ca values was lower than on the measured selected model. Figure 3c shows a point-kriged map of Ex-Ca illustrated using the same 7875 points used to krige Ex-Ca. The point-kriged Ex-Mg values ranging from 0.72 to 2.18 cmol(+) kg^{-1} with a mean of 1.39 cmol(+) kg^{-1} that was somewhat narrower than the range of the measured Ex-Ca (0.68–2.24 cmol(+) kg^{-1} with a mean of 1.37 cmol(+) kg^{-1}). The standard deviation of the kriged Ex-Mg values was lower than on the measured selected model. Figure 3d shows a point-kriged map of Ex-Mg illustrated using the same 7875 points used to krige Ex-Mg.

The range of spatial dependence ranged from 49.7 to 237.6 m, indicating that the grid scale was adequate for assessing of the spatial variability of the exchangeable cations. In this area or a similar field, in soil productivity and fertility research studies to be done about sampling interval can be chosen. Exponential isotropic models were the best semivariogram models for Ex-Na and Ex-Ca, Gaussian model was the best model for Ex-K and also spherical model was the best for Ex-Mg. The information obtained from geostatistical techniques can be used to gain a better understanding of the spatial distribution of soil exchangeable cations in field topsoil. This approach enabled maps to be drawn of soil exchangeable cations in the field-scale. The results suggested that the use of kriging should decrease the required sampling density in the field-scale. Spatial analysis of soil exchangeable cations could be useful for assessing soil fertility and soil quality status, as well as developing appropriate sampling strategies.

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Effect of some pulverization implements and tractor speeds on some machinery unit performance and some soil physical properties *

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Abstract

A field experiment was conducted at Abu-Ghraib during 2007-2008 to evaluate the effect of pulverization implements and tractor speeds on some machinery unit performance, soil mean weight diameter and soil saturated hydraulic conductivity.
In this study, two tractor's speeds included 2.414 and 3.973 km/h, represented the main plots and three pulverization implements included; rotary, disk harrow and spring cultivator, represented sub plots , were used . Tow tractors included; MF-650 and New Holland with pulverization implements, as machinery unit for plowing silt clay loam soil at depth of 17 cm. Slippage percentage, drawbar power, mean weight diameter, and saturated hydraulic conductivity were studied.The experiment was conducted by using Split Plot Design under randomized complete block design (RCBD) with three replications.
The results were as followed:
The rotary was the superior to the other pulverization implements in getting less slippage percentage and drawbar power 1.760 and 45.5 kg.f respectively, whereas spring cultivator effected significantly in getting the highest mean weight diameter 0.905 mm and soil saturated hydraulic conductivity 5.878 cm /h.
Speed of pulverization did not effect on slippage percentage, , drawbar power, mean weight diameter or saturated hydraulic conductivity
The interaction between the spring cultivator and the speed 2.414 km/h was superior in getting higher mean weight diameter 0.918 mm and saturated hydraulic conductivity 6.002 cm/h. While the interaction between the rotary and the speed 3.793 km/h was superior in getting less slippage percentage 1.249 % and drawbar power 37.4 kg.f.
* The research is part of the second researcher thesis

Keywords: Pulverization Implements, MWD, Drawbar Power, Slippage Percentage

Introduction

Plowing operation is considered to be of the primary operations to prepare the soil to be a better growing environment for the seeds. This process increases the soil surface area which exposed to sunlight, by making small soil masses in which facilitates motion of air and water, resulting in a better soil physical properties(Alkazaz,1992 ; and Abass,2004).
Harrowing equipment have been developed and differed into many types and shapes for instance disk harrows, rotary harrows and spike harrows. These equipment have a differ impact from each other. For example spike and net harrows when being used are characterized with a special technical design property, makes them both different from each other in their effect on the soil and consequently the plant(Albanna,1990; and Alfahdawi,2001).
Many considers that the disk harrow is the most utilized equipment in this field of soil processing, for what it has of ability to harrow and pulverize the soil, prepare the seedbed, giving the soil the best technical and physical indications to be a better growing environment,(Mayfield and Robert,1981; and Aday,et al,2001).
Equipment Caliberation, selecting angles, are of the main factors to use this equipment perfectly, harrows can be used after harvesting directly to eliminate residue like cotton maize and sunflower , that’s due to the fact that inversion plowing equipment slips over the plant . In order to get the best soil technical indictors, calibration for the machine must be done, checking the unit through experiments excluded on the field to control and study the factors related to the best soil quality. Rotary tiller is different from chisel and mold board plow in pulverization and disturb the soil, (Alkafaf ,et al,1991;and Mecreery,1959).

Whereby the rotary tiller prepare the seedbed completely, because it does the primary and secondary tillage in one traffic. The research is aimed to determine the appropriate pulverization implement with its speed for having:
1-best performance indicators for the machinery unit
2-best soil physical properties.

Materials and methods

The experiment was conducted in 2007- 2008 in a field belongs to the college of agriculture/ university of Baghdad. Area of the field was 5000m², of a silt clay loam soil (Table 1). Aim of the research was to study the effect of harrowing implement along with the tractor's speed on some performance of machinery unit and some soil physical characteristics. The field wasn't planted in the season before. Weeds were all eliminated. Surface of the soil was leveled using leveling implement.. Soil moisture was 16-17%.
Two tractor's speeds included 2.414 and 3.973 km/h, represented the main plots and three pulverization implements included; rotary, disk harrow and spring cultivator, represented sub plots , were used In this study. Tow tractors included; MF-650 and New Holland with pulverization implements, as machinery unit for plowing silt clay loam soil at depth of 17 cm.
Slippage percentage, drawbar power, mean weight diameter, and saturated hydraulic conductivity were studied.
The experiment was conducted by using Split Plot Design under randomized complete block design (RCBD) with three replications.
(El- Sahooki,et al., 1990).

Table (1) Some soil chemical and physical properties

Character		Percentage
Sand	g. kg^{-1}	98
Silt		582
Clay		320
Soil bulk density	µg.m^{-3}	1.48
Soil particle density	µg.m^{-3}	2.65
Porosity	%	44
Electric conductivity	dS.m^{-1}	3.78
pH		7.76
Weighed moisture content	%	17-16
Mean weighed diameter mm		0.358
Soil hydraulic conductivity	cm/hr	4.813

Calculation of studied indicators:

1-Percentage of slippage %

Slippage percentage was calculated according to the following equation
Suggested by Russell (1980):-

$$Sp = ((vt-vp)/(vt))*100 \ldots\ldots\ldots \%$$

Whereby,

Sp= slippage percentage
Vt= theoretical speed without load (km/h)
Vp=practical speed without load (km/h)

The theoretical speed was calculated by the division of distance on the theoretical time, the following equation was applied:-

$Vt = D / Tt$

Whereby:-

D= the length of the plowed line (m)

Tt=theoretical time without load (sec)

Practical speed (km/h) was obtained from using the following equation

$Vp = (D/Tp)*3.6$

Whereby:

D=distance of the plowed line (m)

Tp=practical time with load (sec)

2-Drawbar force

1) Two tractors were used. First one was drawbaring the second in which the plow was attached to. Both of the tractors were traveling together along with the diameter between them. The plow was at the back in a state in which was almost touching the soil surface in order to measure moving resistance of the machinery unit.

2) Both of the tractors were traveling in the field together along with the diameter of the type of Dillon between them. the plow was in a state of plowing feasibility in different speeds, to ensure the total drawbaring force (pushing force) puf

Plow drawbar force calculation was done in different low speeds according to following equation that was suggested by Al tahan (1991).

Ft = fpu-prm……….Kg.f

Whereby:

Ft: plow drawbar force (kg.f)

Fpu: total drawbar force (push force) (kg.f)

Frm : force of movement resistance (kg.f)

3-Mean weight diameter:

Mean weighed diameter was calculated by using the suggested method by (youker and guiness,1956)

$$MWD = \sum_{i=1}^{n} \overline{xi}.wi / \sum_{i=1}^{n} Wi$$

Whereby:

Mwd =mean weighed diameter

Wi=diameter collection mass

Xi =mean aggregation diameter for each scope

Wi= soil samples mass

N= number of scopes diameter

I= number of scopes

4-Soil hydraulic conductivity:

Constant water head method was used for measuring soil saturated hydraulic conductvity according to klute (1988) equation as follows :

$K = Q/AT*L/\Delta h$

Whereby:

K=gratified water conductivity (cm/h)

Q=water quantity cm^3

A= section area (cm^3)

T=time (h)

Δh=water pressure difference

Results and discussions

Slippage percentage;

Table (2) shows harrowing equipment's influence on slippage percentage. As the process of harrowing using rotivator plow gave less percentage of slippage reached 1.760%. While the spring cultivator gave percentage of 3.313%. That's due to the share of the rotivater, rises up from the soil surface during harrowing to achieve less depth in the soil. This result concurrence with the results of Al fahdawi (2001) and abass (2004) also in comparison with the results of the spring cultivator.
As for the interaction between harrowing and speed, its obvious from the table the privilege of the rotivator and with second speed on spring cultivator, also the privilege of disc harrow and second speeding recording less percentage of slippage reached 1.249%while the spring and disc harrow were 3.472, 4.238% respectively.
That might due to the fact that the rotivater during the process of harrowing rises its shares up the ground resulting in less penetrating into the soil, this result concurrences with Al bassrawie (1997)

Table (2) The effect of pulverization implements and tractor speeds on slippage percentage

Pulverization implement	Practical speed km/h		Pulverization mean
	2.414	3.973	
Disc harrows	2.236	4.238	3.237
Rotivator	2.226	1.249	1.760
Spring cultivator	3.154	3.472	3.313
LSD= 0.05	0.8977		0.2561
Speed mean	2.538	2.986	
LSD=0.05	NS		

Drawbar force (kg.f)
Table (3) illustrates the effect of harrowing equipment in determining drawbar force. As the feasibility of harrowing using rotivator plow obtained less average of drawbar force reached 45.5 kg.f while the spring cultivator obtained 436.5 kg. that might be due to the fact that the shares of the rotivator is being circulating in a direction matches the direction of the tractor resulting in less penetration to the soil, less drawbar force in addition interlocution between the harrowing feasibility and speed has a significant effect on drawbar force whereby the rotivator with second speed exceeded the spring cultivator in having less average of drawbar force reached 37.4 kg while the spring cultivator with the first speed reached 484.1.

Table (3) The effect of pulverization implements and tractor speeds on drawbar force (kg.f)

Pulverization equipment	Practical speed km/h		Pulverization mean
	2.414	3.973	
Disc harrows	376.7	422.4	399.5
Rotivator	53.5	37.4	45.5
Spring cultivator	484.1	388.9	436.5
LSD= 0.05	21.48		18.6
Speed mean	304.8	282.9	
LSD=0.05	NS		

Mean weight diameter
Table (4) illustrates effect of harrowing equipment in the effect of weighed diameter, as treatment of pulverization using spring cultivator
Gave high mean weight diameter reached 0.905 millimeter. While harrowing treatment using the rotivator plow gave less mean weight diameter reached 0.522.
Changing the speed had a specific effect on mean weight diameter. Whereby speed of 2.414 km/h reached higher mean weight diameter of 0.68306melimeter, the second speed of 3.973km/h gave less mean weight diameter reached 0.655melimeter. Those results are in concurrence with the results of Aday,et al, (2001), and that might due to the fact that increasing the speed must lead to

the breakage of soil aggregation. Consequently that’s why speeds increasing lead to less mean weight diameter.
Binary interlocution between speed and harrowing has a specific effect on the characteristic of mean weight diameter. Whereby harrowing treatment, using spring cultivator and the first speed gave higher mean weight diameter reached 0.918melimeter, while harrowing treatment using the rotivator plow and the second speed gave less mean weight diameter reached 0.513melimeter

Table (4) The effect of pulverization implements and tractor speeds on mean weight diameter, ml

Soil saturated hydraulic conductivity

Pulverization equipment	Practical speed km/h		Pulverization mean
	2.414	3.973	
Disc harrows	0.630	0.547	0.580
Rotivator	0.513	0.531	0.522
Spring cultivator	0.918	0.907	0.905
LSD= 0.05	0.004		
Speed mean	0.683	0.655	
LSD=0.05	NS		

Table (5) illustrates effect of harrowing equipment on soil saturated hydraulic conductivity, as treatment of pulverization using spring cultivator gave high saturated hydraulic conductivity reached 5.878 cm/h. While harrowing treatment using the rotivator plow gave less saturated hydraulic conductivity reached 5.118 cm/h. As for changing the speed it had not effect on saturated hydraulic conductivity.
Binary interaction between speed and harrowing has a specific effect on the characteristic of saturated hydraulic conductivity. Whereby harrowing treatment, using spring cultivator and the first speed gave higher saturated hydraulic conductivity reached 6.002 cm/h, while harrowing treatment using the rotivator plow and the second speed gave less saturated hydraulic conductivity reached 5.042 cm/h.

Table 5. The effect of pulverization implements and tractor speeds on saturated hydraulic conductivity, cm/h

Pulverization equipment	Practical speed km/h		Pulverization mean
	2.414	3.973	
Disc harrows	5.761	5.471	5.616
Rotivator	5.194	5.042	5.118
Spring cultivator	6.002	5.755	5.878
LSD= 0.05	0.028		0.023
Speed mean	5.652	5.423	
LSD=0.05	NS		

Conclusions and recommendations
The Previous results indicates to the following
1) _Using the spring cultivator exceeded the other harrows significantly in getting mean weight diameter and soil saturated hydraulic conductivity
Rotivator superiority led to get the less percentage of slippage and drawbar force too
2_superiority of spring cultivator with the speed of 2.414 km/h in getting higher mean weight diameter, And the interaction between the rotivator implement and speed of 3.973km/h in getting less percentage of slippage and drawbar force too
Recommendation:
On the basis of the mention results we recommend the following:
1) _ using the spring cultivator with the second speed because it had given the best soil characteristics and tractor machine performance too
2) _ execution of the study using developed and modern measures equipment like drawbar force equipment to get results of high quantity

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Using EC-Based Maps to Manage Soil and Prevent Soil Degradation

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Abstract
Soil conservation and optimal productivity need to more knowledge and awareness from all interfering agents which are involved in using of soil to appropriate plant nutrition, yield increasing and agricultural products quality. At present, excess fertilizer use has been led to increasing soil salinity irrespective to soil solution materials. In order to reduce this effect and fertilizers sustainable management, It is necessary to provide informative maps that show fertilizer distribution pattern. For this purpose, a field sampling was carried out and 52 points were coordinated by GPS. Related measurements of Soil EC were done and the values were interpolated by Inverse Distance Weighted (IDW) and Spline (SP) methods by using digital elevation model and raster spatial analysis procedures. The thresholds of EC to 50% of yield and EC response threshold were used to classify the filed as appropriate and inappropriate zones. This was done for 54 different plants (including cereals, industrial crops, vegetables, trees and shrubs) and appropriate zones for each plant was determined. This can be useful to determine reasonable cropping and rotation patterns to manage EC as a deteriorate factor on soil. More EC can impose soils to degradation during the time and appropriate cropping patterns, especially selection of plants could be useful to alleviate soil erosion and degradation. These reclassified maps could be useful to clarify the ways of optimized use of soil in respect to salinity challenge.
Keywords: Soil salinity, Soil degradation, Sustainable soil management, Soil classification, GIS.

Introduction
Agriculture is being changed by three fundamental forces: the expanding capacity of personal computers, molecular biology revolution, and developments in information technology like geographical information systems (GIS). Through precision farming, all three technologies can be packaged and delivered to producers. The combined impact is likely to lead to the greatest intellectual transition that has ever occurred in agriculture (Clay, 2011). Soil salinization is an agricultural and environmental concern in many arid and semiarid regions of the world. In Iran, salinity affects large areas because of the inherently clayey and saline nature of the soils, intensive irrigation that results in rising water tables, high evapotranspiration, and inadequate drainage. Excessive soil salinity adversely impacts crop production, soil and water quality, and eventually results in soil erosion and land degradation. In these areas, characterizing the spatial and temporal changes in soil salinity is essential to sustain land quality, optimize crop and water management practices, and recommend adequate soil reclamation. Soil salinity as one of the most important reducing factors affecting crops yield and development, is a serious challenge in Iran which 91% arid and semi-arids in total (Kamkar and Mahdavi Damghani, 2008). Therefore, investigation on the soil status is so important to extend agriculture into marginal lands or conserve other lands from salinization and degradation in the future. Precision farming also has concentrated on site-specific managements on small zones to alleviate the reducing effects of this abiotic factor on crop yields. Soil conservation and optimal productivity need to more knowledge and awareness from all interfering agents which are involved in using of soil to appropriate plant nutrition, yield increasing and agricultural products quality. At present, excess fertilizer use has been led to increasing soil salinity irrespective to soil solution materials. Designing cropping patterns for fields also needs informative plans which can be provided by new techniques such as GIS and interpolation methods. Fertilizers can affect soil salinity by affecting pH. In order to reduce this effects and fertilizers sustainable management, It is necessary to provide informative maps that show fertilizer distribution patterns. On the other hand, no planting in vulnerable lands or better management of soils in high-risk areas can help us to improve soil quality and sustain lands exploiting. This study was aimed to provide EC map of a research farm in GUASNR and assess the suitability of detected zones to different plants with different response thresholds to soil EC.

Material and methods

For this purpose, a field sampling was carried out and 51 points were coordinated by GPS. Field border also was determined by tracking using GPS. Related measurements of soil EC were done and the values were interpolated by IDW (ID) and Spline (SP) methods by using digital elevation model and raster spatial analysis procedures (Wollenhaupt et al., 1997, Franzen, 2011). This experiment was conducted in Research farm of Gorgan University of Agricultural Science and Natural Resources, Iran. The EC response threshold was used to classify the filed as appropriate and inappropriate zones. This was done for 54 different plants (including cereals, industrial crops, vegetables, trees and shrubs) and appropriate zones for each plant were determined. For this purpose, the soil samples were collected based on a geo-referenced point located at the transaction points of a grid cell (100m × 25 m) . At each grid point, random cores were collected. Saturation extraction of soil was provided and EC was determined by EC-meter. Field grids and sampling points are presented in Fig.1.

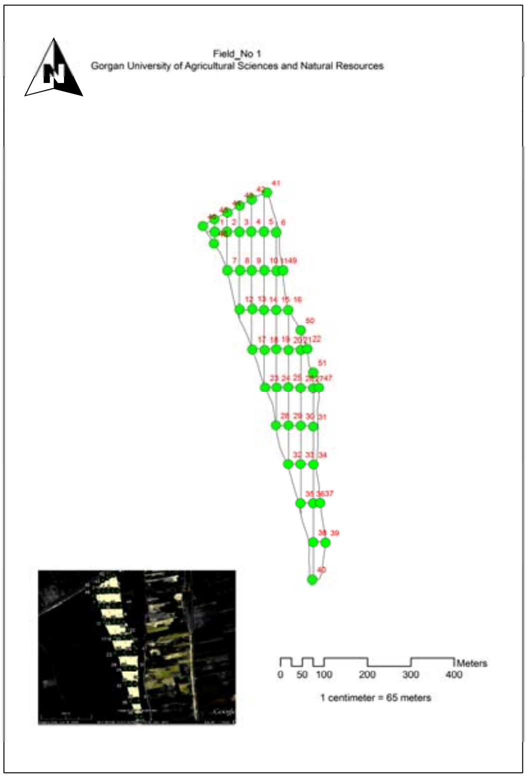

Fig. 1. Field border and sampling points position at the transaction points of grids.

Digital elevation model also was provided by 1/25000 maps and "topo to raster" function with the baselines of contours which were queried from aforementioned maps. Plants which were used in this query have listed in Hall (2001). Raster layers were classified as favorable and unfavorable zones for all 54 studied plants.

Results

Our results indicated that both interpolation methods were appropriate to evaluate EC. Field Ec changed between 0.4-2.48 ds/m and 0.05-2.97 ds/m base on ID and Sp methods, respectively (Fig. 2). Observed against predicted values revealed that SP interpolation method was superior than ID method (RMSE=0.004), but SP method also has reasonable outputs. Therefore, the results of both interpolation methods were provided here. Among 54 plants which were tested here in respect to studied field EC, 49 plants were not faced by EC restriction. 5 plants including (bean, corn, faba bean, turnip and onion) were restricted in many zones which EC value was higher than their threshold. Total field area was 8.502 ha. Our results showed that favorable and unfavorable areas for these plants were not similar (table 1). Therefore, it is advisable that in these zones, cropping of these crops should be avoided, because the yield will reduce by salinity effects, while the soil will be destroyed.

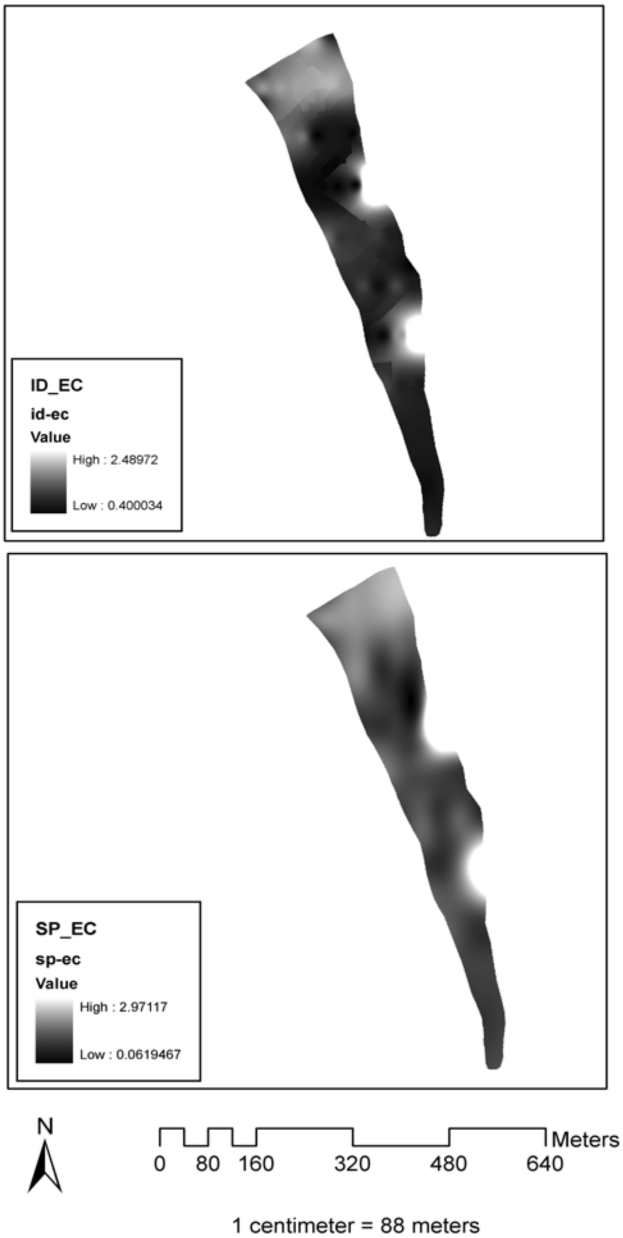

Fig. 2. Ec variability in studied field base on ID and SP interpolationmethods.

Table 1. Plants were used in reclassifying process base on their response threshold and field EC range, total favorable and unfavorable areas and response threshold of plants.

Plants	Favorable (ha)	Unfavorable (ha)	Response threshold (ds/m)
For most of crops	8.429	0.82	2
Faba bean	8.302	0.2	1.6
Bean	8.108	0.4	1
Corn	8.452	0.059	1.7
Onion	8.108	0.40	1.2
Turnip	8.108	0.4	0.9

The map provided for turnip and bean have presented as a sample for both ID and SP interpolation method (Fig. 3 and Fig. 4). The results of other reclassified layers has summarized in Table 1.

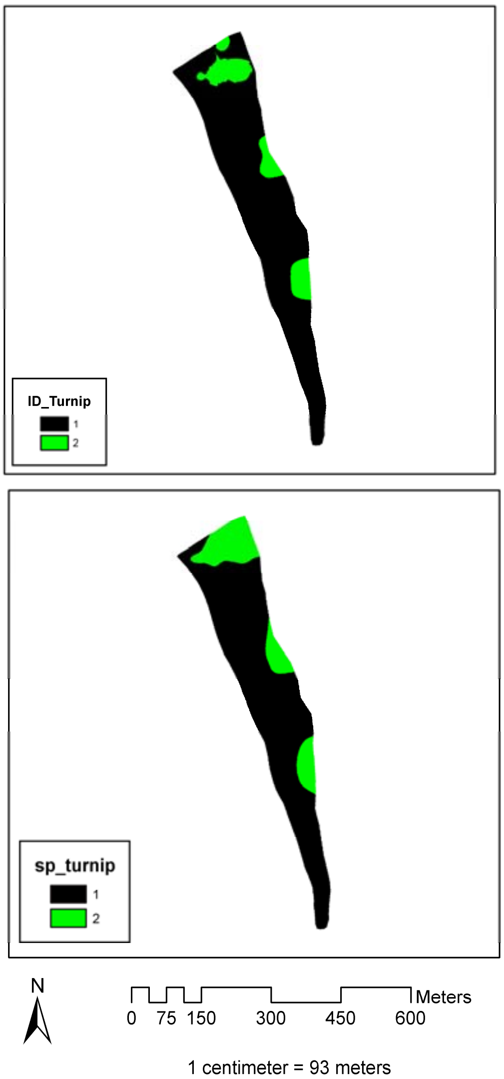

Fig. 3. Favorable (1) and unfavorable (2) areas for turnip cropping in Field #1, Gorgan Univ. Agricultural Science and Natural Resources base on ID and SP methods. For response threshold values refer to table 1.

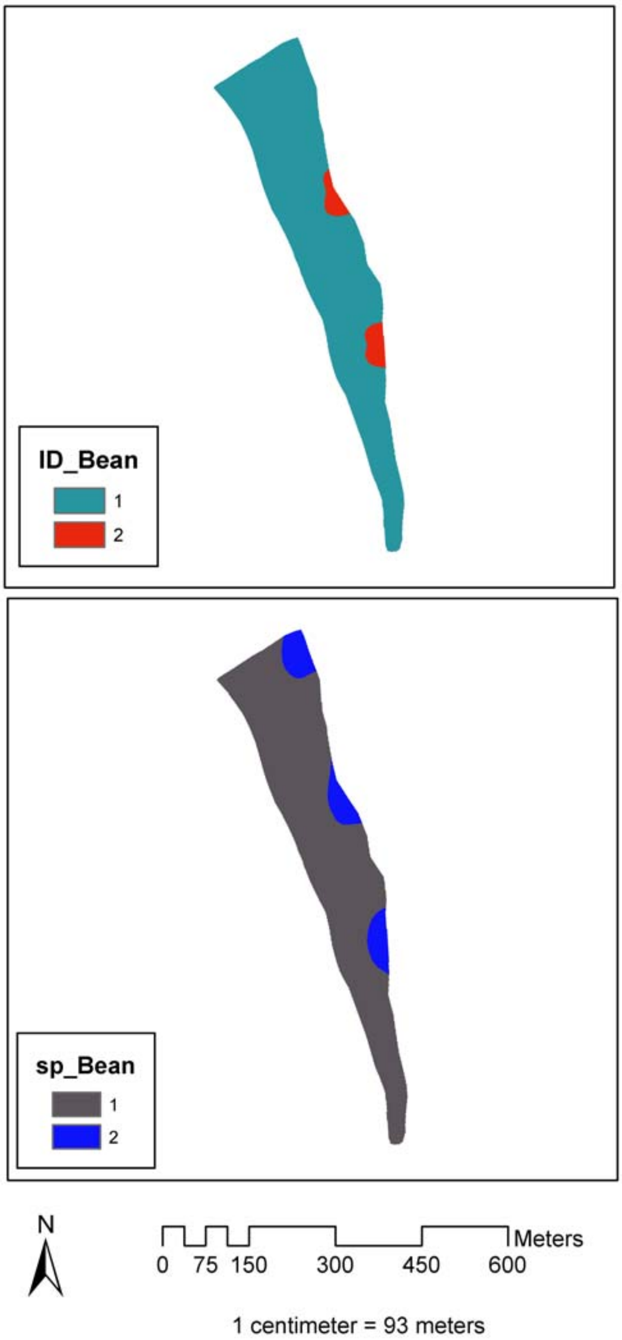

Fig. 4. Favorable (1) and unfavorable (2) areas for bean cropping in Field #1, Gorgan Univ. Agricultural Science and Natural Resources base on ID and SP methods. For response threshold values refer to table 1.

Discussion

Soil degradation is a serious concern which in recent decades has paid much attention on it, because it destroys soil structure and decreases soil quality. Selection of plants that are suitable for a given field is so important to canalize field plans and alleviate negative effects of many other practices such as fertilization or irrigation. Our research revealed that site-specific determination of lands suitability to grow given plants or crops can affect cropping patterns directly and can decrease soil degradation indirectly. Osmotic effects and water uptake interruption are the main effects of soil salinity and consequently yield reduction. Thus, in addition to loss of inputs, and achieving fewer yields, economic reasonability of field will be in question. GIS as a powerful tool can provide applicable maps which can be used as a guideline for growers and policy makers of fields. In this research, different plants showed different plans and this confirmed importance of micro-site detection methods which are the base of precision farming. Thus we can alter crops which are cultivated in these zones or increase irrigation times to dilute salts. Using resistant or tolerant plants also is other option. Spinach family plants are advisable for these areas. These plants are resistant to Na and sodium is an essential element for their growth (sugar beet, spinach, wild spinach). Fertilizers which have chloride, sulfate and nitrate are the main anion compounds which are used in this field. Long-term application of these fertilizers also should be avoided to improve soil situation. It should be noted that these results can be considered in variable rate techniques in respect to irrigation and fertilization aspects.

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Changes in Soil Quality Parameters under Different Land Use Types

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Abstract
Soil quality assessment is essential for sustainable soil management. The objective of this study was to evaluate changes in soil quality parameters in two big soil groups commonly distributed in Erzurum province under different land use types. Disturbed and undisturbed soil samples from 0-15 and 15-30 cm depths of Pellustert and Fluvaquent great groups in cultivated, no till and pasture lands were collected and analyzed for minimum data set for evaluating soil quality. Soil texture, aggregate size distribution and stability, bulk density, penetration resistance, soil organic matter content, soil reaction, $CaCO_3$ content and cation exchange capacity were determined. The results were compared and a quality index value was calculated for each soil type. Soil parameters in cultivated site showed great variabilities than those of no-till and pasture sites. Both soil great groups produced similar results regarding land use type.
Keywords: soil quality, land use type, penetration resistance, soil organic matter, aggregate stability

Effect of Type of Organic Manure and Concentration on the Enzymatic Activities in Some Calcareous Soils from Northern Iraq

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Abstract
Soil enzymes, are very important in the recycling of plant nutrient elements making them mostly available from their organic partner. the effect of four sources of organic manures (alfalfa residue ,cow, chicken and sheep) and their concentrations (0 , 1 and 2%) on the enzymatic activity of three calcareous soils from northern Iraq have been studied in a 45-day incubation experiment at 28C and 90% of the field capacity. During this period ,the activity of urease , asparginase and glutaminase have been measured at a 2-week intervals. Results indicated that the more the concentration of the added manure, The more the activity of the enzymes. Addition of alfalfa residue resulted in more activity, followed by chicken manure, then sheep. The least effect was obtained in soils treated with cow manure.
Keywords: Organic manure, Soil enzyme, Calcareous Soil

Introduction

Enzymes are specific proteins excreted extracellularly (and entracellularly) by soil organisms and plants to break down large organic molecules composing organic manures such as polysaccharides, proteins, lignin and others into smaller molecules to be taken by the decomposing cell, as a source of carbon, nitrogen, and phosphorus(Alexander 1977, Killham 1996). During this process, excess nutrient elements are released to the soil to be used by the growing plants. Among these enzymes are urease, Asparginase , and glutaminase. Urease solubilizes the added urea feltilizer into ammonia and carbon dioxide. Part of the ammonia will be volatilized , while the other part will be used by the growing plant as ammonium or nitrate. Asparginase and glutaminase convert aspargine and glutamine into aspartate and glutamate, respectively making ammonium available to the plat (Zantua and Bremner 1977, Tabatabai 1994).

Since, one of the most important source of soil enzymes are bacteria and fungi, any factor affecting directly their numbers, will affect indirectly on the activity of the enzymes excreted by them. Among these factors are, soil type and it's organic matter contents, pH, moisture, temperatures, and others (Paul and Clark 1989). Since all soil fungi and most of the genera of soil bacteria are chemoheterotrophs, addition of organic manures of animal and plant origins will affect their numbers and so their enzymes excreted (Bergstorm, et al, 1998) .The objective of this investigation is to study the effect of the source and concentration of different organic manures and the soil types on the activity of enzymes represented by urease, asparginase, and glutaminase .

Materials and Methods

Soil types: Soil samples were collected from three different locations in Northern Iraq to represent different properties, as possible, air dried, and ground to pass a 2-mm sieve. Samples of each soil were analyzed physically and chemically, as stated by Page, et al 1982 (table 1).

Table (1) : Some of the chemical and physical properties of the soils studied.

Soil Type	pH	Ec(dsm^{-1})	Organic M%	$CaCO_3$%	Texture
Soil 1	7.13	0.48	0.45	40	Silly clay
Soil 2	7.3	0.84	0.96	28	Silly clay
Soil 3	7.86	1.40	0.34	20	Silly loam

Organic manures: Manures of Chicken, sheep, cow, and alfalfa straw were collected, air dried, and ground to pass a 2-mm sieve. Some of their chemical properties are shown in table 2. (Page et al, 1982)

Table (2) : Some of the chemical properties of the organic manures studied.

Organic source	C%	N%	P%	Ec(dm^{-1})	pH
Chicken	35.3	3.5	1.04	11.6	7.1
Sheep	31.9	2.3	0.68	9.2	7.6
Cow	29.6	2.1	0.91	8.3	8.1
Alfalfa	24.8	4.7	0.39	6.8	6.2

The Experiment: A 500 cc plastic containers, each containing 150g of soil were used as experimental units. The number of containers equal to 72, a result of the combinations of 3 types of soils, (soil 1, 2, and 3) 4 sources of organic manures with 3 concentrations (0, 1%, and 2% w/w). Each replicated 3 times. After the addition of the specific level of each of the organic manures to each of the three soils under study and thoroughly mixed, moisture was added up to 90% of the field capacity of each soil and maintained at this level of moisture by weighing the containers each 4 days. The containers were incubated at 26± 2C° for 42 days during which, the activities of the enzymes urease, asparginase, and glutaminase were measured at (0, 14, 28, and 42) days of incubation according to Tabatabai (1994), and expressed as ,micromole NH_4-N g^{-1} h^{-1}. The plastic containers were opened at 3- days intervals for aeration.

Result and Discussion

Activity of the Enzyme Urease :

Table (3) shows the effect of incubation period on the activity of the enzyme Urease in three soils treated with different concentrations of manures. The data indicate that the activity in the unamended soils at the beginning of the experiment (time 0) differs from one soil to another.

Heigher activity was measured in soil 2, followed by soil 1, while soil 3 was the least (0.52, 0.33, and 0.16 micromole. NH_4-N g^{-1} h^{-1}), respectively. This was in correlation with the organic matter contents of each soil (Table 1). Soil 2 contains the heighest, while, soil 3 the lowest. Microbial biomas is proportional to the organic matter contents of the soil, the more the biomas, the more the enzymatic activity (killham 1996 and Alexander 1977, Dick, et al. 1988). Addition of the lowest level of the organic manures (1%) increased that activity and in some cases it was about double of that obtained in the control. Another increases were also noted when the level of the added manures was 2% . Again, the heighest activity was in soil 2 followed by soil 1, then soil 3. the source of these increases in activity at time 0 are the added manures, since animals and plants excrete urease similer to that excreted by bacteria and fungi (Conn & Stumpt, 1975). Table (3) also shows that incubating the unamended and the manures - amended soils for 14 days gave maximum peak of activity, and any further incubation decreased that sharply. This sharp decreases in activity is probably due to the degradation of the most degradable part of the organic nitrogen composing the organic manures within the first fourteen days of incubation due to the favorable conditions of optimum temperature, moisture, and pH (Alexander 1977, Killham 1996). It was found that, raising the level of the added manures from 1 to 2% increased the activity of the enzyme urease and in some cases, it was double of that measured at the 1% level-maximum activity (5.35 micromole NH_4-N g^{-1} h^{-1}) was measured in soil 1 when amended with 1% chicken manure, followed by alfalfa straw (3.69 micromole NH_4-N g^{-1} h^{1}), followed by sheep manure (2.85 micromole NH_4-N g^{-1} h^{1}), while cow manure was the least (2.69 micromole NH_4-N g^{-1} h^{1}) . Values for soil 1 were (2.88, 3.24, 2.66, and 1.75 micromole NH_4-N g^{-1} h^{1}) respectively. Another increases in the activities were oblained in the soils amended with 2% organic manures (7.29, 7.78, 5.15, and 3.86 micromole NH_4-N g^{-1} h^{1})) for soil 2 and (5.64, 7.19, 4.79, and 2.85 micromole NH_4-N g^{-1} h^{1}) for soil 1. the least activity was measured in soil 3 for both levels of organic manures.

Table (3) : Effect of Incubation Period on Urease Activity (Micromole NH_4-N g^{-1} h^{-1}) in Three Soils Treated with Different Concentrations of Four Types of organic Manures.

A- Chicken Manure

Soil Type	Conc. Of o.m.	Incubation Periods (days)			
		0	14	28	42
Soil 1	0	0.33	0.62	0.39	0.16
	1	0.78	2.88	1.46	0.19
	2	0.94	5.64	2.20	0.08
Soil 2	0	0.52	0.81	0.58	0.32
	1	0.78	5.35	1.82	0.26
	2	1.10	7.29	2.53	0.10
Soil 3	0	0.16	0.52	0.29	0.10
	1	0.38	2.11	1.13	0.16
	2	0.55	5.02	1.72	0.42

B- Sheep Manure

Soil Type	Conc. Of o.m.	Incubation Periods (days)			
		0	14	28	42
Soil 1	0	0.33	0.62	0.39	0.16
	1	0.52	2.66	1.17	0.06
	2	0.64	4.79	1.98	0.58
Soil 2	0	0.52	0.81	0.58	0.32
	1	0.52	2.85	1.59	0.49
	2	0.84	5.15	2.33	0.25
Soil 3	0	0.16	0.52	0.29	0.10
	1	0.26	1.91	0.88	0.23
	2	0.46	4.44	1.42	0.45

C- Cow Manure

Soil Type	Conc. Of o.m.	Incubation Periods (days)			
		0	14	28	42
Soil 1	0	0.33	0.62	0.39	0.16
	1	0.46	2.53	0.97	0.03
	2	0.58	2.85	1.59	0.00
Soil 2	0	0.52	0.81	0.58	0.32
	1	0.45	2.69	1.23	0.26
	2	0.68	3.86	1.72	0.19
Soil 3	0	0.16	0.51	0.29	0.10
	1	0.10	1.75	0.55	0.13
	2	0.23	2.53	1.17	0.23

D- Alfalfa Straw

Soil Type	Conc. Of o.m.	Incubation Periods (days)			
		0	14	28	42
Soil 1	0	0.33	0.62	0.39	0.16
	1	0.84	3.24	1.85	0.19
	2	1.13	7.19	2.30	0.39
Soil 2	0	0.52	0.81	0.58	0.32
	1	0.97	3.69	2.01	0.74
	2	1.33	7.78	2.63	0.10
Soil 3	0	0.16	0.51	0.29	0.10
	1	0.45	2.69	1.46	0.36
	2	0.88	5.61	2.01	0.62

The genera bacteria and fungi which excrete this enzyme are chemoheterotrophs and any increase in the source of carbon and energy will increase their biomass which in turn will increase the activities of the enzymes excreted by them. Many investigators have obtained similar results. Zantua & Bremner 1976 found that urease activity in soil can be increased by the addition of glucose , starch, cellulose, animal manures, and plant materials. They concluded that , although, some of the urease activity produced when treating the soil with organic materials, but , eventually becomes identical to that of the unamended soils. Elder (1993) reported increases in microbial biomass and enzymatic activities after long-term addition of cattle slurry to a grass land. Hojati and nourbakhsh (2006) studied the effect of cow manure and sewage sludge (25 and 100 Tons h^{-1}) on microbial biomas and enzyme activities in a calcareos soil cropped to corn and found that both organic amendments increased the enzymatic activities of glutaminase, alkaline phosphatase, and B-glucosidase compared to the control, and their activities increased with the increases in the rate of application. Comparing the effect of organic manures on the activity of the enzyme urease, it was noted that after fourteen days of incubation, alfalfa straw gave the highest activity followed by chicken manure. Sheep and cow manure was the least, respectively. Table (2) showed that the percentage of the total nitrogen content of alfalfa straw and for chicken manure were 4.7% and 3.5%, respectively, while sheep and cow manures were 2.31 and 2.1% respectively, which reflected on the activity of this enzyme. Again soil 2 gave the heighest activity followed by soil 1, while soil 3 was the lowest, regardless of the type or the concentration of the added manures.

Activity of the enzymes asparginase and glutaminase :

Tables (4 and 5) shows the effect of incubation periods on the activity of the enzyme asparginase (table 4) and Glutaminase (table 5) in three soils treated with different concentrations of manures. The data indicate also that at time 0 the heighest activity was registered in the unamended soil 2 followed by soil 1 , and soil 3 was the least, probably for the same reasen mentioned previously. Again incubating both unamended and manures- amended soils for 14 days gave maximum activity, which decreased after that to reach the activity measured in the control. Quiquampoix et al, 2002 , Yang et al 2006, Laxman and Raman 1999 mentioned that the decrease in the activity may be due to the formation of clay- enzyme or metal – enzyme complexes. Complexation of the enzymes by the clay contents of the soil or metals may affect positively or negatively on the activity of enzymes and some time have no effect. The data of table 4 and 5 indicated that, similar findings to that of urease activity were obtained with regards to the effect of the concentration of the added manures on the activity of both enzymes (asparginase and glutaminase). The heigher the level of the added manure, the higher the activity. Generally, the activity of the enzyme asparginase was more than the activity of the enzyme glutaminase for all the manures tested, except when sheep manure was used, where the opposite was found. Most of the organic N in the organic manures is protein with different sequences of amino acids, among them the amino acids aspargine and glutamine, substrates of the enzyme asparginase and glutaminase, respectively, and probably the concentration of aspargine is more than glutamine (substrate of these enzymes) in all of the manure tested except for sheep manure, which we expect the opposite**.**

Table 4. Effect of Incubation Period on Asparginase Activity (Micromole NH_4-N g^{-1} h^{-1}) in Three Soils Treated with Different Concentrations of Four Types of organic Manures.

A- Chicken Manure

Soil Type	Conc. Of o.m.	Incubation Periods (days)			
		0	14	28	42
Soil 1	0	0.16	0.42	0.16	0.00
	1	0.41	2.64	0.12	0.19
	2	0.84	5.12	1.62	0.10
Soil 2	0	0.42	0.62	0.19	0.10
	1	0.62	3.27	1.26	0.06
	2	0.91	6.29	1.78	0.87
Soil 3	0	0.10	0.29	0.10	0.00
	1	0.26	1.72	0.55	0.10
	2	0.45	3.85	1.46	0.36

B- Sheep Manure

Soil Type	Conc. Of o.m.	Incubation Periods (days)			
		0	14	28	42
Soil 1	0	0.16	0.42	0.16	0.00
	1	0.39	2.09	0.88	0.00
	2	0.38	4.12	1.13	0.19
Soil 2	0	0.42	0.62	0.19	0.10
	1	0.49	2.39	1.03	0.22
	2	0.19	4.51	1.46	0.19
Soil 3	0	0.10	0.29	0.10	0.00
	1	0.10	1.42	0.38	0.00
	2	0.29	3.66	1.12	0.19

C- Cow Manure

Soil Type	Conc. Of o.m.	Incubation Periods (days)			
		0	14	28	42
Soil 1	0	0.16	0.42	0.16	0.00
	1	0.26	1.81	0.68	0.10
	2	0.36	2.43	0.94	0.03
Soil 2	0	0.42	0.62	0.19	0.10
	1	0.42	2.07	0.91	0.19
	2	0.36	3.69	1.17	0.10
Soil 3	0	0.10	0.29	0.10	0.00
	1	0.13	1.13	0.29	0.00
	2	0.19	1.72	0.84	0.10

D- Alfalfa Straw

Soil Type	Conc. Of o.m.	Incubation Periods (days)			
		0	14	28	42
Soil 1	0	0.16	0.42	0.16	0.00
	1	058	3.21	1.13	1.29
	2	0.94	6.71	1.98	0.07
Soil 2	0	0.42	0.62	0.19	0.10
	1	0.78	3.44	1.65	0.38
	2	1.03	6.32	2.14	1.04
Soil 3	0	0.10	0.29	0.10	0.00
	1	0.36	1.91	0.88	0.23
	2	0.62	4.18	1.72	0.24

Table (5) : Effect of Incubation Period on Glutaminasee Activity (Micromole NH_4-N g^{-1} h^{-1}) in Three Soils Treated with Different Concentrations of Four Types of organic Manures.

A- Chicken Manure

Soil Type	Conc. Of o.m.	Incubation Periods (days)			
		0	14	28	42
Soil 1	0	0.19	0.26	0.10	0.00
	1	0.36	2.63	0.91	0.19
	2	0.55	4.76	1.43	0.29
Soil 2	0	0.19	0.52	0.19	0.10
	1	0.49	3.44	0.94	0.25
	2	0.57	5.83	1.46	0.00
Soil 3	0	0.06	0.19	0.13	0.00
	1	0.19	1.13	0.45	0.04
	2	0.36	3.31	1.13	0.29

B- Sheep Manure

Soil Type	Conc. Of o.m.	Incubation Periods (days)			
		0	14	28	42
Soil 1	0	0.19	0.26	0.10	0.00
	1	0.26	1.85	0.65	0.10
	2	0.38	3.74	0.98	0.00
Soil 2	0	0.19	0.52	0.19	0.10
	1	0.23	2.92	0.74	0.17
	2	0.49	4.61	1.17	0.17
Soil 3	0	0.06	0.19	0.13	0.00
	1	0.10	0.91	0.19	0.00
	2	0.19	2.82	0.97	0.10

C- Cow Manure

Soil Type	Conc. Of o.m.	Incubation Periods (days)			
		0	14	28	42
Soil 1	0	0.19	0.26	0.10	0.00
	1	0.13	1.43	0.49	0.10
	2	0.26	2.11	0.62	0.00
Soil 2	0	0.19	0.52	0.19	0.10
	1	0.13	2.56	0.55	0.10
	2	0.36	3.08	1.04	0.19
Soil 3	0	0.06	0.19	0.13	0.00
	1	0.00	0.55	0.19	0.00
	2	0.10	1.62	0.74	0.06

D- Alfalfa Straw

Soil Type	Conc. Of o.m.	Incubation Periods (days)			
		0	14	28	42
Soil 1	0	0.19	0.26	0.10	0.00
	1	0.25	2.82	0.91	0.22
	2	0.74	5.83	1.65	0.17
Soil 2	0	0.19	0.52	0.19	0.10
	1	0.58	4.31	1.17	0.19
	2	0.91	6.81	1.81	0.19
Soil 3	0	0.06	0.19	0.13	0.00
	1	0.36	1.42	0.58	0.10
	2	0.45	3.47	1.46	0.36

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Physicochemical properties of pistachio orchard soils as affected by Magnesium-rich irrigation water in Rafsanjan area

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Abstract
Magnesium is an essential nutrient element for plant growth, but in concentrations higher than Ca, it may cause negative effects on soil properties and plant growth. Rafsanjan valley with an aridic soil moisture regime and high salinity of soil and water resources is located at the southeastern part of central Iran under intensive pistachio orchards. The objectives of the present research were to study the effect of irrigation water on increasing concentration of some nutrients like Mg in the soil, and to compare properties of pistachio orchard soils (4 representative Pedons) with non-cultivated adjacent soils (4 representative Pedons). Results of the study showed that irrigation with a high Mg concentration water (56.3 meql^{-1}) increased Mg concentration and Mg/Ca ratio (from 0.73 to 1.34) and decreased K and Na concentrations and SAR content in soils under study. Besides, irrigation more than leaching requirement decreased K (from 5.56 to 3.7 meql-1) and Na (from 538.7 to 311.9 meql^{-1}) concentrations, EC (from 58.4 to 41.2 dSm^{-1}) and SAR (from 50.05 to 43.9) contents in pistachio orchard soils compared to non-cultivated soils. On the other hand, irrigation without considering leaching requirement has increased EC from 27.8 to 37.9 dSm^{-1}. Irrigation management practices and water quality have significantly affected soil properties.
Keywords: Mg/Ca ratio, Pistachio, Rafsanjan, Soil quality.

Introduction
An extent area of Iran is arid and semi-arid and faces water deficiency. A great portion of water needed for agriculture, industry, and drink purposes is from underground water resources. Rafsanjan area has the most widespread Pistachio orchards in Iran. Deep wells together with irregular and non-sustainable irrigation has lowered water table and increased some elements like Mg in water. Mg concentration increases with salinity and affects soil properties.
Mg levels in soils and irrigation water has increased (Vyshpolsky, et al., 2010). Using Mg-rich irrigation water affects soil fertility and plant growth (Fernández-Cirelli, et al., 2009). Magnesium composed about 1.93 % of earth crust. Mg is absorbed by soil particles in arid and semi-arid soils, that is why toxicity of Mg in these climatic regions is even more important than its deficiency.
There are different ideas about role of Mg on physicochemical soil properties (Norton and Zhang, 2002). The role of Mg on pH is sometimes more than Ca, which increases soil pH up to 8.5. Magnesium reduces soil structure stability directly and increases Na on exchangeable sites, indirectly (Yousaf, et al., 1987; Curtin, et al., 1994; Norton and Zhang, 2002). Type of clay minerals and electrolyte concentration control these negative effects of Mg.
Karimove et al. (2009) reported that higher Mg/Ca in irrigation water replaced Na by Mg on exchangeable sites of soil. Since Pistachio orchards have been highly affected by Mg-rich irrigation water through time, the main objective of the present research was to study physicochemical properties of soils under pistachio orchards compared with non-cultivated adjacent soils in Bayaz area of Rafsanjan.

Materials and methods
Study area
Bayaz area with the elevation of 1410 m asl is located between 55º 16' to 55º 43' longitude and 30º 32' to 30º 52' latitude (Fig. 1). Mean annual temperature and rainfall are 17.7 and 89.4 mm, respectively. Soil moisture/ temperature regimes are aridic/thermic (Banaie, 1998).
There are 63 deep wells and 4 qanats in the area (Geographical Organization of the Ministry of Defense and Armed Forces Logistics. 2003). From geological point of view, the area is part of central Iran unit which is called Anar-Rafsanjan basin. The Anar-Rafsanjan basin is composed of Neogene

sediments. The alluviums are well sorted from coarse fragments in elevations toward salty clay deposits in playa (Geographical Organization of the Ministry of Defense and Armed Forces Logistics. 2003b).

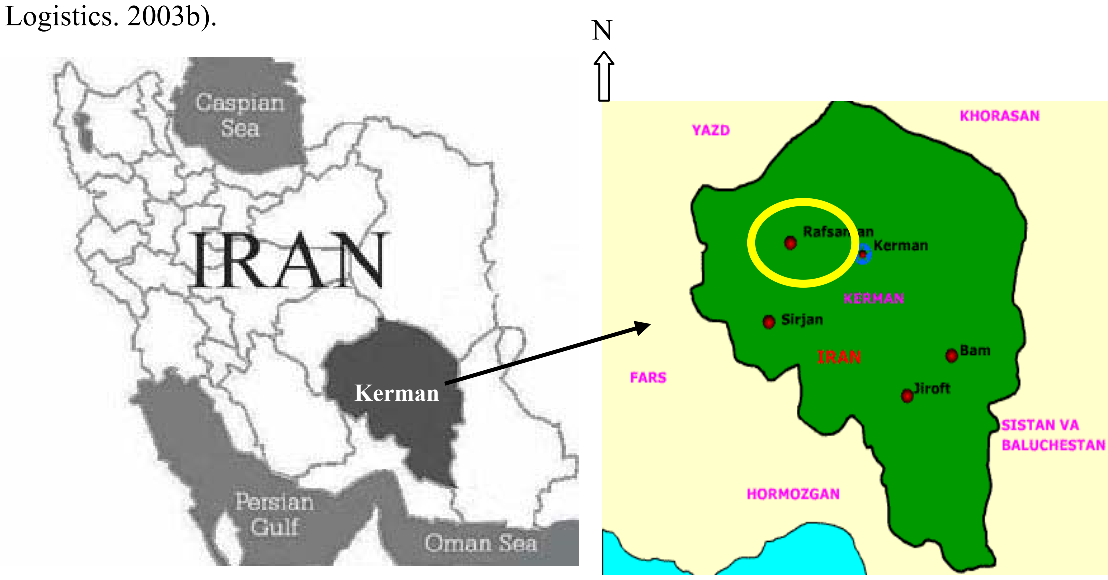

Fig.1. Study area and location of representative pedons

Field work

Chemical analyses of about 500 irrigation water samples were studied and 4 representative water samples with different Mg/Ca ratios were selected. Besides, 4 representative pedons in Pistachio orchards and 4 pedons in non-cultivated adjacent soils were selected, described and sampled (Schoeneberger, et al., 2001).

Lab analyses

Air dried soil samples were meshed and passed through a 2 mm sieve. Particle size distribution (Gee and Bauder, 1986), weight percent of saturated moisture (SP), Calcium and Magnesium in saturated extract (Richards, 1954), Na and K with Jenway Flame Photometer (Richards, 1954), Cl^- by titration with $AgNO_3$ (Richards, 1954), CO_3^{2-} and HCO_3^- with titration method (Richards, 1954) were investigated. Jenway pH and conductivity meters were also used for pH and EC measurement of soil samples. Besides, pH, EC and routine cations and anions were determined in irrigation water samples.

Table 1 - Chemical properties of irrigation water used in pistachio orchards under study

Area	EC	pH	Cations (meq/lit)				Anions (meq/lit)				SAR	Mg/Ca
	(dS/m)		Ca^{2+}	Mg^{2+}	Na^+	K^+	Cl^-	CO_3^{2-}	HCO_3^-	SO_4^{2-}		
Baharestan	15.5	7.1	21.15	15.07	147.5	0.99	135	0	2	47.71	34.7	0.7
Giti Abad	19.4	7.0	38.7	15.5	176.25	0.97	180	0	1.2	50.22	33.89	0.4
Hojat Abad	20.9	6.7	34.6	56.3	186.25	1.13	185	0	4	89.28	27.63	1.6
Mehdi Abad	15.3	7.2	25.9	22.8	132.5	1.06	147.5	0	1.2	33.56	26.87	0.9

Results and discussions

Table 1 shows chemical properties of irrigation water samples studies. Baharestan village has the Mg/Ca ratio of 0.7. Moreover, Mg/Ca ratio in Pistachio orchard (pedon 1) and non-cultivated adjacent soil (pedon 2) are 0.39 and 2.22, respectively (Table 2). It seems that irrigation with good quality water has decreased Mg/Ca ratios from 2.22 to 0.39 in Pistachio orchards under irrigation. The same trend was also observed in pedons 3 and 4.

On the other hand, Mg/Ca ratio in Hojat Abad irrigation water is 1.6. The ratios higher than 1 are called Magnesium-rich water (Qadir and Pauw, 2007). Comparison of pedons 5 and 6 show that the Mg/Ca ratio of orchard soil (pedon 5) is 0.56, which increases to 1.02 due to Magnesium-rich irrigation water used.
Electrical conductivity, Na^+, K^+ and SAR contents in cultivated soils are less than non-cultivated soils except pedon 7, which is attributed to irrigation time intervals. Jalali (2008) has also reported that the use of a saline irrigation water containing high Mg content for a long time increases K leaching in saline and sodic conditions. Moreover, Yong et al., (2005) reported that pedogenesis caused a more Magnesium holding capacity in lower horizons of Anthrosols of China. The same results were also observed in soils under the study of the present research.

Table 2- Mean weight of some chemical properties of representative pedons

Pedon No.	EC (dS/m)	Cations in solution (meq/lit)				SAR	Mg/Ca
		Ca^{2+}	Mg^{2+}	Na^+	K^+		
1	14.3	37.43	14.7	103.6	1.32	20.3	0.39
2	85.7	31	69	1169	16.2	169.8	2.22
3	57.8	29.13	27.07	156.3	1.34	29.06	0.93
4	102.2	127	197	1348	30.6	95.8	1.55
5	37.9	80.12	44.75	315.4	3.09	40.22	0.56
6	27.8	14.97	15.31	300.77	2.83	76.26	1.02
7	41.2	42.5	57.05	311.87	3.68	43.94	1.34
8	58.3	100.9	73.3	538.7	5.56	50.05	0.73

Pedons 1 3, 5, and 7 are located in pistachio orchards of Baharestan, Giti Abad, Hojat Abad, and Mehdi Abad, respectively; pedons 2, 4, 6, and 8 are located in non-cultivated soils of Baharestan, Giti Abad, Hojat Abad, and Mehdi Abad, respectively.

Conclusions

- Magnesium concentration increases with salinity and this in turn increases Mg/Ca ratio.
- Mg/Ca ratio of irrigation water highly affects this ratio in soils under irrigation.
- Parent material, pedogenesis and anthropogenic factors affect Ca and Mg in soils.

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Investigation of Effective Factors on Adoption of "Soil Test" By Potato Growers in Zanjan Province of Iran

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ABSTRACT

The soil test is an important measure of the soil's ability to supply nutrient elements needed for good plant growth also soil testing reveals the current fertility status and provides the information necessary to maintain the optimum fertility conditions for the plants to be grown. Regarding the importance of soil testing to soil health, the main purpose of the study was investigation of effective factors on adoption of "soil test" by potato growers in Zanjan province, Iran. The target population was consisted of potato growers in Zanjan (N= 1500). Stratified Random Sampling was used to select sample population and sample size was included 124 farmers. The main instrument for data collecting was a questionnaire. The questionnaire's validity was confirmed by a panel of faculty members of University of Tehran. Reliability was measured by Cronbachs' alpha and α=0.83 showed the reliability of the questionnaire was acceptable. The results of regression analysis indicated that the adoption of soil test by potato growers in Zanjan province was explained by four variables: attending in extensional courses, respondents' educational level, accessibility to information channels and level of income.

Key Words: Adoption, Soil Test, Effective Factors, Zanjan Province.

Introduction

Fertilizers are the major suppliant of the plant with food elements and are also the most powerful means of maximizing the production of agricultural crops in a hectare. In other words, nutrition elements are critical for increasing the agricultural productions by increasing fertility of the soil. Increasing agricultural production inattentive to food elements of soil without increasing the fertilizers is not possible (Hajimirrahimi et al, 2010).. In another word sustainable soil fertility is key to food and livelihood security of millions of people living in semi-arid regions. (Reddy, 2011)
Although providing the soil chemical and organic materials which mostly done by using different kind of fertilizers in professional agriculture is necessary, in Iran over consumption of chemical fertilizers and imbalance and inappropriate application of them and also ignoring organic fertilizers has became a matter of great concern. Hence Plant nutrition is not used as a scientific tool to increase production in Iran, because over 95% of 2/2 million ton consumed fertilizer is consisted of fertilizers such as urea and ammonium phosphate and other nutrition elements especially micro elements application is very few in fertilization are not observed. The imbalance consumption has led to nutritional deficiency and decrease of soil fertility and also decreasing agricultural produces quality and quantity (Malakoti and Nafisi, 1995).
-Experts believe that sampling and soil test is the only way to restore balance and apply correct fertilization. Paying great attention to soil test results could be important for the further growth of nutrient management in cereal production (Katalin and et al., 2009). Furthermore adoption of some management practices, such as soil testing believed to be an effective way to reduce nonpoint-source pollution from agriculture (Wu and Babckock, 1998)
An agriculture ministry effort has not led to adaption of soil test by farmers. There are unknown different problems that farmers do not adopt soil test. In addition to technical factors, there are

many social issues, which significantly affect the Soil fertility management measures like soli test (Reddy, 2011).

This research has been performed with goal of identification of difficulties and effective factors in adoption of “soil test” by potato growers in Zanjan province.

The study results of Heydari Sareban and Eftekhari (2010) in Ardabil province showed that there is important difference between production cooperative members and non-member farmers due to income and use of regarding of extensional recommendations and their confidence to actions and connection with relation channels and also, there is meaningful and positive relation between independent variables of working-hours of farmer second job, relative advantage and adjustment and test, observable results and soil holding operation complexity and all lands under possess and management and farmer’s work, number of receive debt holding of soil and value, of debt installments and preferring elders to consulting in operation field with value of operation innovation adoption of soil, Also, between know independent variables because of hold operation of soil and numbers of educational terms that a farmer participates in and value of holding the information of radio and educational leaflets with value of operation innovation adoption of soil. The study results of Rostami and et al (2007), indicates that education level and the amount of lands under culture and production various and degree of risk and kind of posses all they are 5 effective factors in acceptance insurance by wheat planters of Kermanshah Hercin City. Yong farmers and lands owners and containing year’s insurance acceptance have tendency toward insurance. Finding research of Arayesh (1998), about farmers raining irrigation technology acceptance behavior in Elam province show that there is not meaningful relation between age and education and lands situation and farmers social characteristics with raining irrigation technology adoption. As there is meaningful relation between agricultural unit size and year of working and economic, cultural and technical factors with adoption of this innovation. Also, studies of yaghobi andchizari (2000), show that behavior adoption of domesticated insurancement in Isfahan province that there is not meaningful relation education independent variables and years of working and age and responded social specifications and hold-domesticates approach to domesticated insurancement and relation to promoter and hold-domesticated company in education actions, but there is meaningful relation between domesticated number independent variables, rate of income, number of debt receive times and amount using of channels and relation resources and domesticated instrument acceptance or adoption. The Research’s results Chizari(2003), shown there is meaningful and negative relation between degree of soil holding technology acceptance or adoption and age variable and years of agriculture, but there is meaningful and positive relation between literacy level, agricultural income, utilized lands extension and agriculture’s approach relative to holding soil and rate of using of debts and variable and governmental credits dependent on soil holding technologies adoption. Other study in Khodabande Town, zanjan province shows that being educated and having more lands are effective on insurance acceptance by cereal planters. Of course factors such as: age, participation in expert’s classes, having unagriculture job and receiving of facilities of agricultural institutes are unable effects on adoption of this innovation. On the other hand study of Dervish and cooperation is explained awareness degree of Agro Forestry innovation and degree of accessing to validities or credits and annual income of family about 50% changes degree adoption, and education and income and access to institutes have meaningful and positive effect on this adoption. The study findings in Ardebil province showed that access to publications and books on irrigation, literacy level, knowledge relation to irrigation and agree of lands under coverage to farmer's behavior is effective in rinsing irrigation (Bagheriand Malekmohammadi, 2005). The studies Hardaker and et al. (2004), did shows that farmer’s decisions in planning is the result of individual experiences, degree of knowledge and ability to face the risks and degree of acquiring information from data recourses. Also membership of farmers in councils and their connection with other farmers have positive affection on their decisions. Also, the studies Barnett and et al (2000), shows that Arkans farmers, miccipi and Louisiana have little trend to innovation adoption of insure men relation to farmers other areas America that it’s reason is expressed innovation increase cost and weakness innovations enforceable designing. The studies Hodges and et al, (1994), shows limitation. Knowledge producers have been caused un-acceptance of holding irrigation innovation. Other study has shown that the meaningful relation exists between subsidies to buying irrigation equipment to farmers with degree of irrigation technology adoption by them

(Dinar and yaron, 1992). The results of study Caroline and et al (2009) in Missouri of America has shown that humanist's motivation was important reason of participation adoption in holding plan environment, and also educational plans have been meaningful effect of increasing of motion and supportive group's management knowledge of environment. The results of another study in Switzerland are shown that perception, communication, and possibilities to participate are the most decisive driving factors influencing the formation of a long-lasting acceptance. Furthermore, acceptance may be based mainly on economic criteria, on usefulness, on ecological or even aesthetic aspects. It could be shown that not all of these motivations lead to a long-lasting acceptance. Ecologically based acceptance seems the most promising because it is founded on general convictions. Economic incentives though important seem to generate only superficial acceptance and do not seem to be as important as is usually assumed (Schenk and et al, 2007). Study of Sattler and et al (2008), shows that despite the general assumption that farmers' decisions are mostly driven by economic rationality, costs were not the most important factor. Other factors, like associated risks, effectiveness, or time and effort necessary to implement a certain measure were equally or even more important depending on the specific situation. Generally, reviewing the research findings results show that individual factors are personality characteristics, and literacy level and farmer's job experiments that have much effect on innovation adoption level. In other word, incentives and materials and spiritual motives and also supports and financial facilities can be determinant on degree innovations adoption.

Materials and Methods

This research involved a descriptive-correlation and a causal comparative survey study. The population of this study consisted of 1500 potato growers in Zanjan province. Using a stratified proportional random sampling technique, 124 potato growers chosen as a sample for the study and were divided into two groups (adopted = 43 and non-adopted = 81) based on their adoption status of Soil Test. Sample size was obtained and supported by the study of Cochran (1977) which offers a formula for determining sample size for a given population. Independent variable of relation characteristics is relative to degree aware and information of innovation "soil test" and kind of impression it's characters and manner potato grower appreciation in extension-educational actions. Dependent variable is innovation level "soil test" by potato growers in Zanjan province. The main instrument for data collecting was a questionnaire. The questionnaire's validity was confirmed by a panel of faculty members of University of Tehran. Reliability was measured by Cronbachs' alpha and α=0.83 showed the reliability of the questionnaire was acceptable. Data were analyzed using Statistical Package for the Social Sciences (SPSS). Descriptive statistics (frequency, percent, mean, standard deviation) were used to describe analyzed data. Independent sample t-test, the man-whitney test and discriminate analysis were employed to analyze the difference among variable.

Result and Discussion

Farmers personal and farming characteristics: The mean age of farmers in the study was 47years (SD=12/8) the majority (68 farmers or 53%) ranged from 55-65 years old. On average, farmers 33/5 years of experience in agriculture (SD= 16/5). The averages of irrigated and arid land amount were 2.54 and 8.65 hectares, respectively. Farmer's educational level average 2.5 years, 76 farmers (63%) had elementary level education. On the other hand, the average distance of farmers land to the nearest agricultural service centers was 6.5 Km.

Table 1: Farmers personal and farming characteristics (n= 124).

variables	Mean	SD	Min	Max
Age (year)	47/02	12/81	24	68
Agricultural experience (year)	33/58	16/55	2	52
Irrigated farm size (Hectare)	2/54	1/48	1	8
Arid farm size (Hectare)	8/65	5/65	1	25
distance between farm and agricultural service centers (Km)	6/5	2/66	1	20
Educational level (year)	2/51	1/98	1	8

Correlation coefficients between selected variables with the rate of adoption of soil test by potato growers

The results of correlation coefficient in Table 2 showed that there was significant and positive relationship between the dependant variable of "the rate of adoption of soil test by potato growers" and independent variables of their: attending in extensional courses ($P<0.01$), respondents' educational level ($P<0.05$) and accessibility to information channels ($P<0.01$). There was also no significant relationship between the respondents' level of income with the rate of adoption of soil test by potato growers at the alpha levels of 0.05 or 0.01.

Table 2- Correlation coefficients between selected variables with the rate of adoption of soil test by potato growers

Selected variables	Correlation coefficients	Significant Level
Attending in extensional courses	0.752	0.000**
Respondents' educational level	0.654	0.031*
Accessibility to information channels	0.369	0.001**
Level of income	0.331	0.441

**: Correlation is significant at the 0.01 level.
*: Correlation is significant at the 0.05 level.

Effective Factors on Adoption of "Soil Test" By Potato Growers

The multivariate regression with stepwise method was used for predicting impacts of independent variables on the dependent variable of adoption of soil test by potato growers. The results of this analysis showed that the variable of "attending in extensional courses" was entered into the equation at the first step. Multiple correlation coefficients (R) were equal to 0.517 resulting in coefficient determination (R2) equal to 0.267: it means that 26.7% of the changes in the variance of the dependent variable are explained by the above mentioned variable.

At the second step, the variable of "respondents' educational level" was entered into the equation. The variable increased R and R2 up to 0.723 and 52.3%, respectively; which describes 25.6% of the changes in the variance of the dependent variable. "Accessibility to information channels" was entered in the equation at the third step. This variable increased R and R2 up to 0.781 and 61%, respectively. Thus, 8.7% of the changes in the variance of the dependent variable were described by the "accessibility to information channels". At the last step, the variable of income level entered in the equation and it increased R2 up to 62.7%. These results are shown in Table 3.

Table 3- Results of regression analysis related to effective factors on adoption of soil test by potato growers

Independent variable	R	R2	R2 Ad.	B	Beta	Sig.
Constant	-	-	-	1.01	-	0.000
Attending in extensional courses	0.517	0.267	0.257	1.78	0.514	0.000
Respondents' educational level	0.723	0.523	0.518	1.12	0.359	0.000
Accessibility to information channels	0.781	0.610	0.606	1.04	0.177	0.001
Level of income	0.792	0.627	0.625	0.56	0.122	0.002

Taking the above results and those in Table 3 into account, the linear equation resulting from regression analysis is as follows:

$$Y = 1.01 + 1.78X1 + 1.12X2 + 1.04X3 + 0.56X4$$

The components of the equation include:

Y = Adoption of soil test by potato growers,

X1= Attending in extensional courses,

X2 = Respondents' educational level,

X3 = Accessibility to information channels,

X4 = Level of income.

According to the results shown in Table 10, the variable “attending in extensional courses” (Beta= 0.514) had the greatest influence on the adoption of soil test by potato growers; the variables “respondents' educational level” (Beta= 0.359), “accessibility to information channels” (Beta= 0.177) and “level of income “(Beta= 0.122) ranked as the second, third and fourth most important factors respectively influencing the dependent variable.

Suggestions

According to importance of attending in extensional courses in adoption of soil test by potato growers, sound educational programs should be planed for making the people informed and encouraged by using various staff development and extension training strategies. Potential educational and training programs might include, among other strategies and methods: conducting workshops and meetings; promoting lecture presentations; supporting publication and distribution of educational materials related to soil test.

According to the results of regression analysis, one of the most important variables affecting adoption of soil test by potato growers was accessibility to information channels. In this regard, providing the information channels particularly new ones including computers and Internet can accelerate the rate of adoption of soil test among the rural people.

A comprehensive and long term planning to presence and continuous connection of promoters and experts with potato growers in center or production units for controlling to agricultural function carry out in applying the innovation.

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Weed suppression ability of soybean (*Glycine max* L.) and maize (*Zea mays* L.) in pure and mixed stands

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Abstract: Weed management is a key issue in organic farming systems. Weed suppression has been found to be greater in intercrops compared with sole crops, indicating synergism among crops within intercrops. In order to investigate the Weed suppression ability of soybean and maize in pure and mixed stand an experiment was conducted as complete randomized block design with 4 treatments and 4 replications at the Agricultural Research Station, Sari Agricultural Sciences and Natural Resources University, Faculty of Agriculture, during 2008-2009. Treatment included: monocultures, intercropping of soybean and maize and a control treatment where weeds grew without crops. Result indicated that total weed number was higher in the soybean stand compare to the weed number in the maize-soybean stand. In corn the number of weeds was significantly lower than that for soybean. This research reports weed suppression had been found to be greater in intercrops compared with sole crops and diversity of weeds was decreased in mixed stand, in comparison with monoculture. Mixed culture was one effective method in sustainable crop production.
Keywords: intercropping, soybean, maize, weeds.

Introduction

Weed science has a long history of solving weed management problems for farmers. Over the last four decades most of the solutions to weed problems have been based on herbicide technology (Raj can and Swanton, 2001). There is a national need to find profitable, weed management systems that help farmers reduce both soil erosion and surface water contamination by sediment, nutrients, and herbicides. Research suggests that a biological advantage to intercropping may result from complementary use of growth resources (Banik et al., 2006). Component crops may differ in their use of growth resources over time and space such that when grown together they make more efficient use of light, water, and nutrients than when grown separately. In addition, competition from weeds may be lessened by a combination of crop species occupying two or more niches in the field (Younie et al., 2002). It is commonly cited that intercropping can be an efficient means of weed control. Aladesanwa and Adigun (2008) Reported that intercropping maize with ground nut (*Arachis hypogaea* L.), mungbean (*Vigna radiate* L.), or sweet potato [*Ipomea batatas* (L.) Lam.] reduced weed growth, yield losses and time required for weeding. The objective of this investigation was to determine the effect of intercropping maize with soybean on weeds compared with pure crops.

Material and Method

Field experiment was conducted in a randomized complete block design with four replications at the field station of Sari Agricultural Sciences and Natural Resources, University (53°4/ E, 36° 39/ N) during 2008-2009 cropping season in the north of Iran. In this experiment maize (SC704) and soybean (032) were intercropped. plots were 4×4 m and consisted of four cropping systems: intercropped maize at 50 000 plants ha^{-1} and soybean at 200 000 plants ha^{-1} (MS_1); sole cropped maize at 50 000 plants ha^{-1} (MS_2); sole cropped soybean at 200 000 plant ha^{-1} (MS_3) and control treatment where weeds grew without crops (W). Seedbed preparation consisted of moldboard plowing, disking, and roller harrowing. The desired populations were obtained by over planting and thinning. Weeds were controlled with hand rouging or by mechanical cultivation as needed. A1cm^2 area from each plot was harvested for determination of stover and plants straw (dry matter minus grain) and 2 m^2 area from each plot was harvested for grain yield. Each sample was separated into component crops and dried at 75 ^{c} until a constant weight was measured. Grain yield was determined for Soybean and corn. For weeds biomass calculations, up to 10 randomly selected plants (depending on their size) of each species, (including roots) within each plot were taken and the average of dry weight (after oven drying at 70$^{°c}$ for 24 h) of each species was determined and multiplied by the number of existing plants within the quadrate in each plots. Using MSTAT-C data were analyzed and the significantly differences between different treatments were determent. Using ANOVA method and Duncan Multiple Range Test (DMRT) the means were compared.

Results and discussion

Cropping system had a significant effect on maize grain yield (Table 2). Intercropped maize yields were higher than sole cropped maize. In Intercropping corn yields were increased when soybean rows were increased. This finding is similar to Alford et al., (2003) and Jiang et al., (1993) that yield of corn can be enhanced by intercropping with pulse crops, possibly because biologically fixed N is transferred from legume to corn plants. Soybean yield was reduced by the presence of corn. These changes in yields were probably due to the success of corn in competing for light, water, nutrients and weed suppression (Bonaparte and Brown, 2004; Silvia, 2007). Davis et al., (2002) reported in a cereal/legume mixture, the legume was severely depressed by cereal competition soon after planting. More soybean yield was produced as the population of soybean was increased in an intercropped (Banik et al., 2006). Soybean yield reduction in intercropped soybean compared to the sole soybean, could be due to a higher degree of interspecific competition in mixed stand and the absence of inter specific competition in the mono crops (Rajcan and Swanton, 2001). The delays in senescence in the monoculture treatment increased the grain filling period therefore increasing the size of kernels contributing to higher soybean yield. Also, Increase in grain yield in soybean sole culture was attributed to increase light interception during vegetative and early reproductive periods. This finding is similar to Jiang and Egli (1993) that shade during reproductive growth reduced pod and seed numbers and yield.

Cropping system affected weed growth, sole soybean allowed the greatest weed production. Maximum Weeds biomass and density was obtained in sole culture of soybean. Weed suppression was observed when intercropping was additive, but the depression in above-ground weed biomass was not due to a synergistic effect of crop diversity, but rather to the effects of crop density and identity. This finding is similar to (Agegnnehu et al., 2006) that weed pressure can also be reduced by intercropping. Reduction in weed density and biomass as planting density of the intercropped species increased, observed by other authors (Fugiyoshi et al., 2007; Aladesanwa and Adigun, 2008).

10 weeds species occurred in the experiment (Table 1). The species redroot pigweed (*Amaranthus retroflexus* L.) occurred in practically all experiment, was the most frequent.

This research indicates that intercropping maize with soybean can be done successfully in temperate zone. This biological advantage to intercropping may result from complementary use of growth resources. Component crops may differ in their use of growth resources over time and space such that when grown together they make more efficient use of light, water, and nutrients than when grown separately. In addition, competition from weeds may be lessened by a combination of crop species occupying two or more niches in the field (Younie, and Litterick, 2002). These finding suggest that intercropping maize – soybean suppresses weeds and it is an effective way to biological weed control.

Table1- Occurrence index (number of plots where a certain weed species occurred/total number of experimental plots) for weed species indentified in the experiment.

Species name	Occurrence index (%)
Amaranthus retroflexus	78
Chenopodium album	45
Oxalis stricta L.	21
Stellaria media L.	10
Ambrosia Artemisiifolia L.	5
Sonchus arevenis L.	12
Abutilon theophrasti	42
Senesio vulgar	8
Veronica persica	2
Cynodon dactylon	28

Table 2: Mean of corn grain yield, soybean yield. Weeds density and weeds biomass in different corn and soybean ratio.

treatments	Corn grain yield (kg/ha)	Soybean grain yield (kg/ha)	weeds biomass (kg/ha)	Weeds density (Plant/m^2)
MS1	10.8 [a]	1997.5 [b]	1335[d]	12,8[d]
MS2	9.8[b]	------	1500[c]	14.7 [c]
MS3	------	2268.6a	1650[b]	15.2[b]
W	----	-----	2559[a]	16.6[a]

Means, in each column, followed by similar letter are not significantly different at the 1% probability level-using Duncan' s Multiple Range Test.

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Effect of Different Tillage Methods on the Germination, Grain yield and Harvest index of Two Barley (*Hordeum vulgar L.*) Cultivars

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Abstract

A field Experiment was performed in randomized complete block design with split-plot arrangement with 4 replicates during 2010-2011 growing season to study the effect of different tillage methods on the germination of seeds, grain yield and harvest index of two barley cultivars. The main plots allotted to four levels of tillage (T1= Chisel + Disc two perpendicular + Leveler, T2= Disc two perpendicular + Leveler, T3= Moldboard plow + Disc two perpendicular, T4= Rotary plow); sub plots allotted to 2 varieties (V1= Rihan, V2= Nosrat). The traits evaluated in this trial, including seed germination, yield and harvest index respectively. The results showed traits between different levels of tillage in all traits, there is a significant difference at 5% level. The highest grain yield of tillage treatment by rotary plow 4,774 kg per hectare, which was quite a significant difference between the yields on the other tillage methods. Another significant difference between three tillage did not yield and three in the second group were statistically. Recent experiments also showed that conventional tillage operations on soil imposes heavy despite some advantages, it is able to provide a perfect platform to aggregate seed and maintain soil moisture; therefore, there is not much pressure in the long run into the soil.

Keywords: Barley, Plow, Germination, Grain yield, Harvest index.

Introduction

Botanists believe that barely is one of ancient plants in the world and they speculate it was first found in Africa. Most ancient cultivars known barely are two wild rows found in excavation in south Europe. Evidences show the plant was cultivate in Stone Age. Mankind has cultivated barely for about 10000 ears. It was commonly seen in Ferrous age in Arabic Republic of Egypt. At that age pilled barely was mainly used for preparing bread. In Iran first barely farms are tracked down to 3000BC, and it was the main source of bread during the Great Cyrus (Poursaleh, 1975). Aase (2000) conducted a research to improve production performance of potato through studying relation between plough and depth and physical properties of soil including superficial specific weight, cone index, water penetration rate, viscosity and water erosion, fringing stream irrigation. The research was conducted in a research farm with loam silty. Tillage samples under study were disk, deep plough made by subsoiler plus disk, deep plough by subsoiler and withough tillage. The study found no significant difference between plough methods regarding runoff and water erosion of soil. Bohari (1995) argued that fuel consumption, energy demand and costs decreased by 55% under tillage condition, while there was no difference between seed and product yield. Dikey (1983) concluded in his researches that operation without tillage on soils with fine particles tissue result in operation reduction due to weak condensation and aerate and utilization of disk mould-board plough 25cm results in significant increase in performance.

Results of tillage and cultivating techniques experiments on maize and soy after wheat harvest by Bicer (1987) showed no significant differences between different samples. However, direct cultivating method is featured with more advantages regarding time and energy consumption. Raimbout (1997) concluded that physical structure of soil significantly affect product yield, though it is adjustable through proper tillage management techniques. Hermowan et, al. (1993) showed in their researches that non-tillage soil results in less soil crust, superficial specific weight and soil compression.Unger (1984) surveyed effects of un-plough and mould board ploughed system on superficial specific weight of soil and reported that with the former, superficial specific weight of soil was higher at 15 to 25cm. Ossible (1992) reported that compression sub-layer of soil results in less seed and yarn yield in wheat on un-plough system. They believed that the reduction of yield product is directly affected by soil mechanical strength, shortage of oxygen or accessible humidity and nutrition.

Materials and method

The experiment was conducted in Shahid Bahonar technical high school research farm located in southeast Tehran, north of Varamin, Pakdasht dist in farming year 2009-10. The place of experiment is at 1180m altitude from sea level and 18º28' north and 51º44' east longitude and latitude respectively. Based on Domerten classification, the area is considered as arid areas with average annual precipitation of 170mm (based on 25years statistics by meteorological department). Maximum and minimum temperature is +44 and -14ºC respectively. The experiment was conducted as split plot (partitioned plots) according to fully random blocks with three repetitions. Main factors are four tillage methods, (-T1 chisel mould + two vertical disks + loler, -T2 two vertical disk + loler, -T3 plough using mould-board plough + two vertical disk + loler, -T4 plough with rotary tiller) and subsidiary factors were comprised of two modified barley types (-V1 Reyhan modified type, -V2 Nosrat modified type). Samples were collected from two depths of 0 to 15cm and 15 to 30cm in soil. Having the results obtained, 100kg/10^4m (K2o), 120kg/10^4m (P2O5) dunk and 40kg nitrogen at cultivating and 60kg/10^4m nitrogen P2o5 was added to soil were added we stem was developing. The land under experiment was a 86 * 34 parcel of land with an area of 2924m2; main plots were 10 * 20m used as tillage samples, each main plot was divided into two subsidiary plots with one meter hedge between each. The two cultivate of barely were cultivated randomly in the plots. Staffs carried out seeding manually on 1 Oct. 2009 and seeds were placed at 2 to 4cm depth of soil by disk, afterward rows were arranged in 18cm intervals by furrower. Bushes were completed gold and clusters were dried on 22 May 2010. Three samples were gathered from each plot using a 1m2 wooden frame to separate the area and harvested by sickle. Number of seed in cluster was determined using 20 bunches collected from different section of plots randomly. Average of number of seeds in each cluster was taken as number seeds in each cluster. Moreover, number of cluster in square meter was counted using the 1m2 frame. A specific portion out of 1 square meter was adopted to calculate blossoming rate, afterward seeds blossoming rate underground was determined for three points in each plot. The rate is obtainable using the following formula:

Blossoming rate = (number of blossomed seeds/total number of cultivated seed) * 100.

Results and conclusions

Germination Rate

Early germination demands proper contact between seed and soil (Hadas 1975). According to variance analysis results (table 1), different tillage methods have significantly different germination rate at statistical level of 5%. Comparison on average germination rate of different tillage methods showed highest rate of 85.97% for rotary tiller tillage process, this 7.33% increase to minimum rate found in vertical heavy disc + lowler with 80.1% (table 2). However, no significant difference were found between three samples processed with rotary tiller, mould-board plough + two turns vertical disk + lowler and chisel plough. Soil humidity is one of the limiting factors of yield product, and waste management and ploughing and tillage processes are highly effective on soil humidity. Rotary tiller does not overturn the soil, but only cut the soil to a specific depth. Of specific features of plough is to prepare the bed for seeding by passing over the land and consequently less total extension. In this way, less power is wasted the plough. The plough decreases total weight of tractor and soil compression, while it keeps more humidity of soil and consequently higher germination rate (Sakine & anjel 2006). Rotary tiller replaces plough, disk and other prunes and leave no groove and ridge behind (Mansouri Rad 1995). Results from other studies on different tillage methods for different products showed that depending on local condition, facilities and objectives, each tillage method such as tradition tillage (Unger, 1977m Rashi & De Datta, 1986, Kheiralla et al. 2004), minimum tillage (Hemmat & Eskandari 2004), no tillage (Ambassa Kiki et al. 1996) may have superiority to others.

Seed Yield

Results from data variance analysis showed significant differences between different tillage methods regarding average seed yield (table 1). Maximum seed yield of 4774 kg/10^4m was obtained for samples with rotary tiller tillage, which is significantly different from other tillage method. No significant difference were found between the three tillage methods (chisel plough, two turn vertical disk + lowler, two turns heave vertical disk + lowler and mould-board plough + two vertical disk + lowler), and all were placed in second statistical group. Minimum average seed yield was obtained from sample with tillage by two turns heavy disk + lowler equal to 3964 kg/10^4m, which is 20.4% reduction in comparison with rotary tiller tillage (table 2). Seed yield, as in index, is a pivotal objective in barely modification plant. In this project, increasing performance and reducing undesired trait simultaneously was the main objective, in other words, improving performance along with perfecting features such as resistance against anti- photosynthesis factors, high resistance to bad characteristics of soil, faster germination, early maturation and resistance to diseases. The third objective entails developing biological models relative to different cultivation condition, that is to modify plants with favorable types and features which affect positively on photosynthesis, growth and seed yield (Sahota 2007). Studies by Dick (1992) on soy and Izaurralde et al (1986) on wheat resulted in less yield in comparison with yield from traditional tillage. It seems that different land preparation methods positively or negatively affect the yield through modifying physical and chemical characteristics (Izaurralde et al.) such as humidity (Dick 1992, Baemumer et al. 1973), soil temperature (Swanton & Willholm 1996). On the other hand, several researches have reported damages caused by applying heavy machines such as mould board plough on soil and consequently plant yield. Researches by Barzgar et al. (2003) showed negative effects by mould-board plough on soil humidity at 1m depth and consequently seed yield comparing to mild tillage machines. Yield differences between varieties of barely were significant at 5% statistical level (table 1). Variety Reyhan with 4410kg/10^4m yield is first in the line with 7.7% higher yield comparing with variety Nosrate 4092.8 kg/10^4m (table 2). The two varieties in this study were of newly modified varieties for mild weather recently adopted from advanced lines in Iran. Selection of the varieties was based on height of bushes, average weight of 1000 seeds, high cultivation index and morphological characteristics. However, seed yield of variety Reyhan was higher in comparison to variety Nosrat regarding number of cluster in surface unit and number of seeds in cluster.

Reciprocal effect between different tillage methods and barely cultivates were significant, which hints that seed yield change caused by tillage methods does not follow a uniform path. Two tillage samples with rotary tiller on variety Nosrate and mould boarad plough on variety Reyhan yielded maximum seed product equal with 4939 and 4856 kg/10^4m respectively. Minimum yield was obtained from mould-loader plough + two vertical disk + lowler on variety Nosrate (diagram 1).

Harvest Index

According to the results of variance analysis (table 1) there were significant differences between tillage methods regarding harvest index. Maximum harvest index was obtained for rotary tiller tillage soil equal to 46.7%, therefore difference with chisel plough + 2 vertical disk + lowler was not significant. Tillage samples with two heavy vertical disk + lowler resulted in minimum harvest index of 40.6%, which shows 15% reduction in comparison with rotary tiller samples (table 2). Frouzandeh Shahraki and Khajepour (2005) reported in their survey on effects of land preparation on sunflower and barely growth in hybrid cultivation a significant difference regarding seed yield and harvest index. Least tillage index regarding measured characteristics resulted in minimum yield. According to the results from the survey, subsoiler + disk plough obtains slightly better results for collection wastes in comparison with other cases for sunflower bed in sunflower-barely hybrid cultivation. No significant difference was observed between the samples under consideration. In addition, reciprocal effect of tillage methods and wheat were not significant. However, variety Reyhan with 43.1% resulted in higher index in comparison to Nosrate with 41.9%.

Table1. Analysis Variance of agronomical characteristic

S.O.V	DF	Germination (%)	Yield (kg/h)	Harvest index (%)
Replication	2	75.29	17114.54	51.71
Factor A	3	27.37*	6685.39**	66.69*
Error(a)	6	58.95	19900.93	27.80
Factor B	1	18.37ns	82968.17*	28.30ns
AB	3	7.81ns	52896.28**	125.29**
Error(b)	8	5.456	11914	3.96
CV (%)	-	8.35	13.8	7.65

Ns,*,**: Non- significant and significant at in 0.05 and 0.01 level of probability respectively.

Table2. Means comparison of agronomical characteristic

S.O.V	Germination (%)	Yield (kg/h)	Harvest index (%)
Factor A	-	-	-
T1	82.10 ab	4197 b	43 b
T2	80.17 b	3964 b	40.6 b
T3	83.67 ab	4071 b	40.7 b
T4	85.97 a	4774 a	46.7 a
Factor B	-	-	-
N1	81.91 a	4410 a	43. 1 a
N2	83.67 a	4092.8 b	41.9 a

Means with the same letter in each column have not statistically significant difference.

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Effects OF Rice Husk Application AND Planting ON Some Properties OF Different Textured Soils

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Abstract
Effects of rice husk application (RH) and barley planting (P) on some soil physical and chemical properties were investigated in three different textural soils (clay, loam and sand). The study was conducted in a factorial experimental design with three treatments (control, RH and RHP) using three different soils. After 5% of RH on dry weight bases was applied into six pots of each soil, barley was sown into three pots of each soil sample. All soils including control treatments were incubated together for 6 months under greenhouse conditions without fertilization. Moisture contents in the pots were kept around field capacity with adding distilled water after weighing pots in 2 days interval. While the highest mean AS (46.75%), EC (0.612 dS m^{-1}) and OC (1.378 %) content were determined in clay textured soil, the highest mean BD (1.413 g cm^{-3}), Ks (17.97 cm h-1) and pH (8.57) values were obtained in sand textured soil. Generally, AS, Ks values and OC contents of the soils significantly increased with rice husk application alone (RH) and with barley planting (RHP) when compared with the control treatments. EC values in clay soil significantly decreased with RH and RHP treatments while EC values in loam and sand textured soils increased with these treatments. While RH application was the most effective treatment on BD and Ks in clay, and OC content in sand textured soils, RHP application was the most effective one on AS in loam and OC content in sand textured soils. The results indicated that RH and RHP applications influenced the physicochemical properties of soils in different levels according to their textural classes.
Keywords: Plant growth, rice husk, soil texture, soil properties

Introduction

Soil is one of the most valuable natural resources in the world. More land, especially marginal lands, has been brought under cultivation. This kind of land is particularly vulnerable to degradation by the processes such as; erosion, depletion of organic matter and nutrients, pollution, waterlogging and salination. The proper management system for the degraded soil in physical manner generally includes improvement of the soil structure, control of infiltration and evaporation, regulation of soil temperature and prevention of erosion (Hillel, 2004). It is known that intensive agricultural practices cause soil structural degradation. One of the most important factors for bad soil characteristic is organic matter deficiency in soils. Addition of organic matter to soils is important to solve this kind of problems. Organic matter generally affects soils physical, chemical and biological properties positively (Flaig et al., 1977).
Cropping system has an important role in soil physical properties due to improving soil structure through several mechanisms such as aggregate enrichment by fine roots and associated fungal hyphae (Tisdall and Oades, 1979; Angers, 1998). Many studies have shown that the potential value of grass in cropping systems has a positive impact on improvement or maintenance of soil structure (Tisdall and Oades, 1979; Stone and Buttery, 1989; Gülser 2006). Natural-wastes used as organic fertilizer are important to increase soil productivity and nutrition values and to improve soil physical, chemical and biological activities (Johnson, 1986; Candemir and Gülser, 2007). The objective of this study was to evaluate the effect of rice husk application and barley growth on some properties of different textured soils.

Material and Methods

Soils used in this study were taken from Kavak, Bafra and Dereköy districts of Samsun. Some properties of the samples are given in Table 1. Rice husk as an organic matter source was supplied from a private factory in Terme, and grounded by 1 mm size. Rice husk had 46.97% organic C, 0.38% N by dry weight basis and 126.6 C:N ratio. Soil samples were air dried and sieved through a sieve with 2 mm size opening. After adding the soil samples into pots, a rate of 5% (w:w) rice husk as a dry weight base was incorporated in each pot and mixed homogeneously. Experiment was conducted in 3 treatments (control (C), rice husk (RH) and rice husk + planting (RHP)) with three replicates. After two months incubation, barley was sown in half number of the pots. All treatments were incubated at field capacity under greenhouse conditions 6 more months. Some soil properties were determined as follows; soil texture by Bouyoucos Hydrometer method, bulk density (BD) in each pot by sampling cylinders (Blake, 1965), organic matter (OM) contents by modified Walkley-Black method (Kacar, 1994), water-stable aggregate (AS), by wet sieving method (Kemper and Rosenau, 1986), soil reaction (pH) and electrical conductivity (EC $_{25°C}$) values in 1:1 (w:v) soil:water suspension by potentiometer and condactivimeter (Bayraklı, 1987).

Table 1. Some properties of the soils used in this study.

Soil Properties	Kavak (Clay soil)	Bafra (Loamy soil)	Dereköy (Sandy soil)
Sand, %	22.08	48.31	89.88
Loam, %	26.29	34.29	3.02
Clay, %	51.63	17.40	7.10
Texture Class	C	L	S
pH (1:1)	8.12	8.25	8.60
Organic matter, %	1.46	1.13	0.20
$EC_{25}{}^{o}C$, dS m^{-1}	0.52	0.32	0.19

Variance analysis of the experimental data was carried out in a factorial experimental design with and without planting on 3 treatments (control, RH and RH+P) with 3 replicates. Pairs of mean values were compared by least significant difference (LSD) and relations between the properties expressed by corelation factors (Yurtsever, 1984).

Results and Discussion

At the end of the experiment; effects of the treatments on some soil chemical (soil reaction (pH), electrical conductivity (EC) and organic carbon (OC) values) and physical properties (aggregate stability (AS), saturated hydraulic conductivity (Ks) and bulk density (BD) values) are given in Table 2 and Table 3, respectively.

Table 2. Effects of rice husk application and barley planting on soil chemical properties.

Soil Texture	pH				EC, dS m^{-1}				OC, %			
	C	RH	RHP	mean	C	RH	RHP	mean	C	RH	RHP	mean
Clay	8,38	8,30	8,28	8,32c	0,771	0,568	0,498	0,612a	0,81	1,71	1,62	1,38a
Loam	8,65	8,40	8,40	8,48b	0,448	0,474	0,483	0,468b	0,56	1,54	1,60	1,23b
Sand	8,80	8,50	8,40	8,57a	0,246	0,298	0,339	0,294c	0,05	0,67	0,55	0,42c
mean	8,61a	8,40b	8,36c		0,488a	0,446b	0,440b		0,47b	1,30a	1,26a	
LSD	0,028**			0,048**	0,033**			0,057**	0,065**			0,112**

Table 3. Effects of rice husk application and barley planting on soil physical properties.

Soil Texture	AS, %				Ks, cm h^{-1}				BD, g cm^{-3}			
	C	RH	RHP	mean	C	RH	RHP	mean	C	RH	RHP	mean
Clay	25,87	65,67	48,72	46,75a	0,16	14,60	2,49	5,75b	1,29	0,98	1,15	1,14c
Loam	3,99	26,73	30,62	20,45b	1,80	4,09	7,22	4,37b	1,34	1,19	1,14	1,23b
Sand	4,47	13,06	12,21	9,91c	11,06	20,77	22,08	17,97a	1,50	1,34	1,40	1,41a
mean	11,44c	35,16a	30,52 b		4,34b	13,15a	10,60 a		1,38a	1,17c	1,23 b	
LSD	2,203**			3,816**	5,965**			7,545*	0,038**			0,066 **

The rice husk treatment and planting significantly increased the organic matter content values of the soils when compared with the controls (Table 2). There was a significant difference among the

OC contents of C, L and S textured soils. While the lowest OC (0.05%) was determined in the control of sandy soil, the highest OC (1.71%) was determined in clay soil with RH treatment (Figure 1A). Numerous studies reported that soils organic matter contents increased with the application of different type and amount of organic residues. (Doran and Smith, 1987; İç and Gülser, 2008; Candemir and Gülser 2011).
The highest pH value (8.80) was determined in sandy soil with control treatment, the lowest value (8.28) was determined in clay soil with RHP treatment (Figure 2). The most significant decrease in pH value according to the control was determined in sandy soil with RHP as 4.47% (Figure 1B). Tang and Yu (1999) reproted that the magnitude in pH changes depends on initial soil pH, organic anion concentration and decomposition degree of residue.
The highest EC value (0.771 dS m^{-1}) was determined in control treatment of clay soil, the lowest value (0.246 dS m^{-1}) was determined in control treatment of sandy soil (Figure 2A). In numerous studies, it was reported that organic waste and compost applications into soil increase EC values of soils (Eignberg et al.,2002; Wang and Yang, 2003). Candemir and Gülser (2011) reported that addition of different agricultural residues in to clay and loamy sand textured soils decreased soil pH, bulk density and increased organic carbon, EC, and AS values of the soils according to the control treatments.

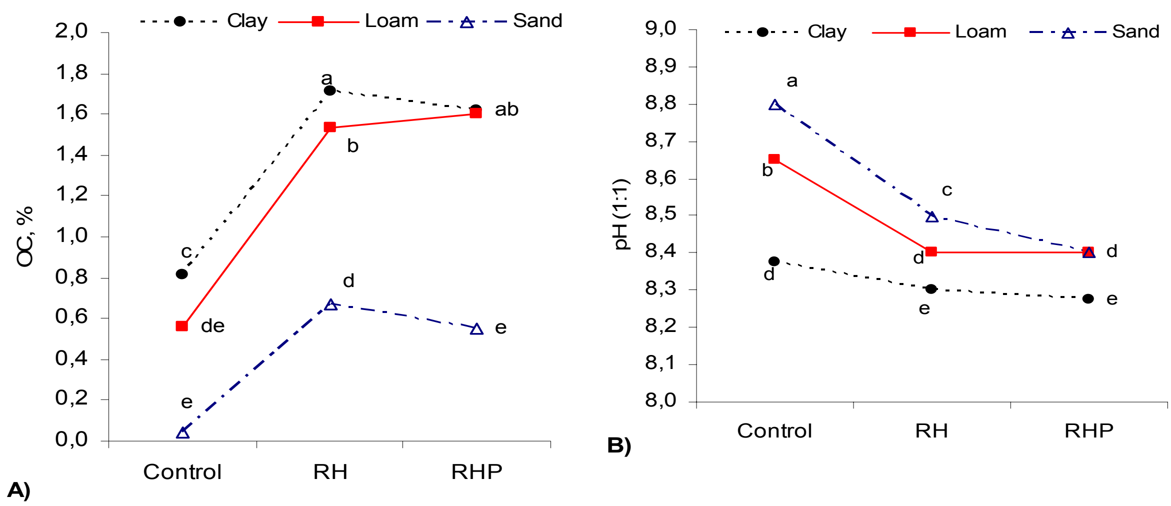

Figure 1. Effects of rice husk (RH) application and barley planting (RHP) on **A)** organic carbon content and **B)** soil reaction (pH) of the different textured soils.

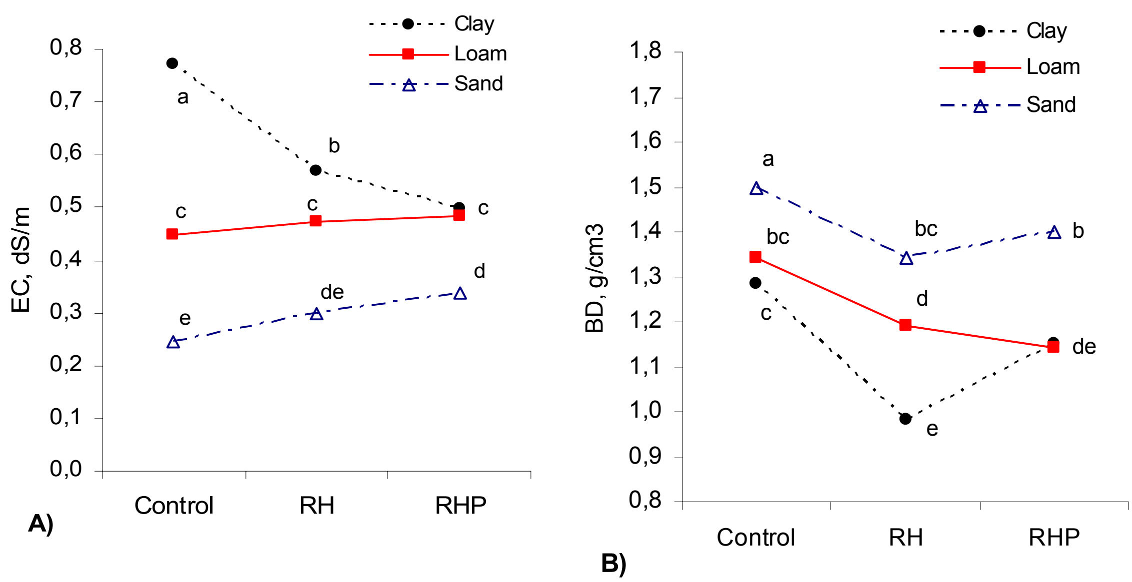

Figure 2. Effects of rice husk (RH) application and barley planting (RHP) on **A)** electrical conductivity (EC) and **B)** bulk density (BD) of the different textured soils.

The lowest BD value (0.98 gr cm^{-3}) was determined in clay soil with RH treatment (RH), the highest BD value (1.50 gr cm^{-3}) was in sandy soil with control treatment (Figure 2B). The highest decrease (24%) in BD according to the control was determined in clay soil with RH application. Soil dry bulk density is often used in soil quality studies as an index of the soil's mechanical resistance to root growth (Reynolds et al., 2003; Drewry, 2006). The lower porosity provides poor aeration, which often is associated with reduced plant growth and may be related to soil borne plant diseases (Miller and Donahue 1995).

Aggregate stability in clay soil was significantly higher than the other soils. The highest AS value (65.67%) was determined in clay soil with RH treatment, the lowest value (3.99%) was in the control treatment of loamy soil (Figure 3A). AS values in clay, loam and sand textured soils significantly increased according to the control treatments with RH application as 153.8%, 569.9% and 192.2%, and with RHP application as 88.3%, 667.4% and 173.1%, respectively. Planting treatment in all soil samples decreased AS values compared with RH application alone. Soil aggregation is important for the resistance of soil surface to erosion and it influences the ability of soils to remain productive (Pinheiro et al., 2004). In a field experiment, Gülser (2006) reported that the growth of forage crops increased macroaggregate size and stability of clay soil, and decreased bulk density compared to a fallow control.

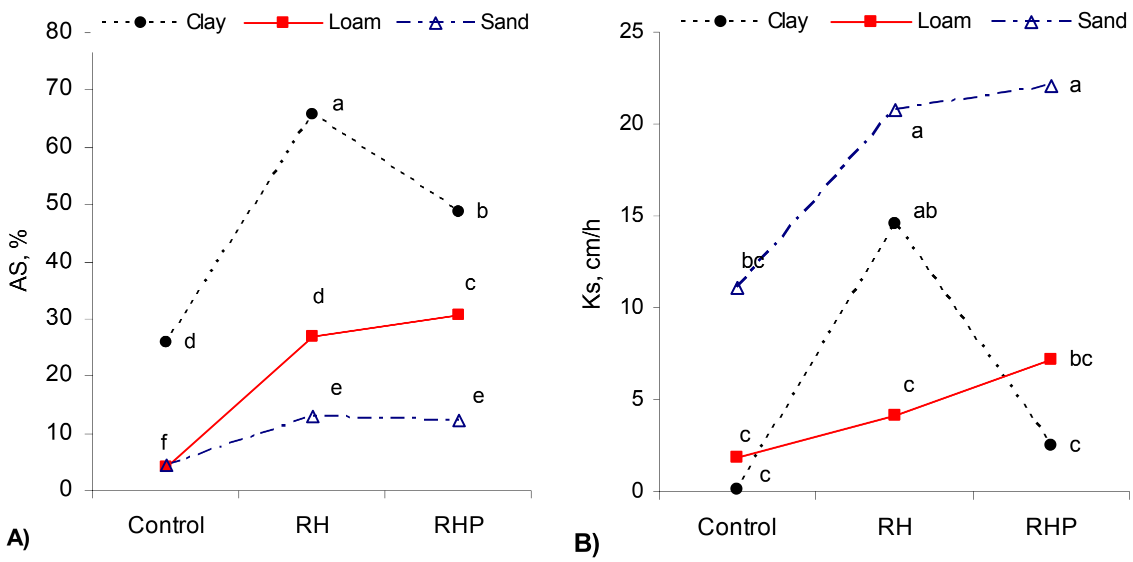

Figure 3. Effects of rice husk (RH) application and barley planting (RHP) on **A)** aggregate stability (AS) and **B)** saturated hydraulic conductivity (Ks) of the different textured soils.

RH treatments significantly increased the Ks values in clay and sand textured soils. The highest Ks value (22.08 cm h^{-1}) was determined in sand textured soil with RHP, the lowest (0.16 cm h^{-1}) value was determined in clay soil with control treatment (Figure 3B). The highest increment in Ks was determined in RH treatment in clay soil. In a complex effect on soil hydraulic conditions, OM not only improve hydraulic conductivity by creating larger soil porosity, but also reduces that by retaining water, allowing less water to flow freely. Organic matter may also affect the pore size distribution of the soil through soil structure development, which also influences hydraulic conductivity (Vishkaii et al, 2010). Additional higher organic material in the soil structural stability, reduce aggregate slaking and dispersion and cause increase in the Ks (Lado et al., 2004).

As a result, this study clearly indicated that rice husk application and barley growth in different textured soils improved the soil physical properties. The increases in aggregate stability, hydraulic conductivity, organic carbon content and decrease in soil bulk density, pH and electrical conductivity values were favourably associated with barley root growth and porosity conditions. The results of the study indicate that the treatments of rice husk and planting sustain a better soil physical health and have a good effect on reducing soil degradation.

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Heavy Metals Content in Urban Soils of Isfahan, Central Iran

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Abstract

Various heavy metals have been reported as dangerous agents to the human health and wildlife when they occur in the environment at high concentrations. The environmental exposure to heavy metals is a well-known risk factor for cancer. Cadmium and lead compounds are classified as human carcinogens by several regulatory agencies. Gastrointestinal cancers (GI Ca) are common malignancies all over the world. Twenty five percent of all cancer-related deaths are attributed to GI Ca. We investigated the levels of two different heavy metals (Cd, Pb) in the soils of the Lenjanat region in Isfahan province, Central Iran where intensive agriculture surrounded by different industries like steel and cement making factories and mining. According to database, many people suffered from gastrointestinal cancers in this region. Two hundred topsoil samples (0–20 cm depth) were collected from agricultural and non-agricultural soils of the region and analyzed for heavy metals. Results showed that the amount of heavy metals decreased with increasing the distance from the factories. The concentration of Pb and Cd was more than 5 and 0.5 mg kg^{-1}, respectively. The total Cd concentration in most of the samples exceeded the suggested Swiss thresholds (0.8 mg kg^{-1}). It seems that aerosols originated from industries and mining activities are the main sources for heavy metals in the agricultural soils of the area. Thus, analyzing heavy metals content in dust could provide us a better insight to solve the problem.
Keywords:Heavy metals, Gastrointestinal cancers, Soil pollution.

Introduction

Human health in towns and cities is strongly dependent on the status of urban soils (Simpson, 1996). In urban and industrial areas, chemical pollution sources are numerous (Madrid et al., 2002; Imperato et al., 2003). Diffuse metal contamination of soils is caused mainly by atmospheric fallout from various sources, the most important being industrial and traffic emissions (Martley et al., 2004; Möller et al., 2005;Rodriguez Martin et al., 2006).
Metal contamination has been shown to reach significantly higher concentrations in urbanized landscapes than in agricultural areas (Imperato et al., 2003;Ordonez et al., 2003), especially in industrial sites (Sterckemanet al., 2000). Direct inputs are other important sources of soil contamination. They mostly come from agricultural activities like sewage sludge or fertilizer spreading (Romic and Romic, 2003). Elevated soil metal concentrations are a serious and current concern for governmental and regulatory bodies for environmental and human risk assessment (Sánchez-Martin et al., 2000; Lee et al., 2006).
Heavy metals such as cadmium and lead present a risk for human health because they are nondegradable pollutants, having a large spectrum of effects (e.g., nervous or digestive system disturbances and carcinogenic effects), especially for young children who are more sensitive than adults (Li et al., 2004).Cadmium is a highly toxic and carcinogenic metal used in metal plating, nickel–cadmium batteries, plastic stabilizers, and pesticides. It can enter the soil by different pathways including atmospheric deposition, and agricultural management activities. Globally, a huge amount of this metal enters the soil due to human activities (Tiller et al., 1999). It is most toxic in its free (ionic) form which easily dissolves and moves through water, threatening groundwater supplies (Vairavamurthy et al., 2000**)**.
The average concentration of Pb in the soils around the world is about 29.2 mg kg^{-1} with the range of <1 to 888 mg kg^{-1} (Ure and Berrow, 1982). Lead is toxic to humans, especially to young children, and to animals. Plant uptake of Pb is very limited. Therefore, soil containing Pb would need to be ingested in order for substantial exposure to occur. Thus the primary route of Pb exposure to humans or animals from soil is by direct ingestion of soil particles or fertilizer rather than via food chain transfer.

Cancer is a serious health problem worldwide, imposing a large economical and psychological burden as well as loss of life and productivity. Cancer is the third most common cause of death in Iran, accounting for 14% of the total death toll.
As both of the cadmium and lead are carcinogenic metal and Gastrointestinal cancers (GI Ca) are common in Isfahan, in this study we investigated the heavy metal levels in urban soils of Isfahan, central Iran.

Materials and Methods
Study Area
The research included 150 km^2 of agricultural top soils of urban and suburban areas of the Lenjanat and Falavarjan regions in Isfahan province. The area located between 51°14′ 29″ to 51°33′15″longitudes and 32°22′50 to 32°37′30″ latitudes. Isfahan is an industrial city in central Iran(Figure 1) in which intensive agriculture surrounded by different industries like steel and cement making factories and lead mining. The soils of this region are Aridisoils. The average annual rainfall and temperature of the region are 150 mm and 15.5◦C, respectively.

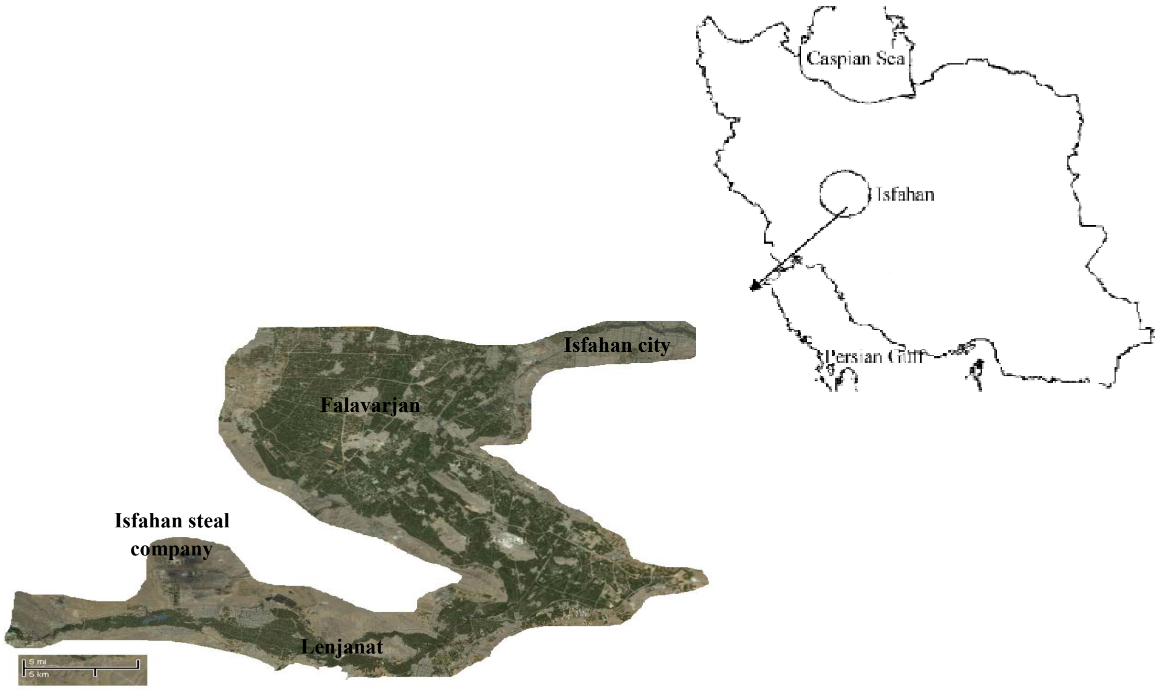

Fig. 1. Location of the study area in Central Iran

Soil Sampling
At each region, soils were randomly sampled from the upper horizon (0-20 cm) and bulked together to form a composite sample. Two hundred topsoil samples were collected and transported to the laboratory.
Chemical Analysis
Soil samples were air-dried and sieved through a <2 mm mesh. Sub-samples were used to measure the physico-chemical properties according to standard procedures. Electrical conductivity and pH of the soil samples were measured in a 1:2 soil to water ratio suspension. Soil texture was determined using hydrometer method. The organic carbon was determined using Walkley and Black's method (Allison, 1986).
For determination of Cd and Lead, 1 gr of the dried samples were digested with 15mL of 4 M HNO3 at 80 ℃ until a transparent solution was obtained (Allen et al., 1986). The solution was then filtered through Whatman No. 42 filter paper and the solution was diluted to 50mL with distilled water. The metal contents of these solutions were determined by flame atomic absorption spectrometry (FAAS).

Statistical Analysis

Descriptive statistics variables including mean, variance, maximum, minimum, coefficient of variation (CV), skewness and kurtosis were calculated using STATISTICA 6.0 software.

Results and Discussions

The summary statistics of the soil properties are given in Table 1. In table 2 descriptive statistics for Heavy metal (Cd and Pb) concentrations in soil was presented. The frequency histograms of the data are shown in Fig. 2.The concentration of Cd showed a nearly normal distribution whereas concentration of Pb had a positive skewed distribution.

Among parameters, coefficient of variability (CV) is the most discriminating factors for describing variability. When CV is less than 10%, it shows low variability; while CV is more than 90%, it shows extensive variability (Zhang et al, 2007). The results in Tables 1 and 2 showed that the CVs of soil Cd and Pb were 44.81%, and 98.37% respectively; indicating soil Pb had high variability in the study area. The CV of Cd in this study indicating moderate variability.

There is no universally accepted safe level for assessing the state of Cd pollution in soils. Therefore, different levels are used in different countries (Kabata-Pendias and Pendias, 2001).In this study, the Environmental standards based on Swiss Federal Office of Environmental, Forest and Landscape were used for the threshold values of heavy metal pollution in the soil (VBBo).

In Table 2, the mean value of Cd concentration in soil was higher than the threshold of 0.8 mg kg^{-1}set by VBBo (FOEFL, 1998) and also the maximum allowable limit (1 mg kg^{-1}) set by United Kingdom (Kabata-Pendias and Pendias, 2001), but the mean value of Pb concentration in soil was less than the threshold of 50 mg kg^{-1} set by VBBo (FOEFL, 1998).Total concentrations of Cd in 80% of soil samples is more than 0.8 mg $kg^{-1}$and more than 1 mg kg^{-1}in 70% of the samples. Nearly 8% of data points have more than 2 mg kg^{-1} Cd and more than 1% of samples have the Cd concentration higher than3 mg kg^{-1}. Total soil Pb content ranged between 1.8 and 115.75 mg kg^{-1}, with a mean value of 16.02 mg kg^{-1}. Nearly 5% of total Pb in soil samples is more than the threshold of 50 mg kg^{-1}.

Table 1. Summary statistics of soil properties

Property	Sample number	Mean	Minimum	Maximum	Variance	Std.Dev.	CV%	Skewness
%Clay	200	39.67	13.28	62.00	122.80	11.08	27.9	-0.31
%Silt	200	33.24	10.00	54.00	52.12	7.21	21.71	-0.14
%Sand	200	27.07	4.72	68.72	177.42	13.32	49.2	0.83
%$CaCO_3$	200	38.57	18.00	74.00	63.99	7.99	20.71	0.60
%O.M.	200	2.26	0.42	4.31	0.55	0.74	32.74	0.38
pH	200	7.48	6.85	7.95	0.04	0.22	2.96	-0.26

Table 2.Descriptive statistics of heavy metal concentration in soil.

Element	Sample number	Mean	Min.	Max.	Variance	Std.Dev	CV%	Skewness	Threshold[a] (mgkg^{-1})
Cd (mgkg^{-1})	200	1.27	0.18	3.12	0.33	0.5	44.81	0.72	**0.8**
Pb (mgkg^{-1})	200	16.02	1.8	115.75	248.44	15.76	98.37	3.73	**50**

[a] Environmental standards in the ordinance of Swiss Federal Office of Environmental, Forest and Landscape.

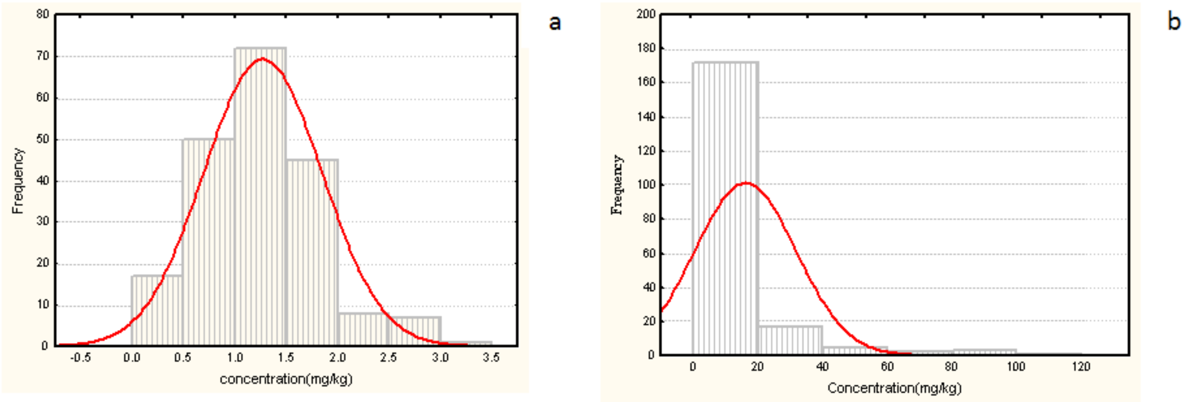

Fig.2.Distribution of total concentration of Cd (a) and Pb (b).

Compared to the threshold values for heavy metals (Cd and Pb) pollution in soils, this investigation (Table 2) indicated that both the studied fields were contaminated especially by Cd. Similar results were also found in the previous studies in china (Liu et al., 2005).

The accumulation of Cd in fields may partly be due to the application of agrochemicals. For example some of the agrichemicals such as fertilizers contain Cd and Pb, which are 0.0005–0.5, 0.0008–0.93 mgkg^{-1}, respectively (Wang and Ma, 2004). Therefore, the long-term application of agrichemicals may result in the accumulation of heavy metals in soils so that heavy metal concentrations in most soils exceeded the threshold values (Table 1). Particularly, the average concentration of Cd was twice the threshold value. Alloway (1990) cites Cd deposition in the EU in urban areas of 3.9–29.6 gha^{-1} year^{-1}, and in rural areas of 2.6–19 g ha^{-1} year^{-1}.One of the other main sources of Cd emissions into the environment was active different industries like steel and cement making factories and mining in the study area.

Automobile emissions are probably the major source of the elevated Pb content in Isfahan urban soils. Highest Pb concentrations were detected in soil samples collected from the border of the city motorway and also from streets areas with high traffic flows (Bargagli, 1998).

In conclusion, high heavy metals content in the soils seems to play important etiological role in the carcinogenesis, Analyzing heavy metals contents in plant, water and dust of the region could provide us a better insight to solve the problem.

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Effect of different isolates of isolated *Azospirillum* from canola rhizosphere on the growth parameters of canola with two different levels of fertilizer under the growth chamber conditions

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Abstract: Nowadays, economic and environmental problems caused by immethodical consumption of chemical fertilizers have caused that trying to biofertilizers production was one of the most important and practical objects of research backgrounds in scientific studies. *Azospirillum* is one of the plant growth-promoting bacteria that has been found in the rhizosphere and the intercellular of the cereals and other plant roots. Releasing the nutrients in soil and also increasing the yield of crops considered as some of the beneficial characteristics of this bacterium. So, a research with the purpose of observation the effect of four superior isolates of *Azospirillum* was carried out on the growth parameters of canola (Hayola 401 variety) under growth chamber conditions, in two fertilizer levels (50% and 100%) in randomized complete block design with three replications and two harvest stages. the effect of bacterial treatments on shoot dry weight, was significant ($p<0.05$) in first harvest, but not significant on the growth parameters in second harvest. The effect of fertilizer levels treatments was significant on shoot dry weight, and root volume in first harvest ($p<0.05$) and also significant on shoot dry weight, root volume, and root dry weight, in second harvest ($p<0.05$, $p<0.01$, and $p<0.05$, respectively).
Keywords: *Azospirillum* spp., Growth parameters, Canola.

Introduction

Canola is a plant which is cultivable in all climates and seasons. As more than 130,000 hectares of the lands of Iran and about 33,000 hectares of the lands of Golestan province have been allocated to cultivate of canola for oil production (Anonymous, 2008). Organic agriculture is based on protecting the ecological balance and strengthening the biological processes by desirable extent (Gholinezhad et al., 2006). In recent decades, sustainable farming systems, conservation of terrestrial organisms community and trying to use of the biological solutions for plant nutrition and providing the health of community have been considered (Saleh Rastin, 2001). PGPR[1] has some positive effects on increasing the efficiency of fertilizer and water usage and ultimately on the crops yield through different mechanisms. This group of terrestrial organisms can play an important role in providing some elements (especially nitrogen) and releasing other elements (such as phosphorus and iron both in soluble and insoluble forms). *Azospirillum* is one of the PGPRs that has been observed in the rhizosphere and the intercellular of the cereals and other plant roots. Nitrogen fixation, plant growth-promoting hormones production, increasing the insoluble-phosphates solubility, siderophores production, improving the water and nutrients uptake, decreasing the ethylene concentration, and ultimately increasing the crops yield considered as some of the beneficial characteristics of this bacterium (Dobereiner and Day, 1976). Plants inoculation with *Azospirillum* is also affective on most of the growth parameters and shoots in addition to root parameters. These changes directly depend on positive effect of *Azospirillum* on increasing some ions uptake such as NO_3^- ، NH_4^+ ، PO_4^{3-} ، K^+ ، Rb^+ ، Fe^{+3} and they are the main reason of increasing the shoots dry weight (Lin et al., 1983; Jain and Patriquin, 1985; Marty and Ladha, 1987).

Materials and Methods

In this research 38 samples of soils and canola roots were used to *Azospirillum* isolation from the different regions of Golestan province. Primary experiments of genus identification, then

[1] Plant Growth-Promoting Rhizobacteria

complimentary experiments of species identification (Tarrand et al., 1978) and also growth-promoting experiments including insoluble-phosphorus solubilizing ability (Arzanesh, 2008; Rajabzadeh, 2009), growth hormone production ability (Bric et al., 1991), and nitrogen fixation ability (Turner and Gibson, 1980) performed on *Azospirillum* isolates. Finally, the effect of 4 superior isolates of *Azospirillum* with control (uninoculated) was carried out on the growth parameters of canola (Hayola 401 variety) under growth chamber conditions, in two fertilizer levels (50% and 100%) in randomized complete block design with three replications and two harvest stages. So, the suitable inoculum of each isolate (with average population of 2×10^8 cfu/ml) was prepared (Vincent, 1970) and inoculated to disinfected canola seeds by 1% sodium hyperchloride solution (Lifshtiz et al., 1987). As the average of bacterial population was 2×10^7 cfu/mg inoculated seeds. After 45 hours, 20 germinated seeds of canola were cultivated in each pot with volume of 10 kg/pot including the mixture of soil and sandstorm (in ratio 8 to 2). Fertilizing was performed based on soil test, before cultivating. So, super phosphate, potassium sulfate, and urea were solved in irrigation water (up to field capacity) and used for 50% and 100% fertilizer treatments (Morshedi et al., 2000). The pots were kept in growth chamber where the light and darkness periods were considered 14.5 and 9.5 hours, respectively. The harvests were performed in 2 stages of 47 days (6-8 foliar) and 103 days (16-19 foliar) after cultivating date. The number of seedlings was brought to six seedlings in each pot before each stage of harvest. In each stage of harvest, seedlings' shoots were sheared from soil surface in each pot. Then, some parameters such as shoot height, root volume, shoot dry weight, and root dry weight were measured in each treatment and replication (Emami, 1996).

Results

Fifty eight *Azospirillum* isolates were isolated from 38 samples of soils and canola roots from the different regions of Golestan province, Iran. Mentioned isolates based on complementary experiments involving growing on NFb[1] medium with 3 percent sodium chloride, the required biotin and the ability of using glucose were separated into 3 related species of *A. brasilense*, *A. irakense*, *and A. lipoferum* (Tarrand et al., 1978; Krieg and Dobereiner, 1986) (Table 1).

Table 1. Related species and the results of growth-promoting experiments on 4 superior isolates of *Azospirillum*

Isolate No.	Related species	Nitrogen fixation (nmol ethylene/ml.h)	Phosphate **solubility** (mg/l)	Auxin production (mg/l)
AC34-III (B_1)	*A. irakense*	0	30.68	1.717
AC39-I (B_2)	*A. brasilense*	0	0	29.297
AC43-III (B_3)	*A. irakense*	0	0	39.799
AC49-VII (B_4)	*A. irakense*	60.64	19.19	9.251

Variance analysis of the effects of fertilizer, bacterial, and both interaction in first harvest on growth parameters of canola (Table 2) showed that effect of fertilizer treatments was significant ($p<0.05$) on shoot dry weight and root volume. Effect of bacterial treatments was significant ($p<0.05$) on shoot dry weight. Also, effect of bacterial and fertilizer interaction was not significant on any growth parameters.

Table 2. The results of variance analysis of effect of treatments on some growth parameters of canola in first harvest.

S.V	Df	Mean of squares			
		Shoot height (cm/pot)	**Shoot dry weight** (gr/pot)	**Root volume** (cm^3/pot)	**Root dry weight** (g/pot)
Replication	2	3.85^{ns}	0.04^{*}	1.36^{**}	0.014^{**}
Fertilizer (F)	1	1.24^{ns}	0.06^{*}	0.53^{*}	0.000^{ns}
Bacterium (B)	4	2.64^{ns}	0.02^{*}	0.08^{ns}	0.004^{ns}
B × F	4	1.79^{ns}	0.01^{ns}	0.06^{ns}	0.004^{ns}
Error	18	1.12	0.01	0.08	0.002
C.V		6.996	14.854	52.413	29.7

*, **, and ^{ns}: Significant at 5%, 1% probability level and no significant, respectively.

[1] Nitrogen Free Blue (NFb)

Results of variance analysis of the effects of fertilizer, bacterial and both interaction in second harvest on growth parameters of canola (Table 3) showed that the effect of fertilizer treatments was significant ($p<0.01$) on root volume and also was significant ($p<0.05$) on shoot and root dry weight. Effects of bacterial inoculation and bacterial and fertilizer interaction were not significant on any growth parameters.

Table 3. The results of variance analysis of effect of treatments on some growth parameters of canola in second harvest.

S.V	df	Mean of squares			
		Shoot height (cm/pot)	**Shoot dry weight** (gr/pot)	**Root volume** (cm^3/pot)	**Root dry weight** (g/pot)
Replication	2	22.7*	2.10**	6.67**	0.09^{ns}
Fertilizer (F)	1	5.2^{ns}	1.41*	19.2**	0.16*
Bacterium (B)	4	10.2^{ns}	0.37^{ns}	0.67^{ns}	0.02^{ns}
B × F	4	3.8^{ns}	0.33^{ns}	0.24^{ns}	0.02^{ns}
Error	18	3.9	0.3	1.02	0.03
C.V		6.12	14.31	17.05	23.87

*, **, and ^{ns}: Significant at 5%, 1% probability level and no significant, respectively.

Results of mean comparison of the different levels of fertilizers on growth parameters of canola in first harvest (Table 4) showed that there was significant difference between fertilizer treatments of 100% and 50% in shoot dry weight and root volume and also amounts of shoot dry weight and root density decreased with reducing the amount of fertilizer. Also, results of mean comparison of the effect of *Azospirillum* indigenous isolates inoculation on growth parameters showed that there was not significant difference between isolates, but this effect was often statistically significant compared to the control.

Table 4. Mean comparison of effect of treatments on some growth parameters of canola in first harvest.

Treatment	Shoot height (cm/pot)	Shoot dry weight (gr/pot)	Root volume (cm^3/pot)	Root dry weight (g/pot)
Fertilizer level of 100% (F_1)	14.889^{a}	0.622^{a}	0.683^{a}	0.144^{a}
Fertilizer level of 50% (F_2)	15.295^{a}	0.533^{b}	0.417^{b}	0.147^{a}
LSD (α=0.05)	0.81	0.066	0.221	0.033
B_0 (control)	13.945^{b}	0.468^{b}	0.521^{a}	0.117^{b}
B_1 (AC34-III)	15.153^{ab}	0.578^{a}	0.5^{a}	0.152^{ab}
B_2 (AC39-I)	15.305^{a}	0.607^{a}	0.5^{a}	0.185^{a}
B_3 (AC43-III)	15.612^{a}	0.627^{a}	0.75^{a}	0.152^{ab}
B_4 (AC49-VII)	15.447^{a}	0.608^{a}	0.479^{a}	0.122^{b}
LSD (α=0.05)	1.281	0.104	0.35	0.052

Values within same column followed by the same letter(s) are not significantly different.

Results of mean comparison of treatments in the second harvest (Table 5) showed that amounts of shoot dry weight, root volume and dry weight decreased with reducing the fertilizer treatment from 100% to 50%.

Table 5. Mean comparison of effect of treatments on some growth parameters of canola in second harvest.

Treatment	Shoot height (cm/pot)	Shoot dry weight (gr/pot)	Root volume (cm^3/pot)	Root dry weight (g/pot)
Fertilizer levels of 100% (F_1)	32.688^{a}	3.982^{a}	6.72^{a}	0.773^{a}
Fertilizer levels of 50% (F_2)	31.856^{a}	3.55^{b}	5.12^{b}	0.626^{b}
LSD (α=0.05)	1.514	0.413	0.774	0.128
B_0 (control)	33.36^{ab}	3.642^{a}	5.4^{a}	0.638^{a}
B_1 (AC34-III)	32.48^{abc}	3.834^{a}	6.2^{a}	0.694^{a}
B_2 (AC39-I)	30.82^{c}	3.398^{a}	5.8^{a}	0.65^{a}
B_3 (AC43-III)	33.66^{a}	3.946^{a}	6^{a}	0.728^{a}
B_4 (AC49-VII)	31.04^{bc}	4.01^{a}	6.2^{a}	0.788^{a}
LSD (α=0.05)	2.394	0.654	1.224	0.203

Values within same column followed by the same letter(s) are not significantly different.

Discussion

Produced phytohormones by *Azospirillum* are effective on respiration rate, metabolism, growth and development of root, so they increase water and nutrients uptake in inoculated plants (Holguin et al., 1999). Bashan et al. (2004) expressed that increasing the efficiency of nutrients uptake through plants was probably due to increase of root surface adsorption as the result of inoculation with growth hormones-producing bacteria such as *Azospirillum*. In fact *Azospirillum* causes to stimulate root branching, increase shoot and root biomass, induce the reproductive cycle by production of plant hormones and causes to increase biomass and nutrient uptake by increasing root permeability (Bashan and de-Bashan, 2005).

The results of this research expressed that effect of *Azospirillum* isolates inoculation was more important in primary stages of plant growth. So, in related experiments of the effect of *Azospirillum* on plant growth parameters, the first time was proposed.

Results of mean comparison of the effect of *Azospirillum* indigenous isolates inoculation (table 4) showed that effect of each isolate was significant on shoot height and dry weight compared to the control. So, effect of phosphate solubilizing, auxin-producing and N_2-fixing isolates can be assessed, positively. But, in second harvest none of the isolates are significantly different compared to the control (table 5). In some cases, *Azospirillum* does not have any affect on the yield and dry material (Bashan and Holguin, 1997; Bashan et al., 2004)

Also, the amounts of biological function in first harvest in each bacterial treatment of B_1, B_2, B_3, and B_4 was 23.5%, 29.7%, 33.97%, and 29.92%, respectively that had increased compared to the control (B_0). Therefore, the growth hormone-producing bacterium had more yield increase than other isolates compared to the control. This afair might be because of becoming the roots mass more capillary and thinner or in other words it might be due to increase of root hairs density which probability the reason is because of producing more auxin in this bacterium. Also, in second harvest the most amount of biological function was related to bacterial treatment of B_4 with the amount of 10.10%. So, N_2-fixing bacterium had the most increase of the biological function compared to the control.

In the case of increasing the yield of inoculated plants with *Azospirillum*, various amounts have been cited, but increase of the yield of inoculated plants in recent usual agriculture has been more than 20%, moderately (Rao et al., 1983; Kapulnik et al., 1987; Kapulnik, 1991; Bashan, 1990).

Increasing nutrients uptake in treatment of phosphate solubilizing bacteria can be due to increase of the available elements (Marty and Ladha, 1987; Lin et al., 1983). This increase can probably be due to the effect of phosphate solubilizing bacteria as *Azospirillum* on increasing root dry weight and consequently, increasing nutrients uptake (Bashan et al., 2004).

Experiments of Biari et al. (2008) on growth stimulation and increasing the nutrients uptake of maize (*Zea mays* L.) by application of plant growth-promoting rhizobacteria (*Azospirillum* and *Azotobacter* strains) in Iran's arid area showed that treating with PGPR(s) significantly increased plant height, shoot and seed dry weight, and number of seeds per row.

Rajabzadeh (2009) surveyed the effect of isolated *Azospirillum* indigenous isolates from rice rhizosphere in different regions of Golestan province, on yield of two cultivars of rice (Neda and Hashemi) in two fertilizer levels under greenhouse conditions. Results showed significant increase ($p<0.05$) in seed yield, number of seed per pot, shoot fresh weight, shoot dry weight, root fresh weight, root dry weight and root density in inoculated treatments compared to the control.

In fact, plant inoculation with *Azospirillum* caused to increase shoots growth and plant height, in addition to affect on roots growth. Even as the result of inoculation, the ratio of shoots to roots increased. Also, the reaction of shoots growth was more than roots growth (Bashan and Dubrovsky, 1996)

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Effect of different isolates of isolated *Azospirillum* from canola rhizosphere on the amount of nutrients uptake with two different levels of fertilizer under growth chamber conditions

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Abstract: In recent decades, economic and environmental problems caused by immethodical consumption of chemical fertilizers have caused to accomplish the special attention and studies on biofertilizers. *Azospirillum* is one of the plant growth-promoting bacteria that can accelerate to release and elements cycle in soil and at last on the yield of crops. The purpose of performing this research was to survey the effect of inoculated canola seeds by *Azospirillum* on the amount of nutrients uptake (N, P, and K) in canola. Therefore, the effect of four superior isolates of *Azospirillum* was carried out on canola (Hayola 401 variety) under growth chamber conditions, in two fertilizer levels (50% and 100%) based on soil test in randomized complete block design with three replications and two harvest stages. The effect of bacterial treatments on potassium and nitrogen uptake was significant ($p<0.05$) in first harvest, but not significant on researched parameters in second harvest. The effect of fertilizer levels treatments was significant on amount of plant nutrients uptake in first harvest ($p<0.01$) and also on plant potassium and nitrogen uptake in second harvest ($p<0.01$ and $p<0.05$, respectively).
Keywords: *Azospirillum* spp., Growth parameters, Canola.

Introduction

Oilseeds are considered as a secondary source of energy in human nutrition after cereals. On the other hand, the meal of canola is used as an important protein-rich food for cattle, poultry and marines (Shahidi and Furouzan, 1997). Canola has attracted the attention in Iran especially in Gorgan province, in recent years. As more than 130,000 hectares of the lands of Iran and about 33,000 hectares of the lands of Golestan province have been allocated to cultivate of canola for oil production (Anonymous, 2008). In recent decades, economic and environmental problems which are caused by immethodical consumption of chemical fertilizers and consider to various inherent capabilities of terrestrial organisms specially microorganisms have caused that trying to biofertilizers production was one of the most important and practical object of research backgrounds in scientific studies (Saleh Rastin, 2001). *Azospirillum* is one of the plant growth-promoting bacteria that has been found in the rhizosphere and the intercellular of the cereals and other plant roots. Nitrogen fixation, plant growth-promoting hormones production, and consequently improving the water and nutrients uptake, increasing the insoluble-phosphates solubility, siderophores and vitamins production, controlling pathogens, synergistic relationship with other useful soil bacteria, nitrite production, bioremediation of the sewage, and toxic residues decomposition considered as some of the beneficial characteristics of this bacterium that ultimately causes to increase the efficiency and yield of crops (Payne et al., 1981; Brock and Madigan, 1991; Stacey et al.,1992). Produced phytohormones by *Azospirillum* are effective on respiration rate, metabolism, growth and development of root, so they increase water and nutrients uptake in inoculated plants (Holguin et al., 1999). Increasing the efficiency of nutrients uptake through plants was probably due to increase of root surface adsorption as the result of inoculation with growth hormones-producing bacteria such as *Azospirillum* (Bashan et al., 2004).

Materials and Methods

In this research 38 samples of soils and canola roots were used to *Azospirillum* isolation from the different regions of Golestan province. Primary experiments of genus identification, then complimentary experiments of species identification (Tarrand et al., 1978) and also growth-promoting experiments including insoluble-phosphorus solubilizing ability (Arzanesh, 2008;

Rajabzadeh, 2009), growth hormone production ability (Bric et al., 1991), and nitrogen fixation ability (Turner and Gibson, 1980) performed on *Azospirillum* isolates. Finally, the effect of 4 superior isolates of *Azospirillum* with control (uninoculated) was carried out on the growth parameters of canola (Hayola 401 variety) under growth chamber conditions, in two fertilizer levels (50% and 100%) in randomized complete block design with three replications and two harvest stages. So, the suitable inoculum of each isolate (with average population of 2×10^8 cfu/ml) was prepared (Vincent, 1970) and inoculated to disinfected canola seeds by 1% sodium hyperchloride solution (Lifshtiz et al., 1987). As the average of bacterial population was 2×10^7 cfu/mg inoculated seeds. After 45 hours, 20 germinated seeds of canola were cultivated in each pot with volume of 10 kg/pot including the mixture of soil and sandstorm (in ratio 8 to 2). Fertilizing was performed based on soil test, before cultivating. So, super phosphate, potassium sulfate, and urea were solved in irrigation water (up to field capacity) and were used for 50% and 100% fertilizer treatments (Morshedi et al., 2000). The pots were kept in growth chamber where the light and darkness periods were considered 14.5 and 9.5 hours, respectively. The harvests were performed in 2 stages of 47 days (6-8 foliar) and 103 days (16-19 foliar) after cultivating date. The number of seedlings was brought to six seedlings in each pot before each stage of harvest. In each stage of harvest, seedlings' shoots were sheared from soil surface in each pot. Then, some parameters such as available phosphorus, potassium, and nitrogen of shoot were measured in each treatment and replication (Emami, 1996).

Results

Fifty eight *Azospirillum* isolates were isolated from 38 samples of soils and canola roots from the different regions of Golestan province, Iran. Mentioned isolates based on complementary experiments involving growing on NFb[1] medium with 3 percent sodium chloride, the required biotin and the ability of using glucose were separated into 3 related species of *A. brasilense*, *A. irakense*, *and A. lipoferum* (Tarrand et al., 1978; Krieg and Dobereiner, 1986) (Table 1).

Table 1. Related species and the results of growth-promoting experiments on 4 superior isolates of *Azospirillum*

Isolate No.	Related species	Nitrogen fixation (nmol ethylene/ml.h)	Phosphate solubility (mg/l)	Auxin production (mg/l)
AC34-III (B_1)	*A. irakense*	0	30.68	1.717
AC39-I (B_2)	*A. brasilense*	0	0	29.297
AC43-III (B_3)	*A. irakense*	0	0	39.799
AC49-VII (B_4)	*A. irakense*	60.64	19.19	9.251

Variance analysis of the effects of fertilizer, bacterial, and both interaction in first harvest on the amount of nutrients uptake of canola (Table 2) showed that effect of fertilizer treatments was significant ($p<0.01$) on phosphorus, potassium, and nitrogen uptake. Effect of bacterial treatments was significant ($p<0.05$) on potassium and nitrogen uptake. Also, effect of bacterial and fertilizer interaction was not significant on any growth parameters.

Table 2. The results of variance analysis of effect of treatments on the amount of nutrients uptake of canola in first harvest.

S.V	Df	Mean of squares		
		Phosphate uptake (mg/pot)	**Potassium uptake** (mg/pot)	**Nitrogen uptake** (mg/pot)
Replication	2	0.23^{*}	112.59^{*}	142.24^{**}
Fertilizer (F)	1	1.22^{**}	236.84^{**}	179.44^{**}
Bacterium (B)	4	0.13^{ns}	93.15^{*}	67.63^{*}
B × F	4	0.08^{ns}	46.61^{ns}	23.89^{ns}
Error	18	0.06	22.36	18.6
C.V		14.64	15.33	14.94

*, **, and ^{ns}: Significant at 5%, 1% probability level and no significant, respectively.

[1] Nitrogen Free Blue (NFb)

Results of variance analysis of the effects of fertilizer, bacterial and both interaction in second harvest on the amount of nutrients uptake of canola (Table 3) showed that the effect of fertilizer treatments was significant ($p<0.01$) on potassium uptake and also was significant ($p<0.05$) on nitrogen uptake. Effect of bacterial inoculation and bacterial and fertilizer interaction were not significant on any amount of nutrients uptake.

Table 3. The results of variance analysis of effect of treatments on the amount of nutrients uptake of canola in second harvest.

S.V	df	Mean of squares		
		Phosphate uptake (mg/pot)	**Potassium uptake** (mg/pot)	**Nitrogen uptake** (mg/pot)
Replication	2	34.53^{**}	4081.82^{*}	3631.52^{*}
Fertilizer (F)	1	15.97^{ns}	8212.12^{**}	3656.57^{*}
Bacterium (B)	4	7.86^{ns}	977.09^{ns}	867.08^{ns}
B × F	4	0.42^{ns}	441.36^{ns}	551.82^{ns}
Error	18	4.58	697.77	634.18
C.V		16.84	14.51	14.56

*, **, and ^{ns}: Significant at 5%, 1% probability level and no significant, respectively.

Results of mean comparison of the different levels of fertilizers on amount of nutrients uptake of canola in first harvest (Table 4) showed that there was significant difference between fertilizer treatments of 100% and 50% in phosphorus, nitrogen and potassium uptake and also amounts of nitrogen, phosphorus, and potassium decreased with reducing the amount of fertilizer. Also, results of mean comparison of the effect of *Azospirillum* indigenous isolates inoculation on the amount of nutrients uptake showed that there was not significant difference between isolates, but this effect was often statistically significant compared to the control.

Table 4. Mean comparison of effect of treatments on the amount of nutrients uptake of canola in first harvest.

Treatment	Phosphate uptake (mg/pot)	Potassium uptake (mg/pot)	Nitrogen uptake (mg/pot)
Fertilizer level of 100% (F_1)	1.81^{a}	33.66^{a}	31.32^{a}
Fertilizer level of 50% (F_2)	1.41^{b}	28.04^{b}	26.42^{b}
LSD (α=0.05)	0.181	3.628	3.308
B_0 (control)	1.36^{b}	24.1^{b}	23.13^{b}
B_1 (AC34-III)	1.62^{ab}	30.88^{a}	28.79^{a}
B_2 (AC39-I)	1.65^{a}	32.41^{a}	30.17^{a}
B_3 (AC43-III)	1.72^{a}	34.01^{a}	31.43^{a}
B_4 (AC49-VII)	1.69^{b}	32.87^{a}	30.84^{a}
LSD (α=0.05)	0.29	5.74	5.23

Values within same column followed by the same letter(s) are not significantly different.

Results of mean comparison of treatments in the second harvest (Table 5) showed that amounts of phosphate, potassium, and nitrogen uptake decreased with reducing the fertilizer treatment from 100% to 50%.

Table 5. Mean comparison of effect of treatments on the amount of nutrients uptake of canola in second harvest.

Treatment	Phosphate uptake (mg/pot)	Potassium uptake (mg/pot)	Nitrogen uptake (mg/pot)
Fertilizer level of 100% (F_1)	13.44^{a}	198.55^{a}	184.05^{a}
Fertilizer level of 50% (F_2)	11.98^{a}	165.46^{b}	161.97^{b}
LSD (α=0.05)	1.641	20.264	19.32
B_0 (control)	11.61^{ab}	172.29^{a}	164.89^{a}
B_1 (AC34-III)	13.26^{ab}	186.88^{a}	176.2^{a}
B_2 (AC39-I)	11.39^{b}	165.04^{a}	156.92^{a}
B_3 (AC43-III)	13.28^{ab}	190.95^{a}	180.22^{a}
B_4 (AC49-VII)	14^{a}	194.86^{a}	186.83^{a}
LSD (α=0.05)	2.6	32.04	30.55

Values within same column followed by the same letter(s) are not significantly different.

Discussion

The high pH in most of the soils of Iran has caused deficiency or fixing some of mineral elements such as phosphorus and potassium. One of the useful ways for increasing the phosphorus solubilizing and decreasing pH around roots is to use the biological fertilizers or phosphate solubilizing bacteria. The studies of some researchers showed that *Azospirillum* has the insoluble-phosphorus solubilizing ability (Arab, 2006; Arzanesh, 2008; Rajabzadeh, 2009). The most amount of phosphate uptake of shoot in first harvest pertained to fertilizer level of 100%. Maybe we can deduce that in primary stages of the growth because of the deficit of phosphorus, the response of canola to complete fertilizer was positive and afterwards the amount of fertilizer level did not influence the phosphate uptake, significantly. Increasing nutrients uptake (including NO_3^{-}, NH_4^{+}, PO_4^{-3}, K^{+}, Rb^{+}, and Fe^{+3}) in phosphate solubilizing bacterial treatment can be due to increase of the available elements (Marty and Ladha, 1987; Lin et al., 1983). Barber et al. (1976) showed that rhizobacteria enhanced absorption of phosphate in young seedlings while causing decreases in older plants.

The most amount of potassium uptake in both first and second harvests pertained to fertilizer level of 100% in inoculated treatments with *Azospirillum* isolates which were significantly different compared to control. Also, bacterial treatments were not significantly different with each other and they all were in the same level. So, maybe we can deduce that in primary months of the growth adding the bacterium with chemical fertilizer caused to increase potassium uptake in canola. Results of the pot and field experiments of Mikhailouskaya et al. (2003) showed that balanced phosphorus and potassium nutrition was the main prerequisite for biological nitrogen fixation of associative N_2 fixing bacteria *Azospirillum brasilense* in association with flax roots. They expressed that phosphorus and potassium had the main role for the formation of beneficial N_2 fixing bacteria-plant roots association. Inoculation technology in complex with P and K fertilization provided a significant increase in flax yield and improvement of fibre quality.

The most amount of nitrogen uptake in both first and second harvests pertained to fertilizer level of 100%. In second harvest there was not significant difference between *Azospirillum* isolates and control. Probably, the reason of limitation to providing fixed nitrogen was that *Azospirillum* does not exude fixed nitrogen into cell like free-living diazotrophs and against symbiosis *Rhizobium*. Also, *Azospirillum* has an effective mechanism to absorb fixed nitrogen that it makes the possibility of ammonium leakage out of bacterium cell difficult. So, it will not be available for plant. Nitrogen fixation just provides a small part of required nitrogen of plant. Besides, N_2 fixation is just observed in the conditions of low pressure of oxygen and afew amounts of nitrogen structures (Michiels et al., 1989). Major part of plants' need of nitrogen can be provided by Biological Nitrogen Fixation (BNF). BNF accomplishes by diazotrophs that have nitrogenase enzyme (Masepohl and Klipp, 1996; Kim and Rees, 1994). Dobereiner and Baldani (1981) reported that there was a difference between two species of *A. brasilense* and *A. amazonense* in nitrogen fixation ability and nitrogen transferring to plant.

The results of this research expressed that effect of *Azospirillum* isolates inoculation was more important in primary stages of plant growth. So, in related experiments of the effect of *Azospirillum* on plant growth parameters, the first time was proposed. Alagawadi and Gaur (1992) expressed that mixed inoculum of *Azospirillum* and *P. striata* caused to increase the yield, N and P uptake in sorghum, significantly. Bhattari and Hess (1993) found that inoculation of wheat with isolated *A. lipoferum* from indigenous varieties caused to increase total nitrogen of shoot up to 39%. They expressed that indigenous bacteria could create better association with indigenous varieties of plants. The reason might be due to their adjustment to environmental conditions and also their more competition ability than non-indigenous bacteria. Experiments results of the effect of two bacteria of *A. brasilense* and *A. chrococcum* on growth parameters of wheat showed that bacterial treatments significantly ($p<0.01$) caused to increase nitrogen and phosphorus uptake. Also, mixed treatments with both bacteria had better effects than single treatments (Mohammadi, 2009). Results of greenhouse experiments of Arab (2006) on effects of isolated *Azospirillum* indigenous isolates from cereals roots in Tehran, Golestan, and Khuzestan provinces on growth parameters and sweet corn yield showed that inoculated plants with N_2-fixing, biosynthesis of auxin hormones, mineral and organic phosphate solubilizing bacteria had high total amounts of nitrogen and phosphate uptake compared to other bacterial treatments.

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Influence of sewage sludge and potassium on macronutrient concentration and partitioning in maize plant

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Abstract

Sewage sludge is a good source of organic matter and plant nutrients. Therefore, its application in most agricultural soils is important to improve some soil chemical and physical properties. To investigate the effect of sewage sludge and potassium (K) on maize yield and macronutrient concentrations and partitioning in maize plant, a pot experiment was conducted in a completely randomized design with three replications. Treatments included four sewage sludge rates (0, 2, 4, and 8%) and four K levels (0, 40, 80, and 120 mg K kg^{-1} soil). Results showed that shoot fresh and dry weights increased significantly following the application of sewage sludge (up to 4%), but root dry weight was not significantly affected, as compared to that of control. Nitrogen (N) and phosphorous (P) concentration in shoot and root, and K concentration in shoot of maize plant increased significantly with increasing sewage sludge levels. Potassium addition significantly increased K in root and N in shoot of maize plants. Translocation factor (TF), defined as shoot/root concentration ratio, indicated that N and K content in shoots were higher than in roots with application of sewage sludge and K. Phosphorous concentration was accumulated in shoots of Maize cultivars (TF>1), but its concentration was higher in roots than shoots of Maize plants (TF<1) treated with 8% sewage sludge and applied K levels. It may be concluded that sewage sludge application have a favorable effect on maize growth. However, soil EC and heavy metals concentrations in plants should be taken into consideration before sewage sludge application in soils.

Keywords: Sewage sludge; K levels; macronutrient concentration; Nutrient partitioning; Maize plant.

Introduction

Most agricultural soils of Iran are generally low in organic matter (OM), usually less than 1%. Therefore, application of sewage sludge in agricultural soils improves some soil chemical and physical properties. Many papers have been published on the beneficial effects of sewage sludge amendment on crop yield and some soil physical and chemical properties, such as improving soil structure, increasing soil moisture, plant nutrients, humus content and cation exchange capacity (Logan et al., 1997; Barzegar et al., 2002). Moreover, amount of K content in sewage sludge is low. Therefore, the aim of this paper was to evaluate the effect of different rates of sewage sludge and K on macronutrient concentration and partitioning in maize plant.

Materials and Methods

Sewage sludge was collected from Industry of Zarghan, Fars province, Iran. The sample was air dried, passed through a 2-mm sieve, and stored in a plastic bag for physico-chemical analyses. Characteristics of sewage sludge and sandy soil (with 93% sand) are given in Table 1. To investigate the effect of sewage sludge and K on growth and macronutrients concentration and partitioning in Maize plant (*Zea Mays* L.), a pot experiment was conducted in a completely randomized design with three replications. Treatments consisted of four sewage sludge rates (0, 2, 4, and 8%) and four K levels (0, 40, 80, and 120 mg K as K_2SO_4 kg^{-1} soil). Plastic pots containing sandy soil and coco peat (1:1 V) were mixed thoroughly with different applied treatments. To prevent nutrients deficiency in Maize plant, based on soil analysis, nutrients were added to each pot uniformly. Maize seedlings were grown for 8 weeks. Roots and shoots of maize plants were harvested, washed, oven-dried at 70°C to constant weight, and dry ashed at 550°C. The ash was dissolved in 2 M hydrochloric acid (HCl) and, after passing through filter paper, was used to determine P and K. Before dry ashing N was measured by Kjeldahl method. Statistical analysis was performed using SPSS and Excel statistical software packages.

Table 1. Selected properties of the sewage sludge and soil.

	pH	EC	TN	OM	K	P	Fe	Cu	Mn	Zn	Cd
		(ds m^{-1})	(%)		mg kg^{-1}						
Sewage sludge	6.79	3.1	2.07	40.2	560	20	1419	60	367	532	9
soil	7.5	0.6	0.005	0.06	30	3	3.6	0.5	1.8	0.6	nd

Results and Discussion

Shoot and root dry weight

Shoot fresh and dry weights of maize plant increased significantly with increasing sewage sludge levels. However, maximum shoot fresh and dry weights were obtained following the application of 4% sewage sludge (Fig. 1). Increase in Maize growth at different sewage sludge rates could be due to elevated availability of nutrients through the sewage sludge application in soil (Bevacqua and Mellano, 1993). El-Dewiny et al. (2006) stated that sewage sludge application could increase fresh and dry weight of radish and spinach plant. Ailincai et al. (2009) revealed that application of 40 Mg sewage sludge ha^{-1} resulted in an increase in yield of winter rape and soybean.

Root dry weight of Maize plant was not significantly affected by sewage sludge levels (Fig. 1). Effect of K and interaction of K and sewage sludge on maize root and shoot weights were not significant (data not presented).

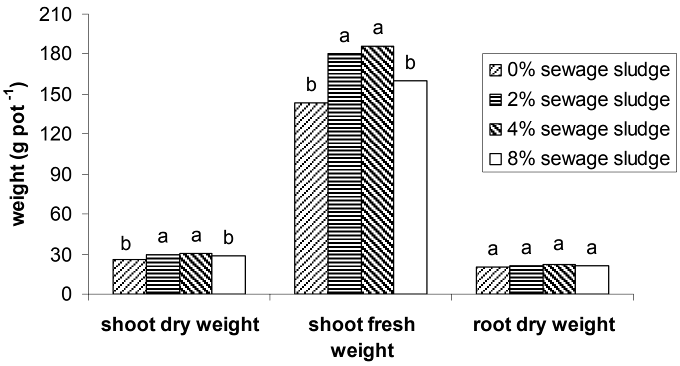

Fig. 1. Shoot fresh and dry weights and root dry weight of maize plant as affected by various levels of sewage sludge

Macronutrient concentration in root and shoot

With increasing sewage sludge levels, mean of N, P, and K concentration in shoot and N and P concentration in root increased significantly, as compared to that of control (Table 2). For example, in comparison to control, addition of 8% sewage sludge increased N, P, and K concentrations in shoot of Maize plant by 2.44, 0.44, and 0.26 fold, respectively (Table 2). Akdeniz et al. (2006) stated that application of 7, 14 and 21 Mg biosolids ha^{-1} increased shoot dry matter yields, N content of leaves, and total N uptake of sorghum plants.

K concentration in root significantly decreased with increasing sewage sludge levels, as compared to control (Table 2). This might be due to the fact that sewage sludge is low in K. Potassium is water-soluble and mostly was removed through treatment process (Lopez-Mosquera et al., 2000).

Potassium addition in soil significantly increased K in root and N in shoot of maize plants, as compared to that of control. However, P and K concentration in shoot of Maize plant were not significantly different between control and applied K levels (Table 1). Mohammad and Athamneh (2004) indicated that application of 20, 40, 80, and 160 Mg sewage sludge ha^{-1} increased N and P concentration in shoots of lettuce plant. However highest K concentration was observed following the application of 20 Mg sewage sludge ha^{-1}. El-Dewiny et al., (2006) showed that organic matter increases N uptake by radish and spinach plants. Our results indicated that sewage sludge is a valuable fertilizer. However, the effect of sewage sludge on soil EC and heavy metals concentration in plants should be taken into consideration before its application in soils.

Table 2. Effect of sewage sludge and K on shoot and root macronutrients concentration in maize plant

	Root			Shoot		
Sewage sludge levels (%)	N	P	K	N	P	K
(each figure is average of four K levels)						
0	0.47 d	1467 c	23520 a	0.91 d	2198 c	45859 b
2	0.96 c	3044 b	17325 b	1.79 c	2874b	47836 b
4	1.37 b	3156 b	16906 b	2.50 b	3630 ab	55968 a
8	1.73 a	3927 a	10112 c	3.13 a	3163 a	57945 a
K levels (mg kg^{-1})						
(each figure is average of four sewage sludge levels)						
0	1.14 ab	2502 b	14417 b	1.90 b	2825 a	50766 a
40	1.17 a	2967 ab	15634 ab	2.11 ab	2898 a	50926 a
80	1.19 a	3358 a	17433 ab	2.12 ab	3022 a	51531 a
120	1.03 b	2768 b	20378 a	2.19 a	3119 a	54384 a

* Means in each column followed by the same letters are not significantly different ($p \leq 0.05$) by Duncan's Multiple Range Test

Nutrient translocation in maize plant

Translocation Factor (TF) was used to evaluate nutrients translocation and partitioning in plant parts. Translocation factor (TF), i. e., the ratio of shoot/root concentration of micronutrients (Kabata-apendias and Pendias, 2001). The TF values for N and K were >1. This indicated that these nutrients concentration were higher in shoots than roots with application of sewage sludge and K, probably due to higher translocation to plant aerial parts (Table 3). Phosphorous concentration was accumulated in shoots of Maize plants (TF>1), but its concentration was higher in roots than shoots of Maize plant (TF<1) treated with 8% sewage sludge and applied K levels (table 3).

Table 3. Translocation factor (TF) of macronutrients in maize plant as affected by applied sewage sludge and K.

Sewage sludge rates (%)	K levels (mg kg^{-1})	TF		
		N	P	K
0	0	1.8	1.3	2.0
0	40	1.7	1.6	2.0
0	80	1.5	1.0	1.9
0	120	3.6	2.2	1.8
2	0	2.4	1.0	3.1
2	40	1.9	1.0	3.4
2	80	1.6	1.0	3.1
2	120	1.7	1.2	1.9
4	0	1.7	1.2	4.1
4	40	1.9	1.0	3.9
4	80	1.8	1.4	3.5
4	120	1.7	1.0	2.5
8	0	1.7	1.1	7.3
8	40	1.8	0.6	5.6
8	80	1.9	0.8	4.4
8	120	1.7	0.8	5.1

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The effect of *Amaranthus blitoides* on growth of *Rhizobium* bacteria

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Abstract

Production of inoculants is an important step to the commercial use of the product. The development of an economic culture medium is necessary to obtain a high quantity of biomass. Amaranth seed is rich in proteins, carbohydrates and lipids. The effect of *Amaranthus blitoides* seeds on the growth of *Rhizobium* isolates was studied. *Amaranthus blitoides* seeds were tested to obtain a growth medium instead of yeast extract. *Amaranthus blitoides* provided maximum growth, Amaranth seed meal can be used instead of yeast extract in medium.
Keywords: Amaranth, *Rhizobium*, medium, yeast extract

Introduction

Rhizobia *(Rhizobium, Bradyrhizobium, Mesorhizobium, Allorhizobium and Azorhizobium)* are unique bacteria because they can fix atmospheric nitrogen by forming symbiotic nodules with leguminous plants. *Rhizobium* spp. are gram negative soil bacteria that have a profound scientific and agronomic significance due to this ability to establish nitrogen fixing symbiosis with leguminous plants, which is of major importance to the maintenance of soil fertility. *Rhizobium* bacteri have useful organisms commercially because of their agricultural application as inoculants for legumens (Buttery et al., 1992).

Production of rhizobial isolates is an important step to the commercial use the product. The development of an economic culture medium is necessary to obtain a high quantity of biomass. The constituens for a medium must metabolite production by providing an adequate supply of energy for biosynthesis and cell maintenance (Bejosano and Corke, 1999; Williams and Brenner, 1995).

Amaranthus seeds contains glutamic acid, tyrosine, valine, leucine, lysine, tryptophan, riboflavin, niacin, biotin, folic acid, lipids, phosphorus potassium, calcium, magnesium, iron and proteins (Bejosano and Corke, 1999; Segura-Nieto et al., 1992; Videira et al., 2002). Usually grain contents fall within the following ranges; crude protein 12-19 %, fat 5-8%, starch 62-69 %, total sugar 2-3 % and ash 3-4% (Williams and Brenner, 1995).

In the present study, we investigated the use of *Amaranthus blitoides* seed meal in isolates of *Rhizobium* V5 and L7 growth cultures instead of yeast extract.

Materials and Methods

Rhizobium V5 and L7 isolates were used in this study. *Rhizobium* V5 isolate was isolated from *Vicia* sp., *Rhizobium* L7 isolate was isolated from *Lathyrus cassius*. Isolates were kept in yeast exract mannital agar medium and stored at 5 °C.

Amaranthus blitoides was used. These materials were previously dried, ground and sieved. Growth in liguid medium with amaranth meals (1, 2, 3, 4, 6 and 8 g/l) were performed according to Grassano et al., (1999). The pH of the media was adjusted to 6.0 with NaOH. Controls contained yeast extract in media. Cultures incubated at 28°C for 3 days on 250 rev. min l^{-1}.

Vicia sp. and *Lathyrus cassius* seeds were inoculated with *Rhizobium* V5 and L7 respectively. Pots were filled 750 g soil. Ten seeds were sown in pots. Seeds were inoculated according to Vincent (1970) as follows; treatment 1; control with no isolates, treatment 2 and 3; inoculations were yeast extract medium and *Amaranthus* seed meals medium, respectively. Plants were grown in pots for 45 days. Roots were separated from shoot sand roots were washed. Plants were then dried for 48 h at 60°C in a forced air oven determine shoot and root dry matter weight.

Results

Table 1 shows the maximum viable cell volues for V5 and L7 obtained in a culture using yeast extract as control and different amaranth concentrations.

Table 1. Effects of treatments on growth of *Rhizobium* isolates

Treatments		Medium pH		Cell / ml^{-1}	
		Rhizobium V5 isolate	*Rhizobium* L7 isolate	*Rhizobium* V5 isolate	*Rhizobium* L7 isolate
Yeast extract medium (Control)		7.35	7.35	1.1 x 10^2	1.4 x 10^2
A.blitoides concentrations (g/l)	1	6.80	6.03	3.2 x 10^2	5.7 x 10^2
	2	6.92	5.51	4.7 x 10^2	4.3 x 10^2
	4	6.73	5.74	7.8 x 10^2	6.9 x 10^2
	6	6.59	5.88	0.2 x 10^2	6.9 x 10^2
	8	6.68	5.88	8 x 10^2	4.8 x 10^2

It has been found that 8g/l of *A.blitoides* seed meal in the media for V5 isolates, 6g/l of *A.blitoides* seed meal for L7 isolate in the media used allowed a higher concentiation of biomass, ranging 0.2 x 10^2 – 8 x 10^2 viable cell / ml, respectively. Media pH values can be seen in Table 1. Values of pH were 5.51-7.35.

The inoculation of *Vicia* sp. with V5 isolate and the inoculation of *Vicia* sp with L7 isolate increased the shoot and root dry weight of *Lathyrus cassius* in the saltly soil as compared to the control (Table 2).

Table 2. Shoot dry weight *Vicia* sp. and *Lathyrus cassius* inoculated with V5 and L7 isolates, respectively.

Treatment	*Vicia* sp.		*Lathyrus cassius*	
	Shoot dry weight	Root dry weight	Shoot dry weight	Root dry weight
Control	100	100	100	100
Yeast extract medium	66.7	89.3	75	96.7
Amaranthus blitoides medium	133*	101.9*	125*	99.0

* Significant at the 5% probability level

Discussion

The results obtained in this study show that amaranth seed meal in medium support rapid growth and high viable cell of *Rhizobium* isolates (Table 1). Several authors suggest amaranth seeds are a good substrate for many microorganisms (Grassano et al., 1999), because it contains proteins and pesticides. The higher viable cell of isolates likely to be obtained with *Amaranthus blitoides* seed meal culture instead of yeast extract (Table 1).

The pH change in amaranth seed meal culture media was different (Table 1). Grassano et al. (1999) found a similar result when bacteria grew in media. *Bradyrhizobium* E109, E110, 5019, 587 and *Rhizobium meliloti* B36, B323, B399, Lq42, Lq51 and U322 grew beter on amaranth meal than on yeast extract as the nutrient source (Grassano et al., 1999). These similar requirements of carbon source confirmed to close relationship of the isolates and the response to Amaranthus seed meal suggests that they require additional growth factors that are not provided by yeast extract. Videira et al. (2002) are the reports to suggest the use of *Amaranthus* seed meal to provide additional nutrients required by rhizobial growth at an industrial level.

Rhizobium inoculants increased the seed germination as compared to the control. Inoculation experiments with *Vicia* sp. and *Lathyrus cassius* showed that *Rhizobium* isolates increased shoot and root dry matter significantly from 133 % to 99 % as compared to the control (Table 2). Shoot growth increased more than root growth. The nitrogen fixing ability of the organism has been postulated as responsible for the increased plant growth and nodulation (Buttery et al., 1992). Maize, pea, lupin, *Amaranthus retroflescus, Echinochloa crus-galli* were colonized with *Rhizobium* isolate. The *Rhizobium* isolate was able to re-establish in the rhizosphere of these plants, after dry storage of the soil for a period up to 12 month (Albareda et al., 2008).

Amaranth seed meal is a low cost substance when compared with yeast extract therefore, future research will be focused on optimizing the growth conditions to obtain higher biomass which is desirable for the industrial development of *Rhizobium* inoculants.

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Growth Promotion of Maize by Rhizobacteria in Salineted Soil

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Abstract
Salinity is a major concern for irrigated agriculture in Şanlıurfa, Turkey. The aim of the research work was to isolate rhizobacteria from cotton, *Salsola crassa, Chenopodium albus* and examined their maize growth promotion properties. Bacterial strains isolated from rhizophere soil of cotton, *Salsola crassa, Chenopodium albus*. All bacterial strains were fast growing, gram positive or gram negative rods. Soil samples of saline soils taken from different areas of Akçakale, Şanlıurfa in Turkey were used in experiments. The investigated were carried out in pot experiments with salinated soil. Tested isolates produced different enzymes. After inoculation with bacterial isolates, the root and shoot growth of maize increased.
Keywords: Maize, rhizobacteria, salinity soil

Introduction
Soil salinity is an important growth limiting factor for most plants. Soil salinity may strongly affect soil biological properties (Zahran, 1997). Harran Plain is located within the Southeastern Anatolia Project (GAP) one of the largest integrated Project of Turkey. Harran Plain has a high potential for crop production. In Şanlıurfa, Turkey, the area of arable salt land is increasing at the rate of 4984 ha (Çullu et al., 2000).
Microorganisms such as bacteria and fungi on the rhizosphere soil were used to plant growth promoting (Zahran, 1997). *Bacillus* and *Pseudomonas* species can produce antimicrobial compounds (De Boer et al., 1998) and enzymes (Nielsen and Sorensen, 1997), they can also be promote plant growth and disease suppression (Zahran, 1997).
Maize is one of the most important crops in Southeastern Anatolia. Inoculation of maize crops with beneficial rhizospheric bacteria may constitute an alternative for improving the availability of nutrients (Riggs et al., 2001). Treatments by using rhizobacteria instead of chemical pesticides and fertilizers, offers a powerful contribution to environmental conservation (Egamberdiyeva, 2005). Use of microorganisms for enhancement of plant production is becoming a more widely accepted praticle in many contries (Egamberdiyeva, 2005).
The aims of this study were (i) to isolates from the rhizosphere of commonly grown plant in saline soils, Harran Plain, Turkey; (ii) to isolate rhizobacteria from maize and examined their plant growth promoting properties.

Materials and Methods
Rhizosphere soil samples were collected from Harran Plain, Akçakale located in the Southeastern Anatolia region of Turkey. Dilutions were made for isolation of rhizosferic bacteria. The identification of relied on standart biochemical and physiological tests according to the classification of Bergey's (Holt et al., 1994).
Cellulase, protease, gelatinase, chitinase, urease activities were detected as described by Egamberdiyeva et al. (2005) and Nielsen and Sorensen (1997). Different amounts of NaCl were adjusted in flasks prior to 10 ml medium to obtain the desired NaCl concentration 0.9, 3, 5, 7, 10, 15, 20 and 25% (w/v) (Mavingui et al., 1992). The inoculation treatment were set up in a randomized design with three replicated. Soils were sampled from irrigated site of Harran Plain, Şanlıurfa. Pots were filled with 750 g soil. Soils were sieved (2 mm mesh) directly after collection. For determination of soil chemical properties, soil samples from the surface to a depth of 30 cm were taken and air dried samples were analyzed for contents of total N, E.C., organic matter, K_2O, $CaCO_3$, and pH. Maize (*Zea mays*) was used as the test plant.
Maize seeds were surface sterilized with sodium hypocloride and distilled water. Ten seeds of maize were sown per pot. The seeds were soaked prior to sowing overnight the both cultures (10^7 cfu ml^{-1}) of bacteria alone. After germination, plants were thinned to obtain seven plants per pot. Plants were grown in pots for 4 weeks. Four weeks after germination, shoots and roots were

separated and dried at 65 ^{o}C before determining the root and shoot dry weight, the criteria for growth promotion were studied as root and shoot dry matter.

Results

We isolated from rhizosphere soil samples isolates belonging to *Bacillus* sp. and *Pseudomonas* sp. *Bacillus* isolates were a Gram positive, motile, spore forming bacterium and *Pseudomonas* sp. isolates were Gram negative, motile. Cells are rod shaped (Table 1).

Table 1.Phenotypic characteristics of isolates

Isolate	Source	Colony morphology	Cell Morphology	Spore	Gram reaction
BM	*Chenopodium albus*	Middle, regular, creamy	Motile, rods, alone	+	+
BM2	*Chenopodium albus*	Small, smooth, creamy, regular	Motile, rods, alone and in pairs	+	+
BP34	Cotton	Middle, rough-swollen regular, creamy, white	Motile, rods, in chain	+	+
BP33	Cotton	Middle, regular, creamy	Motile, rods, alone	+	+
BP12	*Salsola crassa*	Small, smooth, creamy-white, regular	Motile, rods, alone and in pairs	+	+
BM4	*Chenopodium albus*	Large, irregular, creamy-white	Motile, rods, alone and in pairs	+	+
P3	*Chenopodium albus*	Large, creamy	Rods	-	-
P9	*Chenopodium albus*	Large, creamy	Rods	-	-
MH3	Cotton	Large, creamy	Rods	-	-
MK5	Cotton	Large, creamy	Rods	-	-

Tolerant properties of isolates were determined at different concentrations of NaCl. Growth occurs in the range of 0.9-10 %(w/v) NaCl isolates of BM2, BM4, BP12, BP33, BP34, BM, P3, P9, MH3 and MK5 were grown at 10 % (w/v) NaCl; BM, BP33, BP12, P3 and P9 were grown at 15 % (w/v) NaCl; BP12 and P3 isolates were grown at 20 % (w/v) NaCl (Table 2).

Table 2. Resistance of isolates against NaCl concentrations

Isolate	NaCl (% w/v)							
	0.9	3	5	7	10	15	20	25
BM	+	+	++	++	++	++	-	-
BM2	+	+	++	++	++	-	-	-
BP34	+	+	++	++	++	-	-	-
BP33	+	+	++	++	++	++	-	-
BP12	+	+	++	++	++	++	++	-
BM4	+	+	++	++	++	-	-	-
P3	+	+	+	+	++	++	++	-
P9	+	+	+	+	++	++	-	-
MH3	+	+	+	+	++	-	-	-
MK5	+	+	+	+	++	-	-	-

+: weak growth, ++: well growth; -: negative

Table 3 showed results from the extented tested of enzyme production. All of tested isolates demonstrated catalase activity in medium. Amylase activity was positive for BM, BM2, BP34, BP33, BP12 and MK5 and negative for isolates of P3, P9 and MH3. Gelatinase activity was positive for isolate BM and BM2 (Table 3). Isolates showed positive reaction for catalase.

Table 3. Production of hydrolytic enzyme of isolates

Isolates	Enzyme					
	Chitinase	Cellulase	Amylase	Gelatinase	Urease	Catalase
BM	12 mm	4 mm	+	+	-	+
BM2	10 mm	8 mm	+	+	-	+
BP34	10 mm	7 mm	+	-	+	+
BP33	12 mm	2 mm	+	-	-	+
BP12	10 mm	-	+	-	-	+
BM4	10 mm	7 mm	+	-	+	+
P3	-	5 mm	-	-	+	+
P9	-	-	-	-	+	+
MH3	-	+	-	-	-	+
MK5	6 mm	+	+	-	+	+

+: positive, -: negative

Discussion

Ten bacteria were isolated from the rhizospere of cotton, *Salsola crassa, Chenopodium albus*. They were identified as the Gram positive bacteria as *Bacillus* sp. and Gram negative bacteria as *Pseudomonas* sp. (Table 1). Bacterial isolates are salt tolerant. All isolates grow fast in medium supplemented with 0.9 %, 3 %, 5 %, 7 % and 10 % NaCl (Table 2). Also, similar results have been observed in other soil bacteria (Marten et al., 2000; Egamberdiyeva, 2005).

Previous studies reported by Egamberdiyeva (2005); Mavingui et al., (1992) have shown that changes in osmotic potential exerted by salt concentration alter the stucture of lipopolysaccharides and proteins of bacteria in response to salt stress and some bacteria accumulate several solutes to overcome the osmotic stress induced by salt when growing in association with plants. In this study, decreased growth of isolates with increasing salt concentration was registered. Similar to this, Egamberdiyeva (2005) was of the view that increasing salt concentrations may have a detrimental effect on bacterial populations as a result of direct toxicity as well as through osmotic stress. The soil bacteria have evolved a variety of adaptive mechanisms in order to restore the cell turgor pressure between cell and the environment (Mavingui et al., 1992).

In this study, the bacterial isolates that produce different hydrolytic enzymes such as chitinase, amylase, cellulase, catalase, gelatinase, urease. In earlier studies the enzymatic activities have been found to be rhizosphere microorganisms (Nielsen and Sorensen, 1997). Protease, chitinase and cellulase activities have been proposed to be involved in biocontrol processes (De Boer et al., 1998). BM, BM2, BP34, BP33, BP12, BM4 and MK5 isolates showed positive reactions for chitinase (Table 3), which corresponds with soil bacteria reports in the literature (Egamberdiyeva, 2005; Nielsen and Sorensen, 1997).

Soil prior to sowing were analyzed and the clay had the following properties: E.C. 1.73 mmhos/cm; organic matter, 1.64 %; K_2O, 394 kg/da; N content, 0.15 %; $CaCO_3$, 13.6 % and pH, 8.1. Our isolates increased seed germination. Maize yield increases of 18-22 % have also been reported by Rosas et al. (2009) after inoculation with plant growth promoting rhizobacteria. Our isolates can play an essential role in helping plants establish in saltly soil.

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Influence of Different Culture Substrates on the Number of Transferable Tobacco Transplant by Method of Float System to the Main Land

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Abstract

This study was conducted to the purpose of selection of best substrate for the cultivation of tobacco transplant production by method of float system in a completely randomized design at the greenhouse tobacco research center of Tirtash – Iran. In this experiment were performed seven different treatments include of Peat, noshahr's soil, Kurdistan's soil, Peat + vermicompost(2:1), noshahr's soil + vermicompost(2:1), noshahr's soil+cocopeat(2:1), peat + rice bran(2:1) with three replicates. Factors measured at the this experiment are include of the number and percentage of germination, the number and percentage of spiral roots, the number and percentage of transferable tobacco transplanting to the main land, the shoot green weight, shoot dry weight, shoot dry matter percentage, stem length, stem diameter, root length, root fresh weight, root dry weight and root dry matter percentage. Results of analysis of variance showed that effect of different substrate on the factors of the number and percentage of root spiral, shoot dry and green weight, shoot dry matter and root length percentage at level 1% and factors of the number and percentage of transferable transplanting tobacco at 5% level had a statistically significant difference. Comparison of mean the number and percentage of germination showed that Peat beds, Kurdistan's soil and peat + vermicompost have the highest yield and peat + rice bran Bed the weakest yield. According to purpose if number of transplants of transferable to main land is the only criterion, all treatments are suitable for the cultivation of tobacco except peat + rice bran treatment.

Keywords: Cocopeat, Float system, Peat, Transplanting of tobacco, Vermicompost**.**

Introduction

Tobacco is one of the major crops that plays an important role in economics producing countries and income derived from different products of this plant are significantly from national income producing countries. Types of commercial varieties of tobacco is cultured for the production of smoke in Iran. Planting areas of cigarette tobacco Include: The province Mazandaran that is equivalent whit 31% of the total under cultivation area of tobacco, The province Golestan with 24% under cultivation area, Gillan with 21% of under cultivation area, West Azarbaijan with 19%, East Azarbaijan, Kermanshah and Kurdistan 5% rest form of under cultivation area smoking tobacco(Gholizadeh, A., (2008). tobacco is From of the family Solanaceae (Ahi Far, H., (1998) and the genus Nicotiana with scientific name Nicotiana tobaccum(Scientific Bulletin., Research.(1995), n=24. The plant is selfing (Tobacco newsletter,. (1998) that it is cultivated usually annual. Commonly, tobacco growing on beds of peat as main material is formed with different amounts of vermicompost and Perlite. Particle size distribution in terms of size and with their food from is important factor in competency of culture substrate can be considered for transplant production. Particle size is determined in culture without soil, similar is with soil texture and by the relative values and particle size of mixture components (peat, vermicompost and perlite) .distribution size of particles are determined in a substrates, many features as aeration, water-holding capacity, drainage and continuity properties are important that in plant growth. Mr. Beuchat. A. In italy proved that application of peat alone not suitable for transplant production and should be more accurate than select the appropriate combination (Beuchat, A. (2000). He was believed that origin of peats are from natural materials therefore the quality has diverse and are difficult determination their physical and chemical properties. Mr. Harrell N. E. showed that substrate can be appropriate with different sizes constituent particles for transplant production. Shi.x.et.al. reported the beds ratio plays important role in growth and development tobacco transplanting. In mixture peat, vermiculite and perlite, ratios of 50 to 70 percent of peat is suitable for development of transplanting tobacco (Shi, x., G. Liu, and J. chen,.(2001). High ratio of 80% or fewer than 40% of the peat is inappropriate for growth and development of tobacco. This study was performed in order determination most suitable substrate by using internal materials for growth of tobacco seedling in the nursery floating (Caruso,et al., (2001).

Materials and Methods

According to the project, the aim of it is selecting the best substrates replace of peat imported for production of transplant the floating method and review them in the number of transferable tobacco transplanting to the main land. Therefore to perform it was done completely randomized design with 7 treatments. treatments (substrates) include: peat, kurdistan soil, noshahr Forest soil, volumetric ratio 2:1 peat + vermicompost, volumetric ratio 2:1 noshahr soil + vermicompost, volumetric ratio 2: 1 noshahr Soil + cocopeat and volumetric ratio 2:1 Peat + rice bran(ratio 2 for peat and soil noshahr and the ratio of 1 for vermicompost, cocopeat and rice bran) was performed in 3 repeat.

At first were sterilized each of the materials used and were combined together to volume ratios. For this purpose, each of the trays 220 pore filled with the above compounds that each hole has capacity 23.5 ml, after preparation form of tobacco floating nursery and in the middle every house was placed a coated seed variety K_{326} . Viability of seeds was determined before seeding. After trays were placed in the nursery. According to number and repeat treatments was determined dimensions of ponds and inside their to prevent from water out were covered with thick black plastic. In ponds should be 12cm water level and before achieve to this height per cubic meter was used 500gr fertilizer Gromor (20:10:20), 30gr toxin Fungicides Redomil mankozeb, 30gr algae toxin Mishokap (Ranjbar, R., (2009) and to reduce or absorption Bicarbonate water used(5 meq/Lt) 59.9cc concentrated sulfuric acid(Gholizadeh, A., (2008). Three weeks after seeding and at the time the four-leaf seedlings was done similar to the first stage of fertilization. At this stage counting and were calculated number of germination and number spiral roots. In the meantime 3 steps were done header handling operations and other factors were measured 7 weeks after seeding. Time of germination seeds, the number of germinated seeds, the number of usable transplant, transplanting height, weight of dry and green transplanting, stem diameter and etc. were determined. For measuring the above factors was selected randomly of each tray Number of 15 transplant and their average was calculated For the each tray for the analysis of variance and review factors in the statistical software MSTAT-C analyzed were used at level 1 and 5 percent.

Results and discussion:

The results of evaluation of different substrates showed that factors of spiral roots, shoot weight, shoot dry weight, percentage of shoot dry matter and root length at % 1 level and percentage of transferable transplant of factor at level 5% have significant differences but from the statistical point factors of percentages of germination, stem length, stem diameter, root fresh weight and root dry weight showed no significant difference (Table 1).

Table 1: Analysis of variance for different traits affected by the experimental substrates

Resources of Changes	Grading	**Mean Squared**									
		percentages of germination	percentages of spiral roots	Percentage of transferable transplant	root fresh weight	root dry weight	Stem length	Stem diameter	root length	Shoot fresh weight	Shoot dry weight
treatment	6	n.s 326.285	** 14.402	* 1047.047	n.s 2.132	n.s 0.005	n.s 6.175	n.s 0.919	** 21.945	** 646.707	** 1.759
Experimental error	14	194.547	0.862	308.498	0.892	0.003	3.195	0.859	1.592	70.958	0.267

n.s: Showed no significant difference, **: At% 1 level have significant differences, *: At 5% level has significant differences

Table 2: physical and chemical characteristics of the test Substrates

Substrates	pH	Ec (ds/m)	Cl^- (meq/lit)
peat	6.513	1.031	3.4
Kurdistan Soil	7.039	0.948	1.2
Noshahr Forest soil	7.161	2.641	1.2
Peat + Vermicompost	7.067	1.971	2.5
Peat + rice bran	5.737	1.722	7.6
Noshahr Forest soil + vermicompost	7.215	2.850	2.2
Noshahr Forest soil + coco peat	7.038	2.807	7

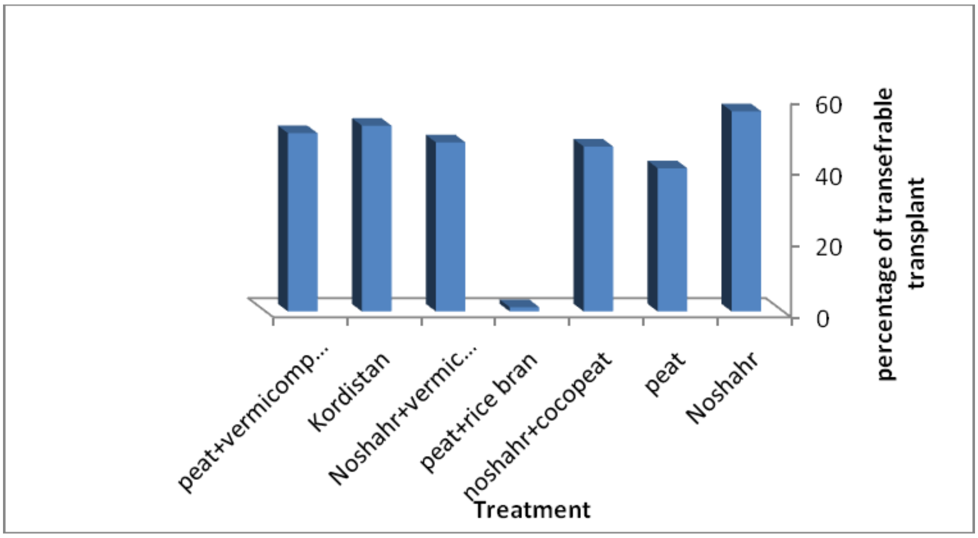

Figure 1. Impact of culture substrate on percentage of transplant transferable

The number and percentage of transplant transferable to the field: Compare data the number and percentage of transplant transferable to the field indicates that all treatments except for treatments peat + rice bran have transmissivity transplant high to the field. It can be cause of related to the composition of rice bran. Because with added rice bran to the peat rate Ec and Cl substrate increased and pH decreased to 5.737 and therefore it causes reduce the number and percentage of transferable transplant to the main land. Finally in between treatments, respectively the best treatments include:1. Noshahr Forest soil, 2. Peat, 3. Peat + Vermicompost, 4. Noshahr forest soil + coco peat, 5. Noshahr Forest soil + vermicompost, 6. Kurdistan Soil, 7. Peat + rice bran. Studies have shown that is better are existed particles with different size in substrate culture. Substrates that their textures have vermiculite or perlite or both these materials are appropriate than to those that 100% their texture is peat.

Suggestions

According to the results, if the objective be only criterion all treatments for tobacco cultivation are appropriate except treatment peat + rice bran. As peat is imported culture and cost, thus should be used other alternatives to be economical.

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Studying some storages of organic carbon as an indicator of paddy soils quality

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Abstract
Labile storage of organic carbon is one of the most important soil quality indicators because of high sensitivity to change in management practices. Particulate Organic Matter (POM) of the labile fraction only represents a small proportion of the total organic matter. They play a key role in the cycling of nutrients, because they are very dynamic. On the other hand carbohydrates are another labile storage of organic carbon that play an important role in forming and sustainability of soil aggregate, so they are considered as valuable indicator in assessing the soil quality. Due to intensive management practices, paddy soils are given a high degree of attention. Therefore to get rid of the threats in rice production, performance the plan of consolidation, equipping and modernization of lands is undeniable necessity. This study was conducted to investigate the possibility of using some of the labile organic carbon storages as the indicators sensitive to management, to assess the impact of equipping and modernization on paddy soil quality. Three locations were selected in Mazandaran province (traditional, consolidated in 9 and 18 years ago). Three replications of soil samples in each field (depths of 0–5 and 5–15 cm) were taken to determine the soil properties. Organic carbon (OC), hot-water-soluble carbohydrate, dilute acid hydrolysable carbohydrate, and POM were measured. Also the amount of organic carbon and carbohydrates extractable by dilute acid and hot-water in POM, and Mean Weight Diameter (MWD) of aggregates were determined. The results showed that the aggregate stability had a significant correlation with OC content of POM. This indicates that OC content of POM can be a suitable indicator for showing paddy soil quality, particularly in relation to soil aggregation and structural stability.

Keywords: Aggregate stability, Carbohydrates, Consolidation of paddy soil, Labile organic carbon storages, Particulate Organic Matter.

Introduction

As an essential indicator of soil quality, soil organic carbon (SOC) and its fractions play an important role in many soil chemical, physical, and biological properties. However, soil total organic carbon might not be sensitive to changes in soil quality resulting from soil management practices changes. As soil organic carbon is a heterogeneous mixture of organic substances, the different forms or fractions of SOC might have different effects on soil fertility and quality. Accumulating evidence suggests that certain fractions of soil organic carbon are more important in maintaining soil fertility and are, therefore, more sensitive indicators of the effects of management practices compared with the soil total organic carbon (Cambardella and Elliott, 1992). Among the different forms of SOM (labile, slow and recalcitrant), labile soil organic matter storages with turnover times of few days to months can be considered as fine indicators of soil quality which influence soil function in specific ways and are much more sensitive to change in soil management practices (McGill et al., 1988).

Particulate Organic Matter (POM) of the labile fraction that represents only a small proportion of the total organic matter, play a key role in the cycling of nutrients, because they are very dynamic. Dissolved Organic Matter (DOM) is a major controlling factor in soil formation (Dawson et al., 1978) that plays a significant role in many chemical and biological processes in soils. The labile soil organic matter storages seem to have a close association with one another and have important impact on soil quality (Laik et al., 2009).

Paddy soil, as an important soil resource, is an anthropogenic soil, and its evolvement and formation are affected greatly by changes occur with consolidation and leveling. The objective of this study was to determine and evaluate the effects of consolidation on paddy soil organic carbon fraction and relationships between soil organic carbon fractions and soil water stable aggregation.

Materials and methods

Three paddy fields were selected in a location of Mazandaran province from Iran. They were included traditional field and consolidated fields in 9 and 18 years ago. Three replications of soil samples in each field (depths of 0–5 and 5–15 cm) were taken to determine the soil properties.

The distribution of water-stable aggregates was estimated by the wet-sieving technique, described in detail by Kemper and Rosenau (1986). In the procedure, 40 g of the < 8mm aggregates were placed on the topmost of a nest of sieves of diameters 4.75, 2, 1, 0.50 and 0.25mm and pre-soaked in distilled water for 10 min before oscillating in water. After sieving the aggregates remain on each sieve were dried at 105°C for 24 h and weighted.

The procedure for particle size fractionation was adapted from methods outlined by Gregorich and Ellert (1993). A 50 g soil sub-sample was dispersed with sodium hexamethaphosphate. The sand size fraction (>53µm) was obtained by wet sieving using a 53 µm sieve. The silt size fraction (2-53µm) was obtained through successive sedimentation-decanting cycles. Dry weights of soil fractions were obtained at 40°C and samples ground in a mortar to pass a 0.5mm sieve screen. Carbon was determined within particle size fractions and whole soil using the Walkley Black procedure.

The carbohydrate distribution in each water-stable aggregate was determined in duplicate in two types of soil extracts, i.e., dilute acid and hot water. In the dilute acid-soluble extract 1 g of soil was mixed with 10 ml of 0.25 M H_2SO_4 and shaken in a rotary shaker for 16 h; in the hot water-soluble extract1g of soil was mixed with 10ml of hot distilled water (85°C) and heated for 2hr 30min. After each extraction and centrifugation, 2 ml of the supernatant solution were used to determine the carbohydrate concentration using the phenol-sulphuric acid method (Dubios et al., 1956). The absorbance was read in a spectrophotometer at 490 nm using glucose standards.

Results and Discussion

From 1950s different soil characteristics or behaviors are used as its quality indicators. Probably most important of them is MWD, which basically used for quantitative description of soil structure (Lovey and Miller, 1997**).**

The result of wet sieve (Fig.1) shows that soil of rice fields with traditional farming systems (A) to consolidated soils (B,C) has lower aggregate stability. Difference between aggregate stability of consolidated paddy soils may be developed due to technique of performance design, the amount of damage caused by land leveling and soil management after that.

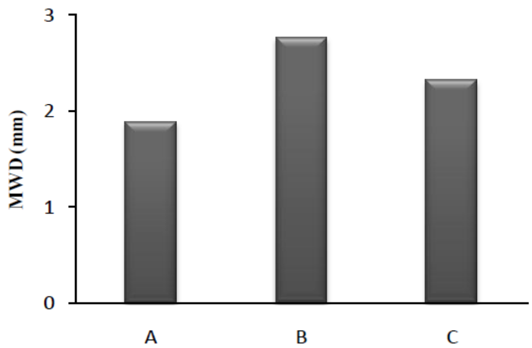

Fig.1. Evaluation of aggregate stability in three fields:
(A) Traditional, (B) Consolidated in 18 years ago, (C) Consolidated in 9 years ago.

Probably, remaining without cover in traditional paddy soils has been leaded to increase of crop residue and fertilizer inputs decomposition, and remove of large amounts of organic carbon of POM from soils after rice harvest (Fig.2).

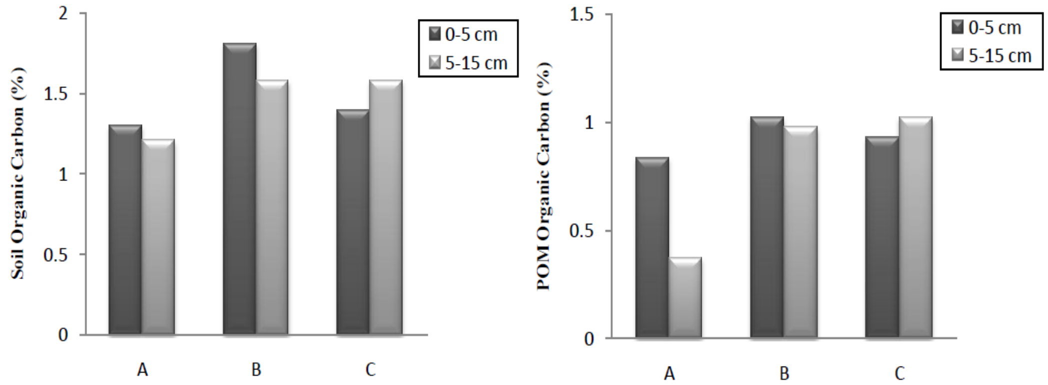

Fig.2. Evaluation organic carbon content of soil and POM in two depths from three fields: (A) Traditional, (B) Consolidated in 18 years ago, (C) Consolidated in 9 years ago.

Fertilization and crop management may influence the feedback between SOC and soil aggregation and thus the protection of SOC (Blanco-Canqui and Lal 2004). Understanding the physical protection mechanism of SOC will contribute to the adoption of appropriate management practices for SOC accumulation and stabilization in paddy fields (Guo and Lin 2001). Great potential for soil C sequestration has been found in rice paddies (Pan et al., 2003; Liu et al., 2006).

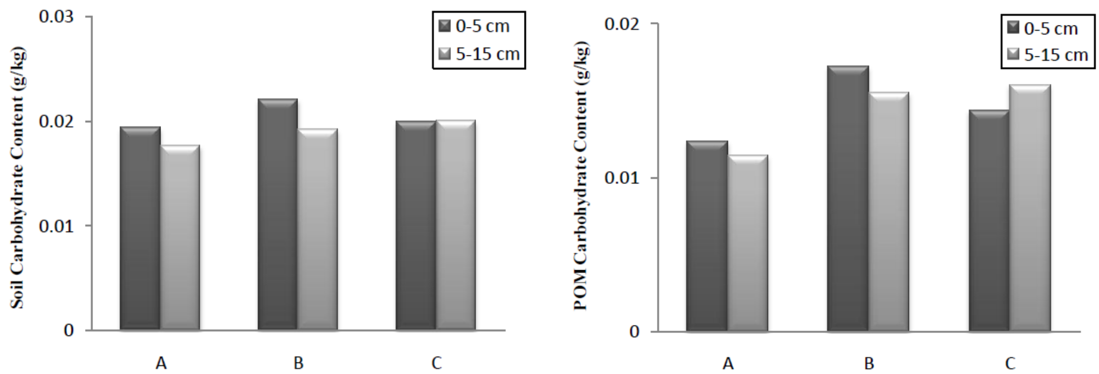

Fig.3. Evaluation carbohydrate content of soil and POM extracted with hot water in two depths from three fields: (A) Traditional, (B) Consolidated in 18 years ago, (C) Consolidated in 9 years ago.

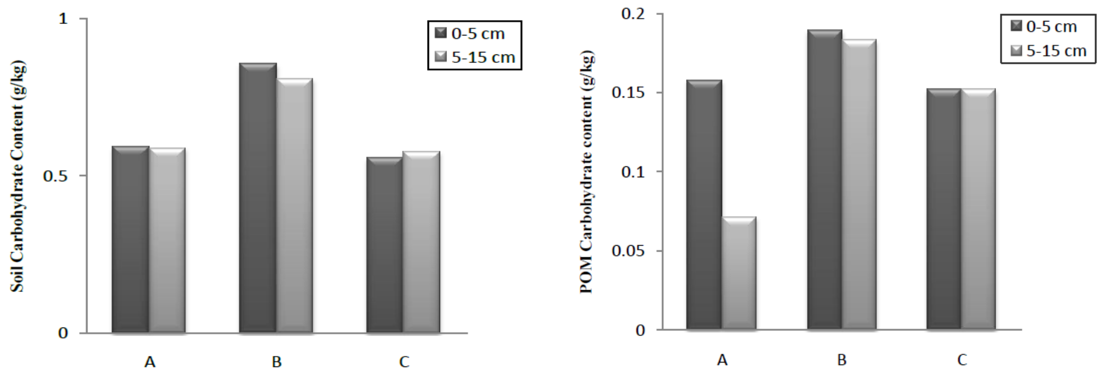

Fig.4. Evaluation carbohydrate content of soil and POM extracted with dilute acid in two depths from three fields: (A) Traditional, (B) Consolidated in 18 years ago, (C) Consolidated in 9 years ago.

In three locations, content of carbohydrate that extracting with dilute acid was more than what extracting with hot water (Fig.3,4).
Haynes and Francis (1993) reported that extracting with dilute acid can also bring out hemicellulose from soil, so content of carbohydrate extractable with dilute acid are more.
Unfavorable consolidation and leveling effects could be observed in the result of consolidated field in 9 years ago (C), because storages of organic carbon in this field have an irregular increase with depth.
One of the basic requirements for achieving a uniform and satisfactory quality of efficient field is precision leveling of field surface. Farmers are particularly interested in the quality of the consolidated plot, in particular, the accuracy of the leveling. Accurate leveling of the paddy field is important for better productivity of the land and labor. When the initial leveling work undertaken for field consolidation is not sufficiently accurate, farmers generally continue leveling as a part of their farming activities. This leveling work entails moving soil from high points to low points. The energy taken to move the earth is related to the volume of earth and the distance it is moved.
In among of evaluated labile storage of carbon in 0-5cm depth, organic carbon of particulate organic matter correlated with MWD at a significant level of $P<0.01$.
Some research workers (Chaney and Swift, 1984; Christensen, 1986) found a direct correlation between soil organic matter (SOM) content and aggregate stability; whereas others (Dormaar, 1983; Hamblin and Greenland, 1977) reported that the fractions of OM rather than the total amount are important in modifying the structural stability of aggregates.
Tisdall and Oades (1982) reported that polysaccharides only act as transient binding agents which are rapidly decomposed by microorganisms and are predominantly associated with the > 0.25 mm transient, stable aggregates. Others (Adesodun et al., 2001; Insam, 1996; Piccolo, 1996; Spaccini et al., 2001), however, have recently shown that carbohydrates cannot be involved in long-term stabilization of soil aggregates because they are also decomposed by microorganisms in soils.
As a result particulate organic matter due to being more sensitive to change management, could be used as a favorable indicator for evaluation of paddy soil quality.

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Effects of Agricultural Waste Amendments on Organic Carbon Content and Cation Exchange Capacity of Soil

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Abstract

Soil organic carbon (SOC) content and cation exchange capacity (CEC) one of the most properties in determining of the soil fertility status. In this study, the effects of different agricultural waste amendments on SOC and CEC of sandy loam textured soil (Typic Xerofluvent) were investigated. Agricultural wastes, such as sugar beet pulp (SBP), apple pomace (AP) and cotton gin waste (CGW) was applied to soil as a fresh material, (dry weight basis 10, 20 and 40 t ha^{-1}), and pot experiments were carried out according to the completely randomized plot design with 5 replicates in greenhouse conditions. This study is consisted of two different stages. The first stage which is consists of 6 months incubation period (1^{st} sample period). Second stage which is consists of other 6 months plus 8 weeks bean (*Phaseolus vulgaris L.*) vegetation period (2^{nd} sample period).

The effects of waste amendments on CEC were significant between first and second stages. At the end of experiment, it is determined that SOC content and CEC of sandy loam textured soil improved by the amendment of SBP, AP and CGW linked to incubation periods.

Keywords: Apple pomace, Cotton gin waste, Fertility, Sugar beet pulp.

1. Introduction

Soil organic matter (SOM) and specifically soil organic carbon (SOC) are known to play important roles in the maintenance as well as improvement of many soil properties (Krull et al.2004). In particular, the suitability of soil for sustaining plant growth and biological activity is a function of physical and chemical properties, many of which are a function of SOM content (Doran and Safley, 1997).

Cation exchange capacity (CEC) is defined as the measure of the total capacity of a soil to hold exchangeable cations and indicates the negative charge present per unit mass of soil (Peverill et al., 1999). A high CEC is regarded as favorable as it contributes to the capacity of soils to retain plant nutrient cations. The use of organic soil amendments has been associated with desirable soil properties including higher plant available water holding capacity and CEC and lower bulk density, and can foster beneficial microorganisms (Doran 1995; Drinkwater et al. 1995). A decline of CEC with soil depth along with a decline in SOM which means that a 1% increase in SOC leads to 1 unit (cmol kg^{-1}) of increase in CEC in variable charge soils (Oades et al. 1989). The largest CEC were associated with the organo-mineral clay fraction, which the authors attributed to the type of SOM associated with minerals Thompson et al. (1989). In addition, functional groups of SOM have been associated with an increase in CEC (Oades et al., 1989). Accordingly, there is generally and McGrath et al. (1988) stated that a good correlation between SOC and CEC and the noted that CEC of a sandy soil increased from 75 to 158 cmol kg^{-1} as SOC increased from 0.46 to 1.39%. The contribution of SOM to CEC can vary between 25-90% (Stevenson, 1994), depending on soil type, but most studies observed a contribution between 30-60% (Tsutsuki, 1993; Loveland and Webb, 2003), 40-50%, respectively (Thompson et al., 1989; Haynes and Naidu, 1998). Eshetu et al. (2004) stated that in forest soils of the Philippines, there was a strong linear correlation between total CEC and SOC content. They also noted that SOC concentrations > 2% increased the CEC of surface soils by up to 4 times compared with mineral soil with SOC concentrations < 2%. Furthermore, at SOC contents < 2%, there was no measurable effect on CEC and they suggest that 2% could indicate a minimum threshold value.

The objective of this experiment is to determine the role of agricultural wastes such as sugar beet pulp, apple pomace and cotton gin waste on organic carbon content and cation exchange capacity of sandy loam textured soil.

2. Materials and Methods

2.1. Materials

Soil (Typic Xerofluvent) was collected from the 0–25 cm of as the name of Tehnelli series at the Land of Research and Practices of Akdeniz University in Turkey. Sugar Beet Pulp (SBP), Apple Pomace (AP) and Cotton Gin Waste (CGW) were obtained from sugar beet, apple and cotton processing company. Results of the analysis of initial (pre-amendment) soil properties and analytical composition of the organic materials are given in Tables 1 and 2, respectively.

Table 1 Selected physicochemical properties of soil.

pH (H_2O)	7,80
EC (dS m^{-1})	0,10
$CaCO_3$ (%)	48,69
Sand (%)	76,4
Silt (%)	4,6
Clay (%)	19,0
Texture	Sandy loam
Field capacity (%)	11,72
Wilting point (%)	4,67
Available water (%)	7,05
Bulk density (g cm^{-3})	1,67
CEC (cmol kg^{-1})	12,29
Organic matter (%)	0,92
Total N (%)	0,049
Available P (mg kg^{-1})	10,52
Exchangeable K (cmol kg^{-1})	0,140
Exchangeable Ca (cmol kg^{-1})	19,40
Exchangeable Mg (cmol kg^{-1})	1,44
Exchangeable Na (cmol kg^{-1})	0,16
Available Fe (mg kg^{-1})	9,21
Available Zn (mg kg^{-1})	1,70
Available Mn (mg kg^{-1})	7,11
Available Cu (mg kg^{-1})	1,10

Table 2 Analytical compositions of the organic materials.

	SBP	AP	CGW
Organic matter (%)	96,95	98,05	81,22
Ash (%)	3,05	1,95	18,78
Moisture (%)	483	436	7
Organic carbon (%)	56,36	57,0	47,21
C/N	39,88	84,82	14,23
pH (H_2O)	3,98	3,84	5,96
EC (dS m^{-1})	0,52	0,37	1,15
Total N (%)	1,413	0,672	3,317
P (%)	0,083	0,079	0,416
K (%)	0,364	0,696	1,371
Ca (%)	0,527	0,236	0,477
Mg (%)	0,323	0,065	0,298
Na (%)	0,119	0,035	0,033
Fe (mg kg^{-1})	481,9	171,53	380,9
Zn (mg kg^{-1})	14,2	10,4	40,8
Mn (mg kg^{-1})	61,5	7,2	25,4
Cu (mg kg^{-1})	8,3	10,7	10,1

2.2. Methods

This study is consisted of two different stages. The first stage consists of 6 months incubation period (1st sample period). Second stage consists of other 6 months plus 8 weeks bean (*Phaseolus vulgaris L.*) vegetation period (2nd sample period). End of the first and second stages, soil samples were collected and the effects of agricultural waste amendments on SOC and CEC determined.

The experiment was carried out in a completely randomized plot design with five replications under greenhouse conditions as a pot experiment. Soil samples were sieved with a 4–mm sieve after being air dried and then 10 kg soil were placed in each pot. On a dry weight basis, three levels of organic materials (10, 20 and 40 t ha^{-1}) were mixed as fresh materials to soil (Table 3) and then incubated for fourteen months. During the incubation period, soil moisture level in the pots approximately was maintained 70% of field capacity.

Table 3 Amendment rates of agricultural wastes in the experiment.

Amendments	Doses	Dry weight	Fresh weight
		t ha^{-1}	t ha^{-1}
Sugar Beet Pulp (SBP)	Control	0	0
	Dose1	10	58,3
	Dose2	20	116,6
	Dose3	40	233,2
Apple Pomace (AP)	Control	0	0
	Dose1	1000	53,6
	Dose2	2000	107,2
	Dose3	4000	214,4
Cotton Gin Waste (CGW)	Control	0	0
	Dose1	1000	10,7
	Dose2	2000	21,4
	Dose3	4000	42,8

The pipette method was used for texture analysis (Baver, 1966). Soil field capacity and wilting point was measured by using pressure membrane (Demiralay, 1993) and bulk density was measured using the cylinder method (Black, 1965). Soil pH was measured in a 1:2.5 (soil: water) aqueous extract (Jackson, 1967) and the soluble total salt was calculated in the saturation extract set out by Bower and Wilcox (1965). Organic matter was determined using the modified Walkey–Black method (Black, 1965) and SOC was calculated by divided the organic matter values with 1.72 (Nelson and Sommers, 1982). Calcium carbonate ($CaCO_3$) content of soil was measured with a Scheibler calcimeter (Çağlar, 1949). Exchangeable Na^+, K^+, Ca^{2+} and Mg^{2+} were determined by extraction with 1N ammonium acetate (CH_3COONH_4), and measured with an atomic absorption spectrophotometer (US Salinity Laboratory Staff, 1954). Cation exchange capacity (CEC) was measured using 1N ammonium acetate (CH_3COONH_4) method (Chapman and Pratt, 1961). Base saturation was calculated by exchangeable bases (Na^+, K^+, Ca^{2+} and Mg^{2+}) divided by total CEC value. Extractable P (with $NaHCO_3$) was determined using the Olsen method (Olsen and Sommers, 1982). Extractable Fe^{2+}, Zn^{2+}, Mn^{2+} and Cu^{2+} were determined using the DTPA extraction method and the atomic absorption spectrophotometer (Lindsay and Norvell, 1978).

Organic carbon was determined using the modified Walkey–Black method (Black, 1965) and the organic matter contents of the organic materials were calculated by multiplying the organic carbon values with 1.72 (Nelson and Sommers, 1982). The pH value of the organic material was measured in a 1:5 organic matter–water mixture (Anon. 1978). Nitrogen was determined using the modified Kjeldahl method (Kacar, 1972). The phosphorus content of the organic materials was determined using the phosphor vanado molibdo phosphoric yellow color in the filtrate method attained from nitric–perchloric acid mixture and the wet oxidation method (Kacar and Kovancı, 1982). The amounts of macro– and micro–elements (K^+, Ca^{2+}, Mg^{2+}, Fe^{2+}, Zn^{2+}, Mn^{2+} and Cu^{2+}) were measured using the atomic absorption spectrophotometer which was obtained in the filtrate after wet oxidation (Kacar, 1972).

One–way Analysis of Variance (ANOVA) was used to compare the effects of different sources of organic materials on soil bulk density. Variables for which significant treatment effects were found and were characterized further using LSD procedure mean at $P < 0.05$ (Anon., 1995).

3. Results and Discussion

The effect of Sugar Beet Pulp (SBP), Apple Pomace (AP) and Cotton Gin Waste (CGW) amendments on soil organic carbon (SOC) was detected after the incubation period (Table 4). The SBP increased the SOC ($P < 0.001$) compared to the control treatment in the both stage. The highest value was obtained from the SBP_3 (0.81%) in the first period. The highest value was obtained from the SBP_3, SBP_2 and SBP_1 (1.15, 1.07 and 1.05%) in the second period, respectively, but amendment rates of wastes were found to be similar. The SBP increased the SOC ($P < 0.001$) compared to the between first and second period. The mean SOC value was higher in the second period (1.05%) than first period (% 0.69).

Table 4 Effect of organic waste amendments on SOC content (%).[1]

Amendments	SOC	
	Period I	Period II
SBP_0	0,61**c**[2]	0,94**b**
SBP_1	0,66**b**	1,05**a**
SBP_2	0,67**b**	1,07**a**
SBP_3	0,81**a**	1,15**a**
Mean	0,69	1,05
$LSD_{Amendment.}$ (%5)[3]	***	***
$LSD_{Period.}$ (%5)	***	
AP_0	0,61**c**	0,94**c**
AP_1	0,63**b**	1,11**b**
AP_2	0,70**b**	1,12**b**
AP_3	0,82**a**	1,37**a**
Mean	0,69	1,14
$LSD_{Amendment.}$ (%5)	***	***
$LSD_{Period.}$ (%5)	***	
CGW_0	0,61**c**	0,94**c**
CGW_1	0,72**b**	1,10**b**
CGW_2	0,74**b**	1,18**ab**
CGW_3	0,93**a**	1,29**a**
Mean	0,75	1,13
$LSD_{Amendment.}$ (%5)	***	***
$LSD_{Period.}$ (%5)	***	

[1] Values of $n = 5$.
[2] The difference between values not shown with the same letter are significant at a $P < 0.05$ level.
[3] Significance,*** Significant at $P < 0.001$.

The effect of AP on SOC was significant ($P < 0.001$) and SOC increased with AP amendment in the both period (Table 4). The highest value was obtained from the AP_3 in the first period (0.82%) and second period (1.37%), respectively. The AP increased the SOC ($P < 0.001$) compared to the between first and second period. The mean SOC value was higher in the second period (1.14%) than first period (% 0.69). Similarly, the effect of CGW on SOC was significant ($P < 0.001$), and SOC increased with CGW amendment in the both period (Table 4). The highest value was obtained from the CGW_3 in the first period (0.93%) and second period (1.29%), respectively. The CGW increased the SOC compared to the between first and second period. The mean SOC value was higher in the second period (1.13%) than first period (% 0.75).

The effect of three agricultural waste amendments on soil CEC was not significant at the both period (Table 5). But, soil CEC value was increased ($P < 0.001$) by the agricultural waste amendments compared to between the first and second period. The mean CEC value was higher in the second period (15.42 cmol kg^{-1}) than first period (% 12.26 cmol kg^{-1}) with the SBP. The mean CEC value was higher in the second period (15.03 cmol kg^{-1}) than first period (12.06 cmol kg^{-1}) with the AP. The mean CEC value was higher in the second period (15.48 cmol kg^{-1}) than first period (12.08 cmol kg^{-1}) with the CGW.

Table 5 Effect of agricultural waste amendments on CEC of soil (cmol kg^{-1}).[1]

Amendments	CEC	
	Period I	Period II
SBP_0	12,27	14,77
SBP_1	11,82	15,98
SBP_2	12,27	15,64
SBP_3	12,69	15,30
Mean[2]	12,26	15,42
$LSD_{Amendment}$ (%5)[2]	ns	ns
LSD_{Period} (%5)	***	
AP_0	12,27	14,77
AP_1	12,48	15,19
AP_2	11,71	15,24
AP_3	11,81	14,95
Mean	12,06	15,03
$LSD_{Amendment}$ (%5)	ns	ns
LSD_{Period} (%5)	***	
CGW_0	12,27	14,77
CGW_1	12,13	14,83
CGW_2	11,44	15,97
CGW_3	12,52	16,36
Mean	12,08	15,48
$LSD_{Amendment}$ (%5)	ns	ns
LSD_{Period} (%5)	***	

[1] Values of $n = 5$.
[2] Significance: ns, not significant ***, Significant at $P < 0.001$.

As results, differences in CEC of soil were related to incubation period. Bulluck et al. (2002) have stated that mean total C and CEC were higher in plots with the cotton gin trash amendments compared to synthetic fertilizers after 2 years. Soil CEC value increased from 6.05 to 7.97 cmol kg^{-1}, and total C increased from 1.17 to 1.90% with the amendment. Most studies show a linear correlation between SOC and CEC. Loveland and Webb (2003) reported that CEC values for agricultural soils can range from 2-50 cmol kg^{-1} and values of around <4 cmol kg^{-1} are common in sandy soils. The effect of cotton gin trash, poultry manure, green manure and synthetic fertilizer amendment on soil CEC were investigated in 2001 and 2002. Levels of soil CEC was all significantly higher in soils amended with cotton gin trash than poultry manure, green manure or synthetic fertilizer. CEC value of soil increased from 3.63 to 6.58 meq 100 cm^{-3} in 2001, and 7.20 meq 100 cm^{-3} in 2002 (Liua et al. 2007). In another experiment, Surekha et al. (2003) have reported that soil CEC value increased from 22.6 cmol kg^{-1} to 25.1 cmol kg^{-1} after the amendment rice husk.

4. Conclusion

As regards the results of the statistical analysis of SOC, there were significant differences between the organic treatments and the control. All amendments increased SOC in the incubation period (especially second period), and OC content of soil has been reach to satisfactory level from very low C content level by the amendments. Differences in OC content of soil were related to amount of the organic material added. Increasing doses of agricultural waste amendment and incubation period had a significant effect on SOC in the experiment. According to the results of the CEC, there were not significant differences between the organic treatments and the control, but there were significant differences between incubation periods. Differences in CEC of soil were related to presence of more humified organic matter linked to incubation period and higher CEC of humic substances. Numerous studies have demonstrated that composts produced from a wide variety of organic materials can improve soil physical, chemical, and biological properties (Brosius, et al., 1998). Therefore, we recommended that used agricultural wastes in experiment may use as compost for the short duration of the waste amendment effects on SOC and CEC.

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Determination of Water-Yield Relationships of Cotton by Using Line - Source Sprinkler System

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Abstract

This research was carried out in order to determine the relationships between water and yield of cotton by the type of Nazilli-84 in 2002-2003. In this research, it was used a line-source sprinkler system and irrigated 4 times on the basis lack of soil moisture. It was used the decrease in the amount of water thrown the title away from the sprinkler at the creation of irrigation treatments. 100, 75, 50, 25 % of the deficient moisture in the soil were applied as irrigation water and also investigated without water treatments. The effect of amount of irrigation water on seed cotton yields was determined at the level of error 0,01 by the statistical analyses in both years. The highest yield were obtained when the missing of soil moisture was 75 %. At this issue, lint+seed cotton yield were obtained as 453 and 463 kg/da, the amount of irrigation water were obtained as 343 mm and 354 mm and the amount of water consumption were obtained as 492 mm and 456 mm in 2002 and 2003.

Keywords: Cotton, irrigation, yield, sprinkler, line source

Introduction

Nowadays, one of the most important issues faced by countries is the increase of demand on agricultural products in the line with population growth and depending on this, the increase of water requirement. While drinking and using water demand is increasing for urban and industrial sector, water resources used for agricultural purposes will decrease. In this case, the efficiency of water and land usage should be increased for higher production. To achieve this, irrigation methods have to turn optimum usage of water and soil resources. The way of using resources effectively is possible by to know current usage to determine bottlenecks and to solve them. Therefore it is necessary to use the methods, that would gain optimum benefit from irrigation water, which is very important input in agricultural production. There are several ways to provide the optimum benefit from irrigation water. These are methods and systems which make irrigation water to be used in a controlled manner. But, while these methods and systems are decreasing water loss or water consumption, shouldn't cause an essential decrease in yield. Today, it is known that sprinkler irrigation method is one of the methods which provide this condition. Giving water to plant root zone by this method provides minimum loss of evaporation and leakage and also low water tension in the root zone provides less plant stress.

Because amount of applied water in sprinkler irrigation is controlled, surface flow and losses decrease, water consumption and efficiency increase, the yield of product increases corresponding to the unit of water.

There are some researches about cotton irrigation in turkey and other countries. According to Yalcuk and Ozkara (1984), it's determined that the yield doesn't decrease with the deficit of 40% in irrigation which applied to witnessed treatment. Ozkara and Sahin (1993) determined that harvest delayed when amount of irrigation water increased. The maximum cotton lint+seed yields, in research done by fixed and mobile sprinkler system, are obtained from the treatments which pan evaporation is used as irrigation water by 100% and 75% (Sener, 1995). Kanber et al. (1996) researched different operating systems and performance of furrow irrigation in free draining and ponding furrows in Harran Plain condition. Kara(1998) has compared line and point-source sprinkler techniques, which used to obtain crop water production functions for the irrigation of second product soybean. Kara and Gunduz (1998) identified that the beginning of flowering and cocoon formation periods are the most sensitive development periods of water in their study, which they researched the deficit irrigation effects on cotton yield in Harran Plain condition. Gunduz and Korkmaz (2007) reported that the first irrigation date of cotton is in the first half of July, the last date of cotton irrigation is in the end of August and approximately cotton should be irrigated 17 days apart and four times in Menemen Plain.

This study was carried out to determine water-yield relationship of cotton by using the line-source sprinkler system.

Materials and Methods

The research was carried out at the land of UTAEM in 2002-2003. Menemen Plain is 33545 ha and its altitude is 10.3 m. Menemen Plain is dominated by Mediterranean climate, summers are hot and dry, winters are mild and rainy. According to the avarage climate data for many years, total annual rainfall were measured as 628-561 mm and the average relative humidity were measured as 69-57% in the years while the research carried out (2002 and 2003). The soil type of research area is loam and some physical, chemical properties of soil are given in Table 1. The electrical conductivity of irrigation water is 1.01 dS m^{-1} which was carried out with the cotton type of Nazilli-84 in the research.

Table 1. Some physical and chemical properties of soil

Soil depth (cm)	Texture	Field capacity (P_w%)	Wilting point (P_w%)	Volume weight (g cm^{-3})	EC (dS m^{-1})	$CaCO_3$ (%)	Organic matter (%)	pH
0-20	L	24.4	12.2	1.31	0.78	4.1	1.4	7.6
20-40	L	23.3	11.8	1.37	1.18	4.5	1.3	7.5
40-60	SiL	25.2	13.2	1.42	1.11	6.4	1.1	7.8
60-80	L	25.9	13.4	1.30	0.99	6.8	0.9	7.7
80-100	L	24.5	13.2	1.30	1.08	6.6	0.7	7.6

The arrangement of the line-sources sprinkler system is practiced on, which were specified the principles by Hanks et al. (1976), Watts et al. (1979) and Kanber et al. (1996). In this system the applied water content is reduced while the line is getting away from the source. It is called ‘‘line-source’’ due to the creation of linear source of irrigation water in the distribution. In this study, sprinkler heads were used which had 4.5 x 4.8 mm double nozzle, 2 atm pressure 1.96 m^3h^{-1} flow rate and 13 m throwing distance. Water collection containers were used, which were placed around the 2X2 m apart of the sprinkler heads and sprinkler testing was performed while the wind speeds were low. According to single sprinkler head test, it is determined that the water distribution was decreased linearly from sprinkler to the periphery on the conditions of 95% uniformity of water distribution and 2 atm operating pressure (R^2=0.965). ıt is obtained that irrigation levels was decreased while getting away from the line source. The determination of harvest area of the parcels was based on the irrigation levels. The general planning of the line source sprinkler system has shown in Figure 1. The water collection containers were placed in all parcels and were provided them at plant height in the irrigation times.

Treatments ;

I_1: Moisture will be enough to lift water field capacity at 0-100 cm depth

I_2: The Treatment of 75% of water applied as irrigation water given to treatment I_1

I_3: Treatment of 50% of water applied as irrigation water given to treatment I_1

I_4: Treatments of 25% of water applied as irrigation water given to treatment I_1

I_5: No irrigation

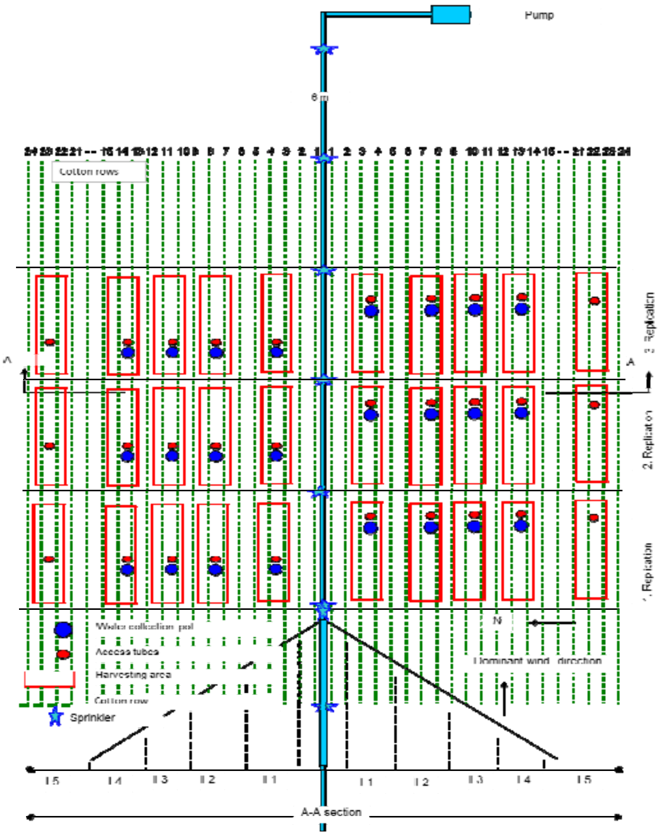

Figure 1. Details of line source sprinkler system (no scale)

Soil samples were taken at the depths of 0-20, 20-40, 40-60, 60-80 and 80-100 cm. And the physical analysis such as the field capacity, wilting point, bulk density, soil texture and the chemical analysis such as soil reaction, electrical conductivity, organic matter, phosphorus, potassium, were analyzed according to given methods. Irrigations were started in mid-July and approximately there were 4 irrigations 15 days apart. The irrigation water was applied to treatment I_1 as favorable moisture will be enough to lift water field capacity at 0-100 cm depth every irrigation (Gunduz and Korkmaz, 2009). Irrigation water which will be given other issues was identified by treatments. Irrigation was continued until the end of August. The amount of irrigation water and soil moisture values were measured in all parcels. The amount of irrigation water, which was identified on treatment I_1, has been fallowed from water collection containers during the irrigation periods. The amount of irrigation water, which were applied to other treatments, were measured from the water collection containers after each irrigation. Plant water consumptions were calculated by the water balance equation for the 0-100 cm of soil depth.

$$ET = I + P \pm \Delta SW - D_p - R_f$$

ET : Evaporation (mm)

I : Irrigation water (mm)

P : Precipitation rainfall (mm)

ΔSW : Soil-water content change (mm)

D_p : Deep percolation (mm)

R_f : Runoff (mm)

The research area was plowed in fall every year. After the necessary fertilizers were applied, when the soil conditions were suitable in spring, soil tillage was done by duplexing and was harrowed. The fertilizer grades were determined according to analysis of soil samples, which were taken in January. The sowing was done after the soil preparation in the spring as row spacing was 70 cm. in

May. Hand hoe and dilution are made after the completion of cotton outputs. Then hoe was done by hoe machine according to the plant development.

Trial treatment's accumulated water consumption was calculated according to the method of moisture reduction (Beyce and Madanoglu, 1978).

Duncan test and analysis of variance were applied to results of cotton yields and results of analysis (Yurtsever, 1984).

Results and Discussion

Amount of applied irrigation water, water consumption and obtained seed cotton yield are given in Table 2. Except treatment I_5, which was anhydrous treatment, in 2002 110-518 mm of irrigation water were applied and in 2003 119-494 mm of irrigation water were applied. Water consumption, depended on applied irrigation water, was 220 mm on treatment I_5 in 2002, 655 mm on treatment I_1. The seed cotton yields, which was obtained in this research were 253-453 kg/da in 2002 and 256-463 kg/da in 2003. The highest yield of cotton seed has been obtained from the treatment I_2 in both years.

The statistical analysis of the obtained seed cotton yield in both years of the research conducted was noticed at 0.01 error. 2 different groups were consisted on Duncan Test. Treatment I_1, I_2 and I_3 were in the first group and treatment I_4 and I_5 were in the second group (Table2,3).

Table 2. Seasonal irrigation water, water consumption, yield and Duncan groups

Treatment	Direction	2002			2003		
		Irrigation water, mm	Water cons., mm	Yield kg/da	Irrigation water, mm	Water cons., mm	Yield kg/da
I_1	N	517	650	416	491	581	443
	S	520	660	425	497	584	452
	Avr.	518	655	421 a	494	582	447 a
I_2	N	333	481	447	340	443	457
	S	354	503	459	367	468	469
	Avr.	343	492	453 a	354	456	463 a
I_3	N	209	362	414	236	339	425
	S	190	345	419	207	313	417
	Avr.	199	354	417 a	222	326	421 a
I_4	N	122	287	300	127	252	324
	S	97	261	315	111	235	291
	Avr.	110	274	308 b	119	243	308 b
I_5 – No irrigation	N	0	220	256	0	172	265
	S	0	220	249	0	172	247
	Avr.	0	220	253 b	0	172	256 b

Table 3. The results of the statistical analysis of seed cotton yield

SV	DOF	2002 lint+seed cotton yield		2003 lint+seed cotton yield		Table F	
		MS	F	MS	F	0.05	0.01
Repetition	2	445	2,82	1922	2,42	19,00	99,00
Direction	1	354	2,24	449	0,57	18,51	98,50
Water	4	43826	125,38 **	50517	107,20 **	3,01	4,77
Direction *Water	4	110	0,31	520	1,10	3,01	4,77
Error	16	350		471			
Total	29	6307		7502			

** : 0.01 error

The amount of applied irrigation water was decreased further from the source. There is a relationship between the applied irrigation water and the distance as y= -55,012 x + 602,94 and it's shown in figure 2.

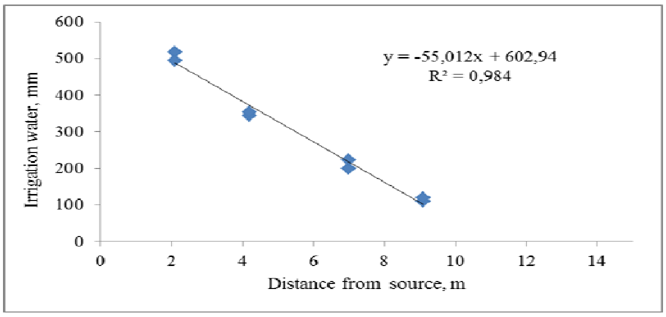

Figure 2. The relationship between the distance from the source and amount of irrigation water

The water consumption and the yields of the treatments were determined in the research's years. The seasonal yield response factor, ky= 0,79, was determined using the relationship between the treatments relative water consumption's decrease and relative yield's decrease, was calculated (Figure 3).

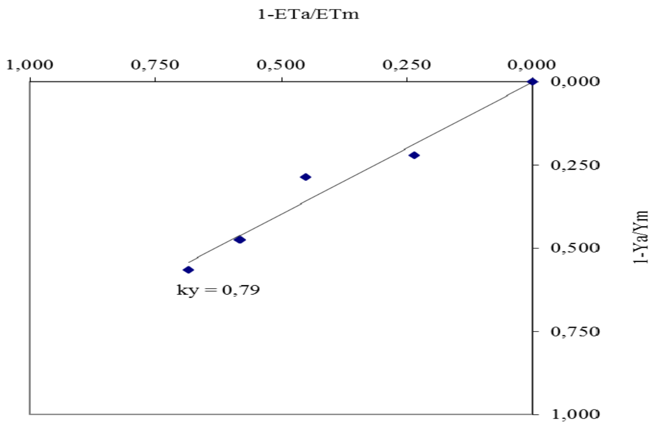

Figure 3. Relationship between relative decrease in water consumption and decrease in yield

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SOIL AND WATER POLLUTION

ORAL PRESENTATIONS

Assessment of nitrification in different soil texture affected by various concentration of pesticides

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Abstract
A laboratory incubation experiment was conducted to examine the effects of difference concentration of some common pesticides on nitrification and ammonia losses in different textured soil. In this study to determinate the rate of recovery NH_4^+, NO_3^- and NO_2^- 100g of air derided soil were placed in 250 screwed conical flask and 37mg $(NH_4)_2SO_4$ (100ppm NH_4) added, then treated with the solution of pesticides of (0,1,2,3L.donum^{-1},0,3,5,7 L.donum^{-1}and 0,05.2,0.5, 1.0 Kg.donum^{-1}) for herbicide (Fusillade supper), insecticide(Atrazin) and fungicide(Brasamid) respectively, about 10ml of each pesticides concentration were applied to sandy laom ,silty laom and slity clay textured soil. The experiment arranged in completely randomized design with three replication. The results indicate that the higher values of recovery NH_4, NO_2 NH_3 loses and lower NO_3 of 19.79 ,4.201,3.95 and 68.659 µ.g^{-1} were observed in silty clay texture soil .However, the result show that most of these pesticides partially decrease the nitrification particularly fungicides, when applied at the rate higher than recommended dosage.
Key words: Nitrification , Pesticides and Soil texture

Introduction
Pesticides are used in a number of human activates to be able to maintain production efficiency. There is a constant demand for a stable crop production in order to support the growing human population and the future usage of pesticides can therefore be expected to increase .Most agriculture are designed to effect only a specific target organism group or processes, but many of them have general toxic effects. The undesired effects that can arise as these substances are transported to surface and ground water. Pesticides are often spread in low concentration over along period time, some are relatively immobile thus deposit in soil .However ,shortly after application, pesticides will leave the application site in dissolved form, hydrophobic pesticides or pesticides with strong ion exchange to clay minerals might also enter the surface run off associated to suspended particles(Kreuger et al.,1999) .Soil microbial response to pesticides have been recommended as nearly warning indicator of ecosystem stress, because of the quick response to changes in environmental condition. One process of considerable ecological importance is nitrification. In most agricultural soils, ammonium (NH_4^+) from fertilizer is quickly converted to nitrate (NO_3^-) by the process of nitrification. This process is crucial to the efficiency of N fertilizers and their impact on the environment, because the net effect is a conversion of fertilizer N from a form that is not normally subject to loss from soil (NH_4^+) into a form that is readily lost by leaching or denitrification NO_3^-. (McInnes and Fillery, 1982., Jan etal.,2009). Nitrification occurs in two steps: NH_4^+ is first converted to nitrite (NO_2^-), and the NO_2^- is then converted to NO_3^-. Both reactions are carried out by bacteria present in the soil. Nitrifying bacteria are chemoautotrophic, in that they produce energy by chemical oxidation of NH_4^+ or NO_2^- and utilize CO_2 as a source of C. Different groups of bacteria are responsible for the two steps involved in nitrification. The NH_4^+-oxidizing bacteria include species from five genera, the most common being *Nitrosomonas*, the NO_2--oxidizing bacteria all belong to the genus, *Nitrobacter*. Because there are only a few species of nitrifying bacteria, nitrification is much more sensitive to environmental conditions than are most other N transformations, which are carried out by a more diverse group of microorganisms (Martikainen,1985). The effect of pesticides on nitrification and number of microorganism depending up on their concentration , rate an d period of application. A study was conducted by Luigi et al,(1996) to investigate the influence of the herbicides bentazon applied at 10 and 100 ppm on soil microbial community .Their results indicate that the higher dosages of bentazon markedly inhibited the nitrification. Investigation carried out by Sebiomo et al.,(2011) to study the effect of four herbicides (atrazine , primeextra, paraquat and glyphosate) onsoil microbial population, soil organic matter and dehydrogenase activity was assessed over a period of six weeks. They concluded that herbicides treatment resulted in asignificant drop in

dehtdrogenase activity when compared to control treatments A studies were initiated to comparison the effect of 10 mg .kg^{-1} soil of benlate ,captan, and lime sulfur fungicides with the nitrification inhibitors nitrapyrin and terrazole on oxidation of NH_4^+ in loamy sand soil, the result showed that the captan inhibited nitrification 21% more than lime-sulfur.(Somda et al., 1991). A laboratory experiments were conducted to study the effect of thimet ,bavistin and diuron on mineralization of urea in some soil of Karnataka , the results of these experiments indicate that the Thimet (phorate), bavistin(carbendazin), and diuron at 10-100ppm inhibited nitrification but no ammonification, the inhibition of nitrification increase with increasing pesticides concentration ,also same study show that nitrification rate was faster in black soils followed by laterite saline soil.(Sarawad,1987). In an incubation experiment revealed result that the nitrification process was inhibited by atrazine application and the inhibition increased wuth increases the concentration to150ppm and the nitrification rate was of the order neutral soil > acid soil>saline soil. Laboratory experiment was carried out to study the effect of some fungicides on leaf litter decomposition in microcosms ,their result refer to that the both fungicides benomyl and diazinon reduce nitrification at higher concentration of 110 and 400µ.g^{-1} for both fungicides respectively(Vink et al., 1999). The rate of nitrification increases with soil temperature up to about 35°C ; below 5°C very little NO_3^- is formed. Soil pH is also important. Below a pH of 6.0, nitrification is inhibited by acidity, and the process virtually ceases at a pH of 4.5 to 5.0. Under alkaline conditions, production of NO_3^- is markedly enhanced. The optimum pH is normally between 7.0 and 8.0(Kinsbursky and Saltzman, 1990; Ibrahim et al., 1995), this range of pH is close to Iraqi Kurdistan soil range, although nitrification in agricultural soils has been studied, most of the studies have been conducted to investigate the effect of some environmental factors such as temperature, relative humidity and soil moisture content, but a few study carried out to examine the effect of pesticides on nitrification in soil of Kurdistan region. Thus, the objectives of this study were: (i) to evaluate the rate of NO_3^- and NO_2^- product from different soil through nitrification; and (ii) to determine whether the effect of different concentration of some common pesticides on nitrification.

Materials and Methods

The soils used were surface (0-30 cm) samples originated from aski-kalak, fields trail of grdarasha and center of agriculture research in Erbil. The samples were sieved 2mm screen, Then analysis to determination some physical, chemical, and biological properties according to methods described by Black (1965) and Allen (1974) Table (1). To determinate the rate of recovery NH_4^+, NO_3^- and NO_2^- 100g of air derided soil were placed in 250 screwed conical flask and 37mg $(NH_4)_2SO_4$ (100ppm NH_4) added ,then treated with the solution of pesticides of (0,1,2,3L.$donum^{-1}$,0,3,5,7 L.$donum^{-1}$and 0,05.2,0.5, 1.0 Kg.$donum^{-1}$) for herbicide (Fusillade supper), insecticide(Atrazin) and fungicide(Brasamid) respectively, about 10ml of each pesticides concentration. Additional water was applied to achieve a soil moisture content equivalent to 60% of the water-holding capacity, 5ml of boric acid indicator solution added to test tube , the test tube putted inside the conical flask and the conical flask was capped and placed in an incubator maintained at 25°C. Measurement of ammonia volatilization loss was carried out daily according to method described by Black et al.,(1985). The ammonia evolved was trapped in boric acid (the color change from red to green) and determined by titration with standard H_2SO_4 0.01N. After 30 days triplicate flask were removed from the incubator, 100 ml of 2 M KCl adding to each flask. The KCl Soil solution was extracted by shaking for 1 h and filtering the resulting suspension under vacuum. The soil extract was analyzed for NH_4^+-N by indophenol blue method (Allen, 1974),while NO_3^- determined by UV spectrophotometer method as described by Richard et al.,(1985),where as NO_2^- estimated by colorimetric procedures (Mulvaney and Bremner, 1979 ; Keeney and Nelson, 1982).

Table (1) some physical, chemical and biological properties of soils under study

Soil proprties		**Iski-Kalak soil**	**Grdarash soil**	**Erbil agriculture research center soil**
PSD $g.Kg^{-1}$	Sand	700	187	51
	Silt	230	646	590
	Caly	70	167	409
Textural name		Sandy Loam	Silty Loam	Silty Clay
pH		7.55	7.73	7.90
ECe dSm^{-1}		0.22	0.54	0.63
Nitare $mg.kg^{-1}$		3.2	2.5	4.3
Ammonia $mg.kg^{-1}$		0.43	1.1	1.8
Organic matter $g.kg^{-1}$		5.4	8.11	11.3
Available phosphorus $mg.kg^{-1}$		1.45	3.2	4.0
Total count of bacteria g^{-1} soil		$102*10^4$	$146*10^5$	$189*10^5$
Total count of fungi g^{-1}soil		$25*10^3$	$62*10^3$	$88*10^3$

Statistical analysis

The experiment was designed in completely randomized design with three replications, factorial CRD. The experimental data were analyzed by using SPSS program and differences between the treatment means were separated by LSD test. (Steel and Torrie, 1969).

Results and Discussion

In the nitrogen cycle of the soil, nitrate has a central position since it is the most bioavailable nitrogen for plants. Nitrification is attest suitable for assessing the side effect of pesticides on soil non-target organisms because of its sensitivity and the agronomic significance of this process. The data in table (2) show the nitrification rate decreased from light soil texture to heavy soil in texture the higher value of accumulated nitrate was found in sandy loam texture compared to other soil texture, the reason may be due to the fact that the sands increase the size of space between particles, thus facilitating the movement of air and drained water, it means that sandy loam soil supply adequate oxygen to meet the requirements of nitrifies microorganisms and stimulate. As well as the result in the same table indicated that the volatilized ammonia is higher in silty clay soil compared to other soil texture which attained 3.95 $\mu\ g^{-1}$, this may be due to the effect of soil pH (7.9) because all study refer to present the positive relationship between ammonia volatilization and soil pH. These results agree with those reported by Rolecke et al., (1996). They reported that the ammonia volatilization increased with increasing g the soil pH. The data obtained with the pesticides and their concentration are reported in table(2) .They show that most of these pesticides partially decrease the nitrification particularly fungicides, when applied at the rate higher than recommended dosage , the drop of nitrification at higher concentration of pesticides in the present study , however , could be the result of a toxic response or it is possible that application of fungicides may result in more disturbed microbial equilibrium than that in herbicides and insecticides application .

Table (2): Effect of soil texture, pesticides and their concentration on the ammonium, volatilized ammonia, Nitrate and Nitrite.

Treatments	NH_4 μ g^{-1}	NH_3 μ g^{-1}	NO_2 μ g^{-1}	NO_3 μ g^{-1}
S_1	15.85	3.170	3.420	73.867
S_2	15.52	3.100	3.355	74.301
S_3	19.76	3.950	4.201	68.659
LSD(0.01)	1.35	0.17	0.21	1.14
P_1	13.47	2.695	2.945	77.034
P_2	14.65	2.929	3.179	75.472
P_3	23.01	4.602	4.852	64.320
LSD(0.01)	1.35	0.17	0.21	1.14
C_0	5.17	1.033	1.283	88.111
C_1	17.06	3.413	3.663	72.248
C_2	18.94	3.788	4.038	69.748
C_3	27.00	5.401	5.651	58.995
LSD(0.01)	1.56	0.20	0.34	1.31

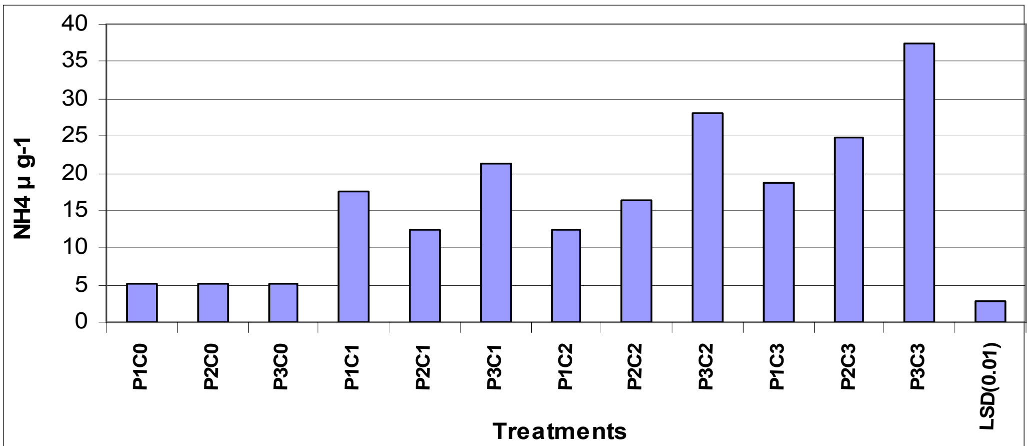
S_1=Sandy loam, S_2= Silty loam, S_3= Silty clay
P_1=Herbicides(L.donum^{-1}), P_2=Insecticides(L.donum^{-1}), P_3=Fungicides (Kg.donum^{-1})

Pesticides		Herbicides	Insecticides	Fungicides
C=Concentration	$C_{0=}$	0	0	0
	$C_{1=}$	1	3	0.25
	$C_{2=}$	2	5	0.50
	$C_{3=}$	3	7	1.00

The data in table (3) and figure (1) revealed that the a pesticides and their concentration have a significant effect on nitrification in different soil texture. Addition of pesticides in higher concentration particularly fungicide to the soils resulted in a lower increase in nitrate production compared to the same soil when pesticides was not added or added in low concentration and recommended rate. The decrease in the ammonium concentration in the presence of the pesticides resulted in large part from volatilization and fixation due to soil reaction and other soil properties. The higher value of recovery NH_4 , NH_3 and NO_2 was recorded from silty loam soil which received 1.0kg.Donum^{-1} of fungicides, it was attained (43.47, 8.60 and 8.50 μ g^{-1}),while the higher value of NO_3 (91.44 μ g^{-1}) was produced in silty clay soil which is not treated by pesticides. This result could be explained on the ground that the heavy soil due to its increase area of surface exposed to solution their particles have the capacity to hold pesticides and other compound also microorganism attract to these area, by this way the present of pesticides in higher concentration may decreases the activity and number of microorganisms in soil , however , heavy soil not only have the capacity to attract and hold nutrients and compounds on their surface , but the also hold much more water may causes too much water holding capacity and not enough aeration . Moreover, it is appear from figure (1) the pesticides with higher concentration significantly decrease the nitrification particularly the fungicides , the inhibitory effect of pesticides may be due to their toxic effect , as well as the results in the same figures suggested that the rate of nitrification decrease with increasing the pesticides concentration , this result may be due to that the higher concentration contributed in disrupted the microorganisms metabolism , then decrease their growth and number.

Table(3):Combination effect of soil texture ,pesticides and their concentration on the concentration of ammonium, volatilized ammonia, Nitrate and Nitrite .

Treatments	NH_4 μ g^{-1}	NH_3 μ g^{-1}	NO_2 μ g^{-1}	NO_3 μ g^{-1}
$S_1P_1C_0$	5.37	1.07	1.32	87.84
$S_3P_1C_0$	2.67	0.53	0.78	91.44
$S_1P_2C_0$	5.37	1.07	1.32	87.84
$S_2P_2C_0$	7.47	1.49	1.74	85.04
$S_3P_2C_0$	2.67	0.53	0.78	91.44
$S_1P_3C_0$	5.37	1.07	1.32	87.84
$S_2P_3C_0$	7.47	1.49	1.74	85.04
$S_3P_3C_0$	2.67	0.53	0.78	91.44
$S_1P_1C_1$	11.03	2.21	2.46	80.29
$S_2P_1C_1$	11.37	2.27	2.52	79.84
$S_3P_1C_1$	30.30	6.06	6.31	54.60
$S_1P_2C_1$	17.13	3.43	3.68	72.16
$S_2P_2C_1$	8.62	1.72	1.97	83.51
$S_3P_2C_1$	11.27	2.25	2.50	79.98
$S_1P_3C_1$	16.53	3.31	3.56	72.96
$S_2P_3C_1$	16.18	3.24	3.49	73.42
$S_3P_3C_1$	31.13	6.23	6.48	53.49
$S_1P_1C_2$	14.10	2.82	3.07	76.20
$S_2P_1C_2$	9.25	1.85	2.10	82.67
$S_3P_1C_2$	13.73	2.75	3.00	76.69
$S_1P_2C_2$	20.40	4.08	4.33	67.80
$S_2P_2C_2$	11.53	2.31	2.56	79.62
$S_3P_2C_2$	17.13	3.43	3.68	72.16
$S_1P_3C_2$	21.97	4.39	4.64	65.71
$S_2P_3C_2$	26.00	5.20	5.45	60.33
$S_3P_3C_2$	36.33	7.27	7.52	46.56
$S_1P_1C_3$	23.67	4.73	4.98	63.44
$S_2P_1C_3$	11.57	2.31	2.56	79.58
$S_3P_1C_3$	21.17	4.23	4.48	66.78
$S_1P_2C_3$	24.93	4.99	5.24	61.76
$S_2P_2C_3$	25.90	5.18	5.43	60.47
$S_3P_2C_3$	23.33	4.67	4.92	63.89
$S_1P_3C_3$	24.33	4.87	5.12	62.56
$S_2P_3C_3$	43.47	8.60	8.50	37.04
L.S.D(0.01)	4.68	0.59	0.65	3.94



Fig(1) Effect of different concentration of some pesticides on recovery NH_4 µ g-1

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Effect of Irrigation with Sewage Sludge on Heavy Metals Movement in the Soil Profiles

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Abstract
The application of sewage sludge on agricultural soils has been widespread in many countries around the world**.** Although sewage sludge contains valuable nutrients, it also contains various toxins, especially heavy metals that can transfer shallow groundwater systems through leaching. The purpose of this study is to assess possible contamination of the Tehran shallow groundwater system with heavy metals under through plot study. For this purpose, four experimental plots were filled with collected soil from Varamin plain. The plots were irrigated with four different kind regimes. After irrigation, the soil samples were taken at 5 cm intervals up to 60 cm from each plot and Cd, Ni, and Pb concentrations in each sample were measured by atomic absorption spectrophotometer**.** Results showed that the metals accumulated in surface layer of the soils or they had low vertical movement that this property is related to their high sorption capacity by soil. The accumulation of pollutants within the first 18 cm of the 4 plots showed that accumulation percentages for different plots of soil were 66 to 90%. Therefore, pollution of this area groundwater by these metals is unlikely.
Keywords: Heavy metals; Sewage sludge; Irrigation; Movement; Soil profiles

Introduction

Increasing urbanization and industrialization have resulted in a drastic increase in the volume of wastewater produced worldwide. One way of sludge disposal is its application on land. The application of sewage sludge on agricultural soils has been widespread in many countries around the world also in Iran, and this has been shown to improve soil properties such as soil organic matter, nutrients, porosity, aggregate stability, bulk density, and water retention and as a result, to increase plant productivity. In the European community, over 30% of sewage sludge is used as fertilizer in agriculture. For example in UK, Gove et al. (2001, 2002) reported increasing use of enhanced treated sewage sludge in agriculture. However, although sewage sludge contains valuable nutrients, it also contains various toxins, especially heavy metals that have excessive use in industrial applications. Heavy metals are very harmful because of their non-biodegradable nature, long biological half-lives and their potential to accumulate in different body parts. Even low concentrations of heavy metals are toxic because there is no good mechanism for their elimination from the body. Excessive accumulation of heavy metals in agricultural soils through wastewater irrigation, may not only result in soil contamination, but also lead to elevated heavy metal uptake by crops, and thus affect food quality and safety. In addition, there is also the possibility of transfer of these metals into environmental media, most especially shallow groundwater systems through leaching. Metal transfer from sewage sludge to soil and subsequently to groundwater represents one of the most critical long-term hazards associated with the application of these wastes to soils (McBride et al., 1997). However, research carried out on the potential migration of heavy metals in sewage sludge-amended soils shows controversial results. The authors of some studies showed that heavy metals migration in soils is practically insignificant as metals remain at the site of input (Welch and Lund, 1987; Dowdy et al., 1991). This is consistent with the poorly soluble nature of heavy metals in most soils, especially basic soils (McLean and Bledsoe, 1992), in which the metals tend to accumulate at the soil surface and become part of the soil matrix. However, several fiel trials and column studies (McBride et al., 1999; Ashworth and Alloway, 2004) have demonstrated migration of metals at depth, and have highlighted the concern regarding groundwater contamination by these pollutants.Large lands in south of Tehran city (approximately 6900 ha), are irrigated with treated and untreated wastewater, and with attention to high water table in these areas, there is a need to assess the possible environmental impacts of such application, from viewpoint of pollution of this area groundwater, that have increasing concern, both in agricultural and environmental studies. Based on the above background, the focus of this study is to assess possible contamination of the Tehran shallow groundwater system with heavy metals under through plot study with 4 different irrigation regimes. Three heavy metals were selected in this study: Cd, Pb, and Ni.

Materials and methods

Soil sampling

Composite soil sample were collected from upper horizon of Varamin plain in south of Tehran (51° 38′ to 51° 48′ N, 35° 14′ to 35° 11′S). The samples were air dried and passed through a 2mm sieve. Main soil properties such as pH, electrical conductivity (EC), cation exchange capacity (CEC), $CaCO_3$ equivalent, gypsum, organic matter, saturated moisture, and texture were measured in lab (Black, 1965 and Page, 1965).

Plots preparation

Four experimental plots (2 m * 6 m) were filled with collected soil. The first plot was irrigated with effluent from Shoush wastewater treatment plant, the second one with sludge and pipe water, the third plot with simulated wastewater with heavy metals (the concentration of each element in solution was 10 times concentration of heavy metals in drainages of south of Tehran and equal to several years irrigation) and the fourth one with sewage sludge and effluent. The sludge was incorporated into 0-20 cm plow layer before irrigation. After irrigation, the soil samples were taken at 5 cm intervals up to 60 cm from each plot. Then Cd, Ni, and Pb concentrations in each taken sample were measured by atomic absorption spectrophotometer (Varian model 200) after the samples were digested and extracted in HNO_3 4N solution.

Results and Discussion

Results of physico-chemical analyses of soil sample showed that texture of soil was loamy; pH, CEC, PO_4^{--} , total nitrogen, calcium carbonate, organic matter, moisture, and porosity were 7.76, 36.62 cmol/Kg, 20 mg/Kg, 1.4 mg/Kg, 12.25%, 36%, 35.8%, and 0.52% respectively.
The distribution in depth of 4 pollutant elements (Ni, Pb, Cr, and Cd) in plots is presented in Fig. 1. This Fig. shows that:

1. Except plot 1 (irrigation with effluent of treatment plant) in other plots, the highest concentrations were consistently within the first 17 cm, i.e. they had low vertical movement.
2. In plot 1, the highest concentrations was within 5 cm.
3. Below a certain depth (20 cm for plots 1, 2, and 3; and 10 cm for plot 1), the concentration in pollutant elements reached values similar to those of uncontaminated soils.
4. Nevertheless, the studied elements, showed small but significant accumulations at 20 to 30 cm (except plot 1).

Lower migration of these pollutants in plot 1 in comparison with other plots, is because of their low concentration in effluent of Shoush plant, that is used for irrigation of plot 1.

Ni

Pb

Cd

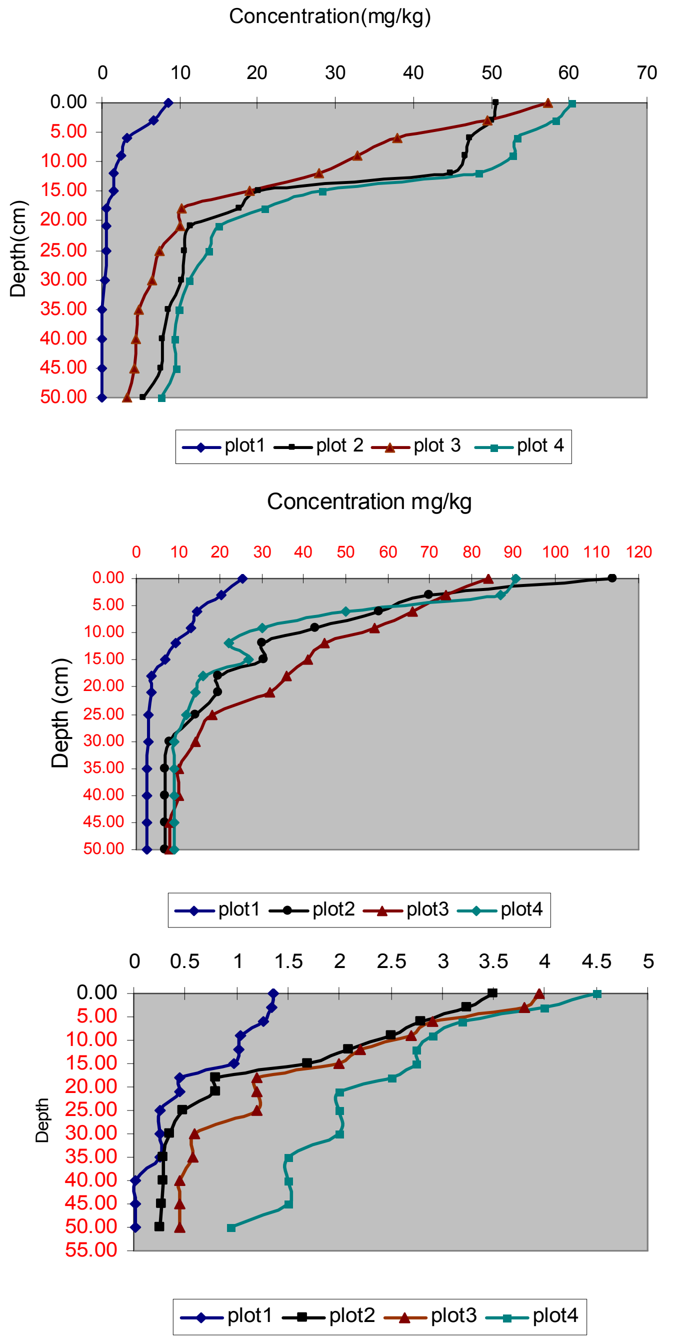

Fig. 1: Depth distribution (mean values) of Ni, Pb, and Cd in the soil profiles

5. Below a certain depth (20 cm for plots 1, 2, and 3; and 10 cm for plot 1), the concentration in pollutant elements reached values similar to those of uncontaminated soils.

6. Nevertheless, the studied elements, showed small but significant accumulations at 20 to 30 cm (except plot 1).

Lower migration of these pollutants in plot 1 in comparison with other plots, is because of their low concentration in effluent of Shoush plant, that is used for irrigation of plot 1.

This accumulation of the metals in surface layer of the soils, is related to their high sorption capacity by soil which is a result of chemical reaction between soil solid phases including silicate clays, oxides and hydroxides of metals especially Fe and Al, amorphous minerals and also lime and organic matter and firm bounds with these compounds (Corey et al., 1987 and Emmerich et. al., 1982). This property accompanying with high rate adsorption of these elements by soil solid phases, causes their retention in surface layer of the soil. Migration of our studied elements in soil, is a product of 4 parameters including soil sorption capacity, reaction rate of these elements with solid phase, water movement rate in soil and their primary concentration. Combined effect of these

4 parameters, determine distribution of these metals in soil profiles and depth of movement. These results are consistent with previous observations. For example, Plant and Raiswell (1983) indicated that under neutral to basic conditions, relative mobility is considered low or very low for all of the elements. However, movement of these elements is observed in some soils. Soil acidity, light texture, and structural features such as soil cracks, were important factors in this case (Alloway, 1990). Also, migration of these elements is observed in soils under lagoons, an area that huge volume of sludges is disposed. Especially if wastewater contains high chelating agents, these agents can form soluble complexes with Cd, Pb, and Ni, thereby causing more movement of them in soil profile.

In Fig. 2, the accumulation of Cd, Pb, and Ni within the first 18 cm of the 4 plots is compared. This Fig. shows that Ni had the greatest accumulation and the least vertical movement in plot 4. This Fig. shows that accumulation percentages for different plots of soil were 66 to 90% for different plots and elements. According to results of this study (low movement of Cd, Pb, and Ni in the plots and their accumulation in the 18 cm soil depth), it can be concluded that once soil is polluted by treatment plant effluent or sewage sludge containing Pb, Cd, and Ni in south of Tehran city soils, these 3 elements tend to accumulating in topsoil quickly and not easily desorbing from the soil, so they will move downward to groundwater very slowly. Soil acidity, light texture, and structural features such as soil cracks, were important factors in this case (Alloway, 1990). Also, migration of these elements is observed in soils under lagoons, an area that huge volume of sludges is disposed. Especially if wastewater contains high chelating agents, these agents can form soluble complexes with Cd, Pb, and Ni, thereby causing more movement of them in soil profile.

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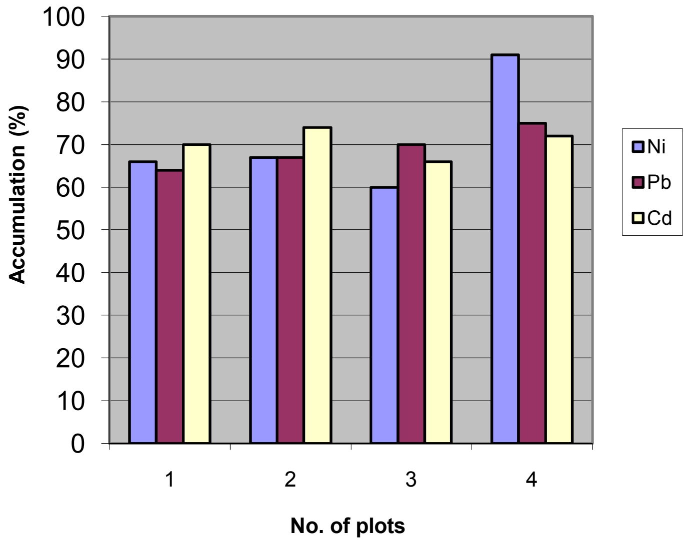

Fig. 2: Ni, Pb, and Cd accumulation percentages in the first 18 cm of the soil

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Chenopodium album can remediate contaminated soils.

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Abstract
Soil contamination with heavy metals in many parts of the world is expanding, and it has so many hazardous to human healths. Phosphate fertilizers which use in agriculture of Iran have cadmium contamination. One way to improve contaminated soil is using plants to remediate it. The objective of this study was to eliminate the potential of a high biomass halophyte weed *chenopodium album* L. to extract cadmium from soil to the plant. This plant is native to Middle East and Mediterranean country. A randomized complete block experiment in four replications was designed. Soil was contaminated by 5, 10, 20,40, 60 and 100 $mgkg^{-1}$ by cadmium (cadmium chloride).The results showed that at 100 mg kg^{-1} of soil cadmium contamination, the Cd concentration in shoot was 40 mg kg^{-1}.The high biomass production in high Cd-treatments indicates that the Cd-removal from soil which is a major factor for phytoextraction technology, was high (184 gr ha^{-1} at 60 mg kg^{-1} soil Cd-content in one harvest). High Cd concentration and high biomass in this plant shows that *chenopodium album* L. have a high potential to remove Cd from contaminated soils.
Keywords: Cadmium, Contaminated soils, Phosphate fertilizers, Phytoextraction , *chenopodium album l.*

Introduction
From a global perspective, the climate, the soil is considered as the third component of the human environment. Soil has two major functions: Supply of plant growth and other life forms and the other is the repository for the waste of nature. This feature enables the soil to have considerable ability to deal with external factors. The earth is always in search of a balance it can be concluded, when the soil is contaminated this confrontation and conflict can be impaired or goes out of its natural norm (baibordi, 2001). Increasing human activity on earth has caused the soil to earth in case of continuous partial of pervasive be impaired. Obviously, soil pollution is an undesirable phenomenon. Refinement of heavy metals from the soils that minimal losses to the environment and then are of your soil in to a category can be verified (Mazhari, 2004). One way of improving soil contaminated with heavy metals is phytoremediation. Phytoremediation means the use of herbal plants to remove pollutants from the environment or reduce their risk (Cunningham and et al 1995). phytoremediation can be viewed as a Green Revolution in the field of new technologies in refining the expression. Various forms of inorganic and organic compounds from phytoremediation that can be covered there. This is one of the types of phytoextraction. Derived primarily from Phytoextraction to treat contaminated soil and removal of contaminants from the soil are(Henry,J.R.2000). In this way the plant uptake, concentration, and sequestration of soil contaminated with oxic metals in the above ground plant biomass (stems, leaves, etc.) Is used (Nacimento, 2006).
chenopodium album L is a plant of the family (Chenopodiaceae) and a year and with stems and branches of the static, often with pink lines and its height varies according to climate and soil type. This plant is a plant species like salt.
In this research effort is uptake and phytoremediation of cadmium element is evaluated using a*chenopodium album* L.

Material and methods
Composite samples were taken from a depth of 0-30 cm. The samples were air dried and then were passed through 2mm sieve. Physical and chemical testing of soil samples containing, characterized by the relative frequency of particles (clay, silt, sand) to the hydrometer method, electrical conductivity of saturated soil extract (EC), with the gauge, saturated soil's acidity (PH) c. Using the device. PH meter, methods of soil organic matter (Walkely and Black) were measured glomerate and the apparent specific gravity method and paraffin and cadmium in soil using DTPA and field capacity moisture content (θFc) by the suctioned vice 33 pages Kp pressure measurements were obtained. The bulk density of soil; layouts for each pot 1/3 $gr/cm^3$was calculated. The allowable

concentration of cadmium concentration was chosen as the concentration range from zero to several times the concentration allowed to cover. Therefore, concentrations, 5,20,40,60,100 mg / kg cadmium in the soil was for contaminated soil from ($CdCl_2.H_2O$) was used to spray the soil and thoroughly mixed with it soils then were left to Pollution and contamination of soil and the interaction is more natural.

Approximately three months after the final growth took samples of soil and plant pots were. Crops were harvested from crown and separate the roots by placing the pot in a tub full of water were collected. The samples were dried at 70 ° C for 48 hours and then dried using an electric mill with steel grinding chamber were all. Two grams of soil weight and Meyer Erlenmeyer flask (conical flask) 120 ml and poured into 20 ml DTPA solution added and two hours of the rotational model with Shaker (IKA Werk KS580) with 250-degree flat 42 What man filter paper and then we move. Cadmium levels in soil were measured with atomic absorption Specter AA.200 model. For total extractable Cd, more plant oxidation method was used. For this study, a factorial design in randomized complete block with four replications was used for data analysis, statistical software SPSS (version 16) was used.

Results and discussion

The soil was sandy clay loam (sand 50%, silt 26% and clay24%) in texture with initial pH 7. 8 (1:2 soil: water), electrical conductivity (EC) 6.71 ds m−1 (1:2 soil: water), organic carbon 0.7%, calcium carbonate 7.5% and initial Cd 1.23 mg kg−1 (Table 1).

Table 1.Selected physical and chemical properties of the experimental soil and water (Mean ± SD, n=4)

Soil Parameters	Unit	
Soil texture		sandy clay loam
pH(1:2)		7.8±0.38
Bulk density	(gr/cm3)	1.334± 0.1
EC(Soil)	(dS/m)	6.71±0.68
Initial Cd (soil)	mg kg−1	1.23±0.011
N	%	0.07±0.002
CaCO3	%	7.5±1.06
OM	%	0.7±0.02
Plant biomass(Shoot	Kg ha-1	4380±640
Applied water Parameters	Unit	
EC	(dS/m)	1.2±0.02
Initial Cd	mg kg−1	0.013±0.00

Cadmium uptake by the *chenopodium album* L. is shown in Figure 1.This chart gives the amount of cadmium uptake by soil cadmium concentration of 100mg/kg is 140gr/ha shoot.

In a similar study that Jan Mertens and colleagues (2007) on the phytoextrction of cadmium accumulation with hyper accumulator plants were similar to the results achieved. The results of the experiment and the amount of cadmium accumulated in the shoot by increasing plant biomass was increased. .The measurements showed that in the third harvest plant significantly reduced the amount of cadmium in soil.

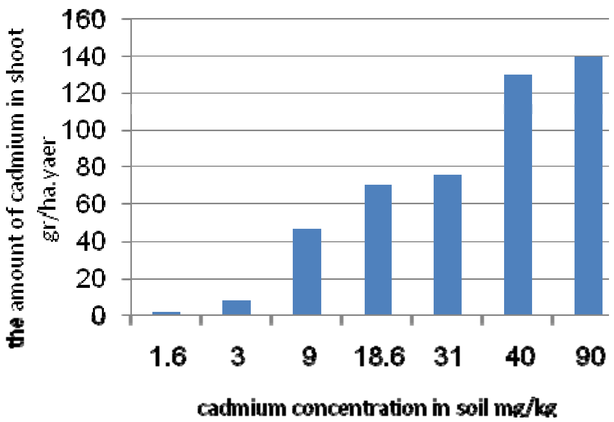

Figure 1.amount of cadmium uptake with chenopodium album

Cadmium changes in *chenopodium album* shoot's are given in Figure 2 which tells the amount of cadmium accumulated in the soil cadmium concentrations in crops 90 mg/kg to 40mg/kg reaches its maximum.

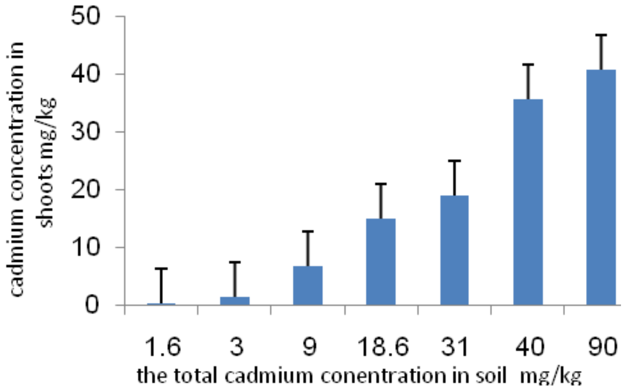

Figure 2. Cadmium changes in chenopodium album's shoot

Also according to the results based on dry weight equivalent weight 600 gr/m^2 cadmium levels in soil is close to zero. The equivalent weight 370 gr/m^2 cadmium levels in soils is 90 mg/kg. (Figure 3).

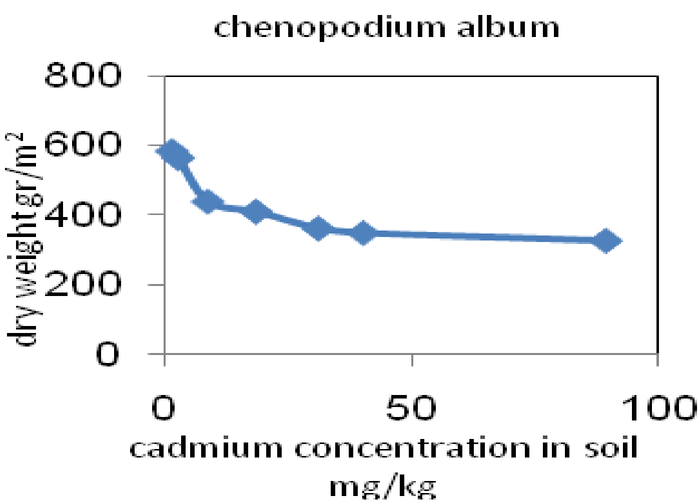

Figure 3. Amount of cadmium based on dry weight

Conclusions

Over all the results of experiments and statistical analysis shows that phytoextraction with hyperaccumulator plants has great potential for improving soils contaminated with heavy metals like cadmium and *chenopodium album* is a hyperaccumulator plants and has the ability to absorb cadmium from contaminated soil and High concentrations of cadmium contamination in soil and uptake by this plant increases. Cadmium accumulated in the measurement of crops this is evidenced and can be refined from plant to soil contaminated with heavy metals including cadmium used.

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High Biomass *Chenopodium album L.* is a Suitable Weed for Remediation of Lead-Contaminated Soils

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Abstract
Phytoremediation is a modern and useful method used to remediate the contaminated soil by heavy metals such as lead. This technology has the lowest cost and greatest effect. To carry out usefulness of Phytoremediation, choosing an appropriate plant is an essential step. The objective of this study was to find out if the high biomass producing halophyte *chenopodium album* L. can be used for phytoremediation of lead from contaminated soils. Accordingly, an extensive test was carried out to assess the phytoremediation capability of this species. The soils used in the experiment were contaminated with 150, 300, 600, 900 and 1200 mg Pb on kg^{-1} soil, in the form of $PbNO_3$. The results indicated that at the highest level of lead contamination in the soil, the concentration of lead in the root and shoot were 128.93 and 9.021 ppm, respectively. The lead uptake increased with Pb contents in soil and the maximum accumulation occurred in the roots. The average Pb accumulation in shoots was not significant. Its high biomass production and high Pb concentration of lead in the plant, justifying that this plant can be a potential candidate for pb phytoremediation from contaminated soil.
Keywords: lead, contaminated soils, heavy metals, *chenopodium album* L.

Introduction
The toxicity of heavy metals is a problem for ecological, evolutionary and environmental reasons (Nagajyoti et al., 2008). Lead is one of the heaviest elements that cause many of the environmental pollutions. It has extreme toxicity effects on humans and other living organisms, and then provides its considerable role in environmental pollutions. Using Pb in various industries has increased its pollution rate in different ecosystems. These toxic substances are released into the environment and contribute to a variety of toxic effects on living organisms by food chain (Chhotu et al, 2009). It indirectly contaminated soil and produce toxicity in agricultural products consumers directly (Nagajyoti et al., 2008).
Phytoremediation, the use of vascular plants to remove pollutants from the environment or to render them harmless (Cunningham,1993) has emerged as an alternative technique for removing toxic metals from soil and offers the benefits of being in situ, cost-effective and environmentally sustainable. In recent years, great interest has been developed for the identification of autochthonous plant species, which can accumulate elevated amounts of heavy metals in their tissues, with the aim of employing them for phytoremediation of contaminated soils (Garcıa, 2003). These plant species have special mechanisms to cope with higher levels of metals in growth medium.
Some plants have considerable potential in uptake and accumulation of heavy elements; these plants are applied to remediation of contaminated soil (Bradley and Vielly, 2001). Phytoremediation is a cost-effective method that is used to remediate the contaminated soil without any adverse effects to the environment (Shump 1993).
The selection of plant species for phytoremediation is the most important issue in the remediation of contaminated soil. Today because of difficulty in culture and plants adaptation from other region of contaminated soil, and also, for insufficiently of growth, some challenges are appropriated to use domestic and native plants for phytoremediation.
Successful phytoextraction of metal from contaminated soil requires the plants to tolerate the metal. Plants must also be able to produce sufficient biomass besides accumulating a high concentration of heavy metals.

Even though developments were done in phytoremediation, techniques are needed for future investigations on different soil sciences fields such as, microbiology; physics and soil chemistry (Bennett et al. 2003).

Phytoremediation was intended firstly by Raskin (1991) in a conference about approaches to decreasing pollution in the environment, then was defined according to Cunningham and Berti (1993); followed by Schnoor (1995) who introduced a more appropriate definition.

Generally, in Phytoremediation, the selection of a suitable plant species, that has high potential to reduce soil contamination, is very important. Iran is an arid to semi arid country and has extant of halophytes plants. For high tolerability of halophytes plants in saline and arid conditions, and also, for limited numbers of Pb hyper-accumulator plants, Lambsquarters (*Chenopodium album* L.) is known as one of the accumulator plants for heavy metal such as lead. This plant specie has ability of Pb accumulation not only in their roots, but also can transfer it to their shoots, that are the phytoextraction end. Instead, the amount of Pb accumulated in roots is higher than its shoots.

Pb concentration that was reported in various plants by researchers is very various. This variation may be due to environmental and genetic factors. Although concentration of Pb varies in different parts of plants, but occasionally in some plants usual concentration of Pb is lower than 3 $mgKg^{-1}$ (Henry, 2000). The main object of this paper is to study the potential of halophyte lambs quarters tolerability in Pb contaminated soil and in its accumulation.

Because lambs quarters plants have enough biomass in high concentration of Pb, even though the Pb accumulation in its aerial organs is not as it in hyperaccumulator plants, it can be used as a phytoextraction plant in contaminated soil.

Material and methods

Experiments were done in a set of 28 pots (1 element × 4 repeats × 7 treatments) according to Complete Randomly Design. Soil was passed on a 400 µm (No. 40) sieve. Initial specific gravity of soil was 1.344 $g\ cm^{-3}$. All pots were filled by 7 Kg soil. Soil was contaminated by 0, 150, 300, 600, 900, 1200 µg $PbKg^{-1}$soil; respectively, named T_0, T_{150}, T_{300}, T_{450}, T_{600}, T_{900} and, T_{1200}. Pb was applied as $PbNo_3$. In this study, amount of transpiration was measured by gravity methods, and plants were harvested after the end of shoot and roots growth. Fresh weight of plants in all pots was measured, and then harvested plants were washed by distilled water. Washed plants dried in oven at 130 °C, and then ground. Pb concentration in ground Plants samples was obtained in wet oxidation method by ICP instrument..

All values reported in this work are means of four independent determinations. The mean values are given in tables. All the data has been statistically analyzed by one way analysis of variance (ANOVA) in randomized complete block design to check the variability of data and validity of results. Comparison between control and treatment was done by LSD test (Gomez and Gomez, 1984). All tests were performed with SPSS software.

Results and Discussion

Obtained results from concentration of total Pb, soluble Pb, and Pb in roots and shoots of lambs quarters plants, by ICP instruments are shown in table1. Data presented in this table show amounts of Pb measured in soil and in plants.

Table1. Concentration of total Pb, soluble Pb, and Pb in roots and aerial organs of lambs quarters plants

Pb in soil (µgr)	Pb in shoot (ppb)	Pb in root (ppb)	Total soluble (%)	Soluble Pb (ppm)	Total Pb (ppm)
0 (T_0)	3371.349	10132.776	1.24%	0.46	37.25
150(T_{150})	3792.035	50329.010	0.35%	0.52	150.25
300(T_{300})	4251.595	90802.200	0.20%	0.6	286.09
600(T_{600})	5539.026	99634.750	0.12%	0.68	547.50
900(T_{900})	6792.106	120477.089	0.09%	0.76	827.50
1200(T_{1200})	9020.986	128935.185	0.07%	0.84	1182.00

Fig1 shows trend of Pb amount in shoots of lambs quarters plants in respect to Pb concentration of soil. Results shows, except of T_{1200}, increase in Pb concentration of soil causes increase in amount of Pb in shoots. This result indicated that although Pb was identified as contaminated agents for plants, but in this case to some levels of Pb contaminations has increased growth of plants. Absorbed Pb in plants to certain concentration (900µg) was acted as a factor to improving plants growth and accounted as a nutrient substance by plants, which applied in cellular interactions.

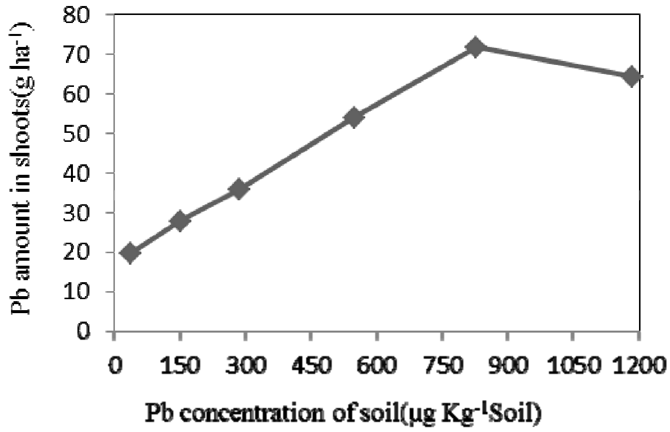

Fig1. Amount of Pb in shoots of lambs quarters plants in respect to Pb concentration of soil.

Despite the existence of toxic and or stress factors, if other growth factors are optimum, they could reduce productivity in plants, and according to previous studies, high concentrations of Pb are toxic and cause stress on plant growth (Porębska and Ostrowsk, 2001). Then, the existence of Pb toxic ions, NO_3^{-2} can't increase plants growth. In other examples, even though other factors such as soil humidity and nutrient elements are in optimum levels, salinity stress decreased plants productivities.

Results in fig.2 show that the increase in Pb concentration in soil causes a rise in the concentration of Pb in shoots and roots. But this increasing in roots is higher than shoots. Plants differ widely in their capacity to uptake and accumulate heavy metals in various organs.

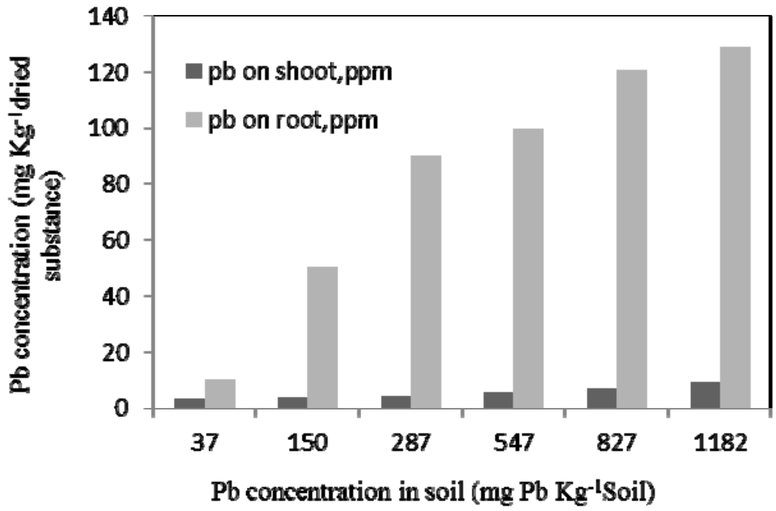

Fig2. Concentration of Pb in shoots and roots of lambs quarters plants in respect to Pb concentration of soil.

Even thought in high concentration of Pb in soil, T_{1200}, any toxic effect wasn't observed in this plant. In conformity with the observations of earlier researches, Asadi and Homaii (2009) found that the Pb concentration in shoot and in roots of *Chenopodium botrys* was respectively 5.6-54 mg

Kg^{-1} and 22.8-97.7 mg Kg^{-1} . Pb concentrations of roots and stems were respectively, 1.8-18 and 7.6-32.2 rather than usual Pb concentration of same plant (Asadi and Homaii, 2009).

In other studies some differences were also observed in the distribution pattern of Pb in *C. album* and *C. botrys*, which shows *C. album,* has more effective results for Pb accumulation than *C. botrys*. This indicated that *C. album* with efficient biomass has ability on translocation of Pb from contaminated soils, which is very important factor in phytoremediation.

Comparison of Pb accumulation on roots and shoots of *C. botrys* shows that the increased Pb concentration in the soil enhanced significantly the Pb accumulation of root than that of shoot (Fig3).

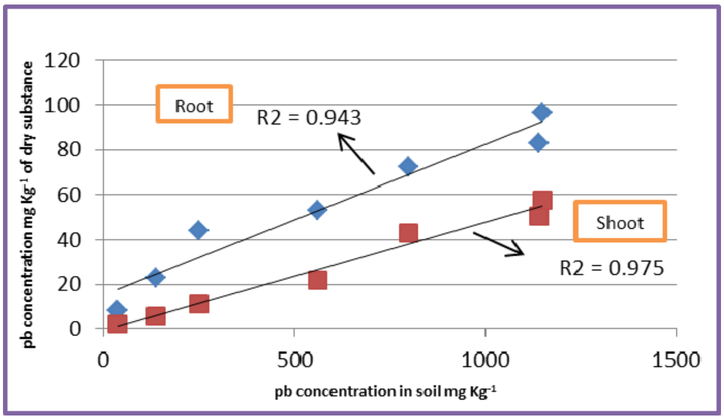

Fig3. Comparison of change in Pb accumulation on roots and shoots of *C. botrys*

It is also well demonstrated that the increased concentration of Pb in soil may lead to a change in accumulation pattern of Pb in the root and shoot of *C. botrys*.

Fig (4) shows that in *C. album* Pb accumulation on roots is higher than that on shoots. Also, it shows that the increased Pb concentration in the soil enhanced significantly the Pb accumulation of root than that of shoot (Fig3).

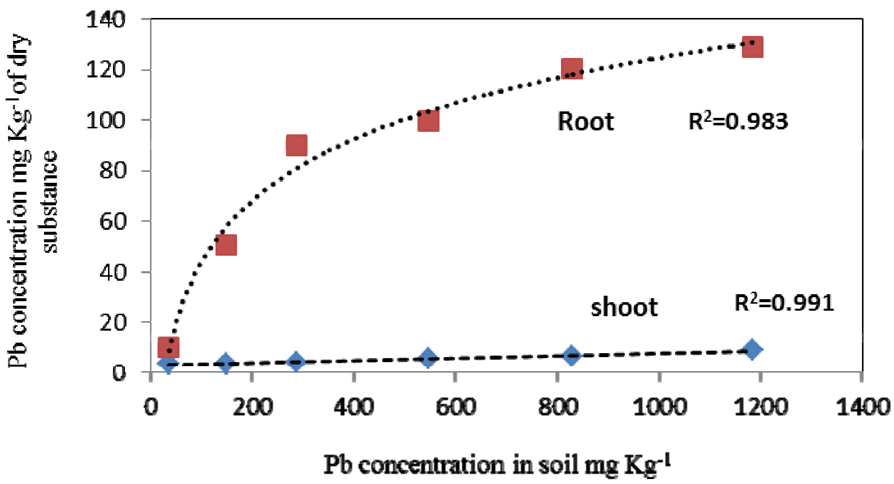

Fig4. Comparison of change in Pb accumulation on roots and shoots of *C. album*

According to these results, portion of Pb remediation in root higher than shoot of *C. album*, then we can conclude that *C. album* is an appropriate option to remediate in soil contaminated by Pb. Also, in phytoremediation of heavy metals, several factors such as plant tolerability and its ability in translocation of heavy metals from roots to shoots, high growth and biomass producing must be considered (Gomez and Gomez, 1984).

It is also well documented that *C. botrys* has effective results in soil remediation than *C. album,* especially in their shoots, but portion of Pb accumulation in roots of *C. album* can not be ignored.

Results from present study showed that roots of the *C. album* plants have great potential to tolerate and absorb Pb from the growth medium.
Fig (5) shows that ratios of Pb accumulation in shoot to root of *C. album* has reduced by increasing in Pb accumulation in roots, justifying that this plant can be a potential candidate for pb phytoremediation from contaminated soil.

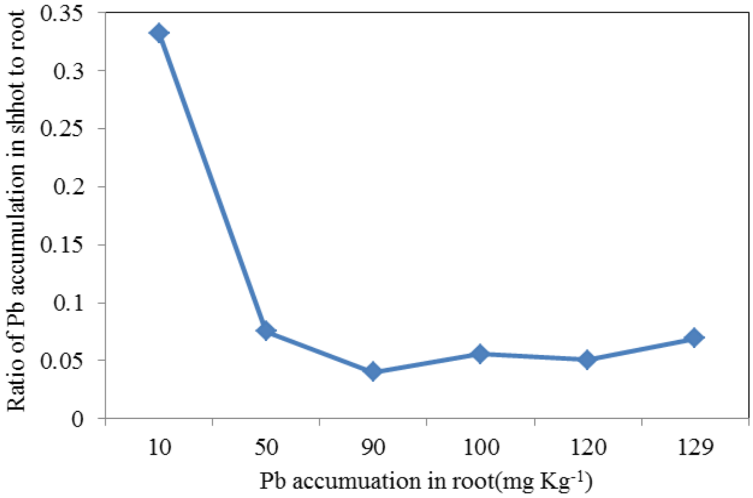

Fig 5. Ratios of Pb accumulation in shoot to root of *C. album*

This plant yields high percent of above ground organs and can be introduced as a halophyte plant which can be successfully used in phytoextraction of soil contaminated by Pb.

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Interaction between Cross-Linked Polyacrylamide and Water and Solute Flow in a Sand and Loam

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Abstract
Superabsorbents such as cross-linked polyacrylamides (PAM) are frequently used to increase water availability to plants. Commercial PAM providers suggest that besides its increasing water availability, PAMs also increase nutrient availability to plants. To test this premise, four application rates (0.0, 0.33, 1.66, 3.33 and 6.66 g kg^{-1}) of a PAM were mixed with sandy and loamy soils to evaluate its effect on nutrient leaching and retention in these soils. Miscible displacements of chloride were conducted on columns of PAM-soil mixtures and results were evaluated by an equilibrium CDE model. Increasing the PAM rates up to 1.66 g kg^{-1} resulted in increased early appearance and dispersive transport of chloride in sand. In addition, increasing the PAM rates caused gradually increased tailing of the breakthrough curves of chloride in both soils. These suggested that PAM increased preferential transport of chloride while it increased retention of chloride in soils. Effect of PAM on preferential transport and retardation of Cl was greater in sand than loam.
Keywords: PAM, Equilibrium CDE model, Preferential flow, Miscible displacement, Breakthrough curve.

Introduction
Cross-linked polyacrylamides (PAM) are used to increase water use efficiency in especially sandy soils in arid and semiarid regions worldwide (Bhardwaj et al., 2007). Some studies (Gallaghan et al., 1988; Baker, 1990; Silberbush et al., 1993; Yetgin Uz et al., 2010) showed that use of PAM increased water availability to plants, while others (Austin and Bondari, 1992) reported its detrimental effects on plant yield and survival.

Efficiency of PAM depends soil conditions and on balancing cation in PAM (Bharwaj et al., 2007). When mixed with soil, efficiency of PAM is decreased as its complete swelling is restricted by confining soil particles. Buchholz (1998) reported that positive results were found when small amounts of PAM were used with dry soils. In addition, efficiency of PAM is decreased by salinity of soil water since saline water decreases the water holding capacity of PAM. Kazanskii and Dubrowskii (1992) suggested that adding polymers to sandy soils may improve water holding capacity of soils due to polymer swelling and decreased hydraulic conductivity of sand and lowered drainage water below the root zone.

Commercial PAM providers often declare that PAM not only increases water availability to plants but also nutrient availability due to that it absorbs nitrate-like anionic nutrients and slowly release when plants need. To our knowledge, no study has been conducted to test this premise. Therefore, the objective of this study was to investigate the effect of PAM on retardation and leaching of Cl in a sandy and a loamy soil.

Material and Methods
Material
The experiments were conducted using disturbed columns of a sand and loam. The loam samples were taken from 0-30 cm of a wheat field and sand samples from an uncultivated area on an alluvial terrace, 300 m away from the first sampling site, in northeast of Çankırı, located in north of Central Anatolia, Turkey. Some selected properties of soils are given in Table 1.

Table 1. Some properties of soils used in the miscible displacement tests

Soil	Sand %	Silt %	Clay %	OM %	$CaCO_3$ %	pH	EC
Loam	45	18	37	0.47	10.5	7.4	4.5
Sand	70	6	24	1.75	13.8	7.6	5.4

OM: Organic matter content, EC: Electrical conductivity (mmhos cm^{-1})

Methods

The soils were transported to laboratory, sieved with 2-mm screen and mixed thoroughly with a cross-linked polyacrylamide (PAM) in 0.33, 1.66, 3.33, and 6.66 g kg^{-1}. The samples were packed in 30 cm long plastic columns with 8.4 cm id. The lower end of the column was supported with a fabric. To achieve an adequate packing, bottom of the columns were hit gently 20 times to the laboratory bench during packing. Each PAM rate was repeated three times with each soil. Therefore, 2 (soils) x 5 (PAM rate) x 3 (replicates) = 30 soil columns were packed in total.

Miscible Displacement Tests

The column was gradually saturated with 0.01 M KBr solution from the bottom of the column (van Genuchten and Wierenga, 1977). Upon saturation, the column was positioned on an upright stand. The inlet at the top of the column was connected to Marriotte system, and the outlet at the bottom was connected to an automatic fraction collector. Saturated hydraulic conductivity of the column was determined with the bromide solution after steady state flow rate was established. Approximately three pore volumes of tracer solution of 0.05 N $CaCl_2$ in 0.01M KBr solution were introduced into the column with the Marriotte system followed by approximately four pore volumes of 0.01 M KBr to leach the tracer Cl from the column. The effluent was analyzed for chloride using an ion specific electrode (Abdalla and Lear, 1976).

Relative concentrations (C/C_o) of Cl were calculated by dividing the concentration of Cl of the effluent collected by the concentration of in the stock solution. The data from the flow experiments were quantitatively evaluated using the computer program STANMOD (Deterministic Equilibrium CDE). Coefficient of hydrodynamic dispersion D (cm^2 h^{-1}) were fitted and the pore water velocity ν (cm h^{-1}) and pulse duration T_0 (dimensionless) were measured. Retardation coefficient R (dimensionless) was assumed unity for nonreactive Cl. Dispersivity λ (cm) was calculated by $\lambda = D/\nu$.

Results and Discussion

The experimental BTCs showed both early breakthrough and tailing (Figs. 1 and 2), which is indicative of both preferential transport and mass exchange between mobile and immobile regions in the columns. In general, early appearance of Cl increased with increasing PAM rate in both sandy and loamy soils. However, extent of early appearance was greater in the sand. The early appearance is resulted from nonuniform transport of solute in macropores and similar pathways where water flows more rapidly than the adjacent regions. In general, these preferential pathways can form by physical (cracks, fissures, funnels, etc.) and biological (decayed soot channels, earthworm channels, animal burrows, etc.) means (Beven and German, 1982). In this experiment, preferential pathways would form between swelled PAM and surrounding soils particles, through which water and solutes moved rapidly and appeared earlier at the exit of the columns.

The effect of PAM rate on preferential transport of Cl in sand columns was more drastic than in loam. This was attributed to the fact that in greater rates, PAM particles were subjected to increasingly greater pressure from the soil particles in loam that resulted in decreased macroporosity. As the PAM rate increased, more pressure exerted by surrounding soil particles on swelling PAM particles and this caused limited space available for macropore-like conduits in the column, ultimately resulting in decreased preferential transport of Cl in the loam.

Table 2 shows that increased PAM rate had no significant effect on *v*, D, and λ in loam, while it significantly affected these variables in sand. This was attributed to that within-treatment variation was greater in loam columns compared to sand columns. This high within-treatment variation resulted that the means for treatments were not significantly different in the former. Compared to loam, increasing PAM rate had a significant effect on *v* at low PAM rates, and it consistently increased λ in sand.

Table 2. Means and standard deviations for pore water velocity, coefficient of hydrodynamic dispersion, and dispersivity obtained on repacked sand and loam columns used in this study.

PAM $g\ kg^{-1}$	***v*** $cm\ h^{-1}$	**D** $cm^{2}\ h^{-1}$	**λ** cm
		Loam	
0	^{a}24.44±8.38	^{a}148.6±95.1	^{a}5.16±3.13
0.33	^{a}16.28±9.86	^{a}143.5±128.6	^{a}9.27±5.05
1.66	^{a}25.15±2.58	^{a}187.3±52.8	^{a}7.27±2.48
3.33	^{a}25.23±3.10	^{a}158.7±58.8	^{a}6.53±3.05
6.66	^{a}16.47±3.08	^{a}102.3±5.51	^{a}6.47±1.42
		Sand	
0	^{a}25.46±19.79	^{a}101.0±103.2	^{a}3.51±0.96
0.33	^{b}49.79±1.61	^{b}406.6±49.24	^{b}8.15±0.74
1.66	^{b}54.33±6.30	^{c}391.3±33.84	^{b}7.22±0.53
3.33	^{a}25.71±0.21	^{c}270.3±23.03	^{c}10.51±0.90
6.66	^{a}20.77±0.12	^{c}316.3±33.08	^{d}15.23±1.88

Means for the same soil with same letter in the same column are not different at the significance level of 0.05.

v: pore water velocity, D: coefficient of hydrodynamic dispersion, λ: Dispesivity (D/*v*)
n= 3 for each mean. Numbers following ± are standard deviations of means.

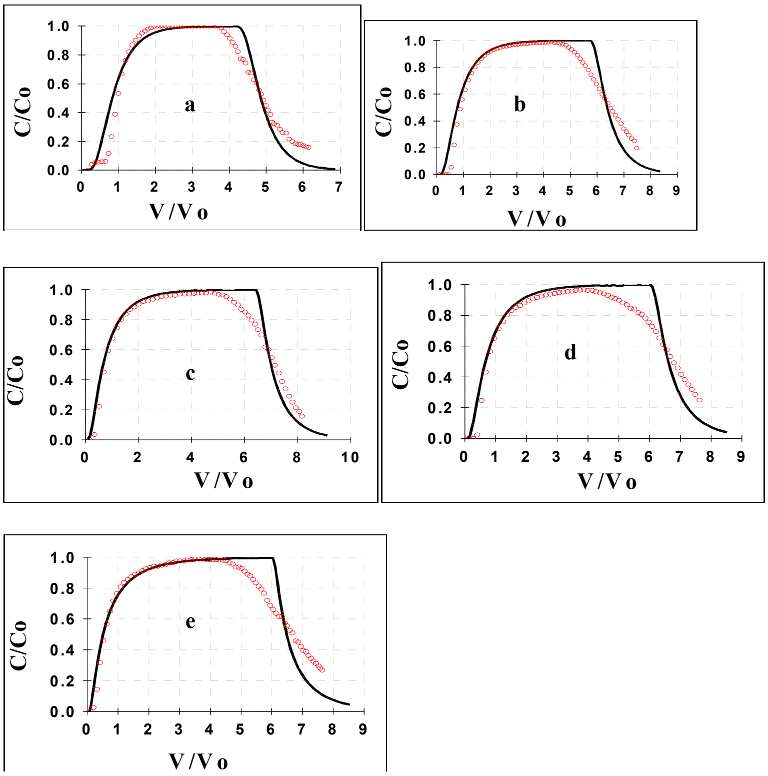

Fig.1. Breakthrough curves of chloride at disturbed soil columns of loam with a) 0.0 g kg^{-1}, b) 0.33 g kg^{-1}, c) 1.66 g kg^{-1}, d) 3.33 g kg^{-1}, e) 6.66 g kg^{-1} PAM rates. Dashed lines are experimental BTCs and continuous lines are BTCS estimated with equilibrium CDE (CXTFIT) of STANMOD.

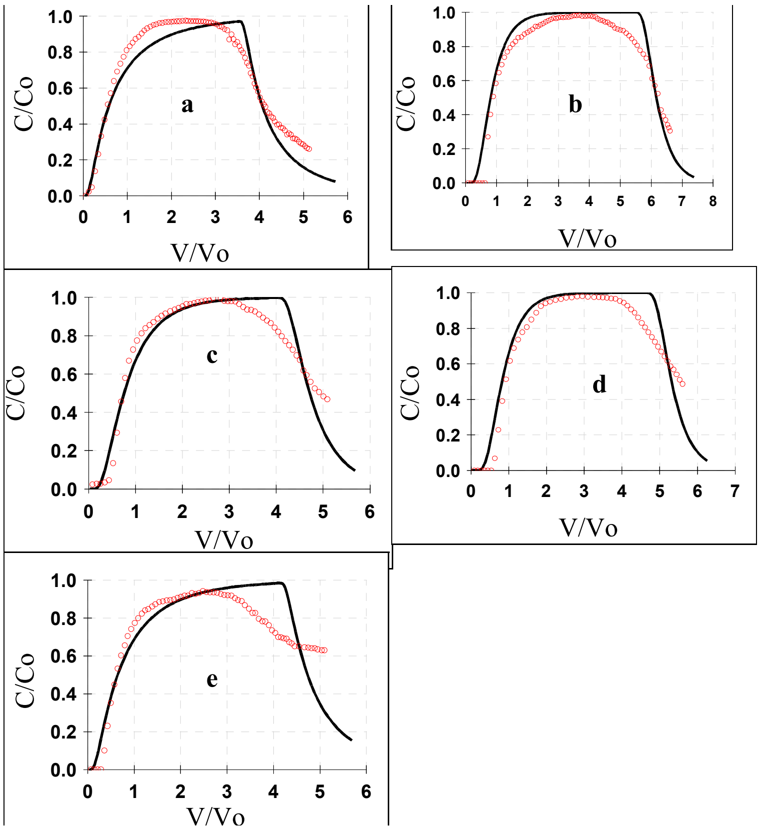

Fig.2. Breakthrough curves of chloride at disturbed soil columns of sand with a) 0.0 g kg^{-1}, b) 0.33 g kg^{-1}, c) 1.66 g kg^{-1}, d) 3.33 g kg^{-1}, e) 6.66 g kg^{-1} PAM rates. Dashed lines are experimental BTCs and continuous lines are BTCS estimated with equilibrium CDE (CXTFIT) of STANMOD.

Pore water velocity (v) increased drastically until PAM rate of around 2 g kg^{-1} in sand and then decreased with greater application rates (Fig.3 left). However, a dissimilar behavior of v was observed for loam.

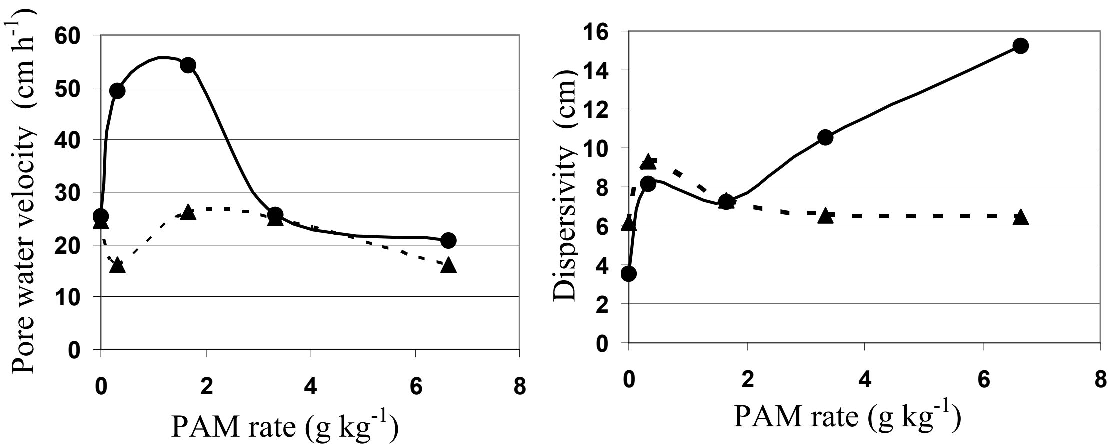

Fig.3. Change of pore water velocity (cm h^{-1}) at the left and dispesivity (cm) at the right as a function of increasing PAM rate. The continuous lines indicate sand and dashed lines loam.

Hydrodynamic dispersion is caused by diffusion of solute from greater concentration region to lower concentration region and nonuniform flow of water and solute in the flow paths (mechanical dispersion). Combined effect of these two processes is represented by hydrodynamic dispersion coefficient (D) in equilibrium CDE model. The variable dispersivity (λ), calculated by $\lambda = D/v$, indicates whether the transport is dispersive or convective. A value of λ greater than unity suggests that the transport is dispersive and otherwise it is convective. In our study, the transport was increasingly dispersive against increasing PAM rates in sand columns contrary to in loam columns (Fig.3 right one). This was attributed to interaction between swelled PAM particles and pore size distribution in sand columns. This was also attributed to the high salinity (Table 1) of sand, which would prevent the complete swelling of PAM as suggested by Taylor and Halfacre (1986). In loam, application of PAM in small amounts increased and further rates of PAM decreased the dispersive transport of Cl as indicated by dashed line.

An asymmetrical BTC with a right tail indicates incomplete transport caused by diffusive mass exchange between mobile and stagnant regions in the column. The experimental BTCs in Figs 1 and 2 (circles) showed that incompleteness increased with increasing PAM rates in both soils, indicating that some Cl diffused into the swelled PAM particles and was preserved there against leaching. In the sand, effect of PAM on retardation of Cl was more drastic than in loam as thicker right tails of experimental BTCs for sand (Fig. 2) indicated.

Conclusions

Mixing a cross-linked polyacrylamide (PAM) with a sandy soils in increasing rates accelerated the transport of Cl in repacked columns up to about 2 g kg^{-1} rates and decreased at greater rates. However, increasing the PAM rate promoted gradually the dispersive transport of Cl. Part of applied Cl was retarded by PAM as the Cl diffused into the swelled PAM particles where it was preserved against leaching. Tick right-tailing of BTCs indicated that the backward diffusion of Cl from the PAM particles to soil was slower than its forward diffusion.

Contrary to sand, mixing the PAM with a loam slightly affected the transport of Cl in the soil columns. Mixing the PAM with loam at increasing rates resulted in inconsistent values of v as a function of PAM rate and dispersive transport Cl in the soil columns was independent of PAM rate although PAM increased the dispersive transport slightly at the low application rates. In addition, PAM retarded some of the applied Cl in loam columns.

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The Effects of Cd Polluted Soil on Nitrogen Fixation of Native Strains of *Sinorhizobium Meliloti* and Yield of Alfalfa

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Abstract

The effects of heavy metals, as soil pollutants, on *Rhizobia* and symbiotic system of *Rhizobium*-legume are not clearly known. The aims of this study were to isolate heavy metals resistant strains and determination the effects of Cd on nitrogen fixation of native strains of *Sinorhizobium meliloti* in calcareous soils. To isolate and obtain pure cultures of *sinorhizobium meliloti* strains, root nodules were collected from different alfalfa farms of Zanjan province. Microscopic, biochemical and plant tests carried out on different strains, rhizobia were identified and strains capable of nodulating were determined. Based on effectiveness of symbiosis and shoots dry weight of alfalfa sown in greenhouse, nitrogen fixation efficiency of different strains was examined and compared with each other. The results showed that different strains have different nitrogen fixation efficiencies and the strains were classified into four groups namely as very effective, effective, partialy effective and not effective. In the second part of the experiment, resistance of different *sinorhizobium meliloti* strains to Cd were examined using selective medium of H.M containing different amounts of Cd. The highest concentration of heavy metals supporting growth for each strain was determined as the maximum resistance level (MRL) and based on it the strains were classified into three groups namely as sensitive, relatively sensitive and tolerant. The results showed that significant differences between the strains in this respect. In the final experiment, the effective strains (capable of nodulating) with low and high tolerance to heavy metals were selected and used for a pot experiment. In this experiment, the effects of five *sinorhizobium meliloti* stains (S_6, S_{12}, S_{17}, S_{41} and S_{51}) as inoculants, and five concentrations of heavy metals were examined on growth of alfalfa plant using a mixture of perlite and sand as media. A control and a treatment receiving 70 mg/kg N were also included in this experiment. The highest and lowest biomass yields of alfalfa were obtained for control and those treatments inoculated with resistant strains to Cd. The S_{51} strain was tolerant to Cd and had higher nitrogen fixation ability than the other strains. The S_{17} and S_3 were the most tolerant and the most sensitive strains to Cd respectively.
Keywords: Alfalfa, Cadmium, pollution

Introduction

Today the world is facing increasing pollution in the environment and damage to natural ecosystems such as soil. The use of chemical fertilizers brings about environmental issues and problems such as the contamination of subsurface strata with heavy metals namely Cadmium (Miola *et al* 1980). Some soils of Zanjan province are polluted as a result of foundries and the creation of industrial by-products (Jebeli 1378). Cadmium is the main pollutant in soil which can enter the soil via chemical fertilizers. Rhizobium has played a role in the increase of fertile layers for centuries. Rhizobium creates a connection for the endosymbiotic nitrogen fixing of the legominus plant. Legumes have a plenteous need for fertilizer which when coupled with the excessive use of chemical fertilizer is a probable cause of Cadmium contamination of subsurface strata. Research shows that Cadmium has a negative effect on the order of nitrogen fixation symbiosis. (Ibko et al 1995). The fixation of nitrogen in white clovers in polluted soils with different amounts of metal, it showed that plants in native fields and all areas testes the symbiotic bacteria are able to fix the nitrogen (Koleri 1997). Gilor McGrath (1989) pointed out that with a lack of rhizobium leguminosarum bv. Trifolii in order to fixate nitrogen in polluted soil 40 percent less output of white clovers. The effects of heavy metals on the number of micro organisms are more sever in sandy soil than clay soil. In a report from Brookes et al (1986) the fixation of nitrogen by heterotroph sianobacterias on the contaminated soil with a heavy metal decreased by 30 percent. Rother et al (1983) investigated the effect of heavy metals on white clovers and found out that the node and nitrogen fixation had a small reduction the amounts of Cadmium, Zinc and Lead were 216 mgKg, 20,000 mg Kg and 30,000 mg Kg respectively. One field experiment results of polluted soil with 10mgKg Cadmium, 52mgKg Nickel and 120 mgKg Zinc showed no effect on the nodes or nitrogen fixation. The great deal of pollution in Zanjan's agricultural fields as a result of Lead and Zinc factories there has been no experiments to determine the effects on the biological fixation of nitrogen in this province. This research aim is to investigate the effects of Cadmium pollution to the native fields, the sinorhizobiom symbiosis and nitrogen fixation.

Materials and Methods

Due to the close proximity of lead and zinc factories to the fields in Zanjan province, some of the fields in this province are polluted with heavy metals. Alfalfa root samples were collected and the nodes are separated from the roots, sinorhizobium meliloti was isolated from the nodes. The ability of each strain to symbiosis with alfalfa was studies (plant infection test) (Vincent 1982). Then the nitrogen fixation of the rhizobium strains are studied in a greenhouse with 45 strains of rhizobiom in 3 reps. the symbiotic effectiveness was devised (Yek et al 1993). In the next stage of the research the effects of of Cadmium was assessed in solid media. The various strains of rhizobiom based upon its resistance against heavy metals (Sensitive, relatively sensitive and tolerant), levels of Cadmium were investigated on growth, nodulation and nitrogen fixation of the sinorhizobiom meliloti. 5 levels of Cadmium (0,2,10,20 and 30 mgKG) and 5 strains of bacteria (strains with a higher SE which has various resistance to heavy metal were used (Table 1). 6 plants were sown into each pot and were incubated for 90 days, after this period plants were cut off , then put in an oven at 65-70 degrees Celsius to dry out. The of nitrogen content of the shoot was measured as well nitrogen uptake.

Table1. Characteristics of used strains

strain	S.E.	Resistance to Cd
S6	153	Tolerant
S12	57	Tolerant
S17	86	Tolerant
S41	139	Tolerant
S51	73	Partially sensetive

Results and Discussion

Normally the final determination of any rhizobium is based upon is symbiotic ability, from here it is that 45 strains of pure rhizobium could host the vegetation and pollution tests. The comparison of levels of Cadmium on the shoot dry weight showed that with an increase of Cadmium concentration the shoot weight decreases. The amount of this in treatment showed maximum (0.6) and treatment 30 mgKg Cadmium (at least 0.2) this treatment witnessed 67 percent less output. The comparison of levels of Cadmium shows that with an increase in Cadmium the nitrogen concentration in the shoot was reduced, the highest concentrate of nitrogen (2.94%) was related to the witnessed treatment of Cadmium., the least cadmium (2.15%) is treated 30mgKg Cadmium(Table 2). The dry weight of the shoot of the inseminated plants with the experimented strains in various levels of cadmium are as follows: Relatively sensitive strain to cadmium with a symbiosis higher <(S51) strains with maximum symbiosis ,(S6) Strains resistant to cadmium,(S17 & S12) with regards to the results obtained its visible to see that the most and least amount respectively of the resistant strains and those with a high symbiosis higher are (S41 & S17) and the witnessed treatment.

Table 2. Effects of Cd on some plant indices

Cd concentration(mg/kg)	Shoot dry weight(g/pot)	N-Uptake (mg/pot)	N concentration(%)
0	0.6077a	2.943a	1.817a
20	0.5056b	2.871b	1.481b
10	0.2693c	2.781c	0.7548c
20	0.2187d	2.729c	0.5975d
30	0.1990e	2.643d	0.5295e

The interactive effects of the strains and the levels of cadmium on the shoot dry weight (Table 3) showed that with an increase of cadmium levels the effect of the strains on increase of shoot dry weight was reduced, and this reduction was different for each of the strains, in the way that the most dry weight (0.7408 gr /pot) was related to the inseminated plant with strain S6 (high SE and resistance to cadmium).

Table 3. effects of Cd and Strains on shoot dry weight(g/pot)

Cadmium concentration(mg/kg)					
Strains		5	10	20	30
Blank	0.2833mn	0.2901	0.2440mn	0.1893o	0.159p
S6	0.7408c	0.3584k	0.4185j	0.3290l	0.156p
S12	0.4288ij	0.4148ij	0.4757h	0.2975mn	0.140p
S17	0.6425e	0.4390ij	0.6860d	0.4442i	0.5706f
S41	0.6773d	0.6234e	0.3255l	0.6203e	0.3612k
S51	0.6887d	0.5728f	0.4956	0.3010m	0.2867mn
N-70	0.7324b	0.8526a	0.8124b	0.5122g	0.5667f

At 0 level of cadmium and the least amount (0.1401 gr/pot) in treatment S12 (high SE and resistance to cadmium) and at level 30mgKg was observed. In general the resistant strains with high concentrations of cadmium in comparison with the sensitive strains were less affected by high concentrations of Cd (Table 3). The results showed that the levels of cadmium affected the nitrogen fixation of the strains, the highest percentage of nitrogen (3.033%) was related to the inseminated plants with strain S41 (with a high SE and resistance to cadmium) at level 0 cadmium and the least amount of (2.367%) in treatment of S6 which had a high SE and resistance to cadmium and at level 30mgKg cadmium was observed (Table 4).

Table 4. effects of Cd and Strains on plant nitrogen concentration(%)

Cadmium concentration(mg/kg)					
Strains	0	2	10	20	30
Blank	2.66fghi	2.53ijkl	2.60hijk	2.66fghij	2.66fghi
S6	3.00c	2.76de	2.50jkl	2.340kl	2.36l
S12	2.73efghi	2.86cdef	2.73efghi	2.73efg	2.56hijkl
S17	2.96cd	2.93cde	2.83cdefg	2.76defghi	2.73efghi
S41	3.03bc	2.93c	2.73efghi	2.70fghij	2.53ijkl
S51	2.86cdef	2.76defgh	2.83cdefg	2.63ghij	2.60fghij
N-70	3.33a	3.30a	3.23a	3.20ab	3.03bc

An important point to remember here is that the 2 strains S41 and S6 both have high SE and is in the high resistant group of strains however in alfalfa pots with a high level of cadmium they didn't show consistent reactions and the effects of no cadmium to S6 strain were greater. Similar to this situation is of the dry weight and total nitrogen was observed. In general with an increase of the levels of cadmium the effect of the strains on the nitrogen concentration was less however this reduction was different in the strains.

Conclusion

The results obtained with this research shows the studied strains have around 60% resistance to cadmium and only show growth in 10-30 mgKg cadmium. While 8.9% of the strains are able to tolerate levels 70 -80 mgKg Cadmium and have good resistance. 4.4% of the strains had a very high resistance in tolerating cadmium that even to the last level (150mgKg) of cadmium. The rhizobium symbiosis systems the effects of increased concentration of heavy metals in the soil decreased the amount of bacteria and symbiosis and this made nitrogen fixation reduction. In relation to the strains resistant to heavy metals for the fact that they are tolerant to the metals the growth is more active than the bacteria in nitrogen fixation. It is visible in this research and results that with an increase in cadmium from level 0 the nodes and dry weight of the shoot weight are reduced. Other researchers have also commented on the effects of metals and rhizobiums in soil and their resistances and they believe metals in the soil affect plants (Angel et al 1989). Ibko et al (1995) also reported that plants have protective elements with rhizobiums against the toxins present in metals. The results given alongside Angel et al 1989, results about the toxins in cadmium over alfalfa and rhizobioms symbiosis are similar reported the main reason are the genetic differences of the strains in the resistance of metals (Angel et al 1989).

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Emergent Pollutants in Soil Amendments: Treatment of Antibiotics in Sewage Sludge and Animal Manure

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Abstract
Sewage sludge and animal manure have been recognized and used as soil amendments to improve soil characteristics, and provide nutrients for crop growth in agricultural applications. However, the presence of antibiotics and some other emerging pollutants both in sewage sludge and animal manure from confined animal feeding operations has raised much concern. Although adopted as the best available technology, biological treatment does not permit the removal of most emerging pollutants. It has become evident that the application of advanced treatment methods can be crucial for the safe recycling of these wastes.
This study aims to investigate the chemical oxidation of antibiotics in sewage sludge and animal manure in order to prevent the transfer of antibiotic pollution to soil and the development of bacterial antibiotic resistance. Laboratory experiments with artificially contaminated manure and sludge revealed the importance of pretreatment that provided the extraction of antibiotics from solid phase for the efficiency of the subsequent chemical oxidation process. Ozone and Fenton oxidation processes were examined over a wide range of experimental conditions to assess the value of process parameters. The application of the integrated process consisting of extraction pretreatment and chemical oxidation with either Fenton or ozone yielded oxytetracycline removal efficiencies over 90% both in manure and in sludge.
Keywords: soil amendments, sludge, manure, antibiotics, advanced oxidation processes

Introduction

Utilization of manure and biosolids for soil amendment is desirable due to the possibility of nutrient recycling. Land-application of biosolids is stringently regulated for specific organic and inorganic substances. In the US, the regulations generally cover heavy metals (HM) and pathogens. A limited number of states also regulate the polychlorinated biphenyls (PCBs) (NEBRA, 2007). The EU regulations also include some priority organic pollutants (PCBs, adsorbable organohalogens-AOX, linear alkylbenzyl sulfonates-LAS, diphthalate(2-ethylhexyl)-DEHP, nonylphenols-NPE, polycyclic aromatic hydrocarbons-PAH, polychlorinated dibenzodioxins/dibenzofurans-PCDD/F) (Jones-Lepp and Stevens, 2007) along with HMs and pathogens. The Turkish biosolids regulations similarly set standards for the limits of HMs, pathogens and for the previously listed organic compounds regulated in the EU (OG. 04.08.2010/27661). The pharmaceutical and personal care products (PPCPs) have not been included in neither of the mentioned legislation programs. However The US National Council (NRC) has recommended that occurrence of these in biosolids shall be thoroughly investigated (Jones-Lepp and Stevens, 2007). With improved analysis techniques in the recent decades, detection of PPCPs at significant quantities has increased concerns leading to the need of more extensive research as was demonstrated by the NRC report. As a result, implementation of new regulations to cover these is expected in the future.
Utilization of raw manure directly as fertilizer by the farmers is very common. For compost manure, in the US, the states have set regulations based on the best management for nutrients (Brington, W.F., 2000). In the EU, some countries have implemented eco-label standards, which regulate components such as HMs, organic matter, N content, pathogens, etc. In some of the EU countries, the labeling of compost is required by law (EC, 2012). However, again no emphasis has been put on the PPCP content of either raw manure or compost yet.
Significant concentrations of the non-regulated compounds have been detected, which include endocrine-disrupting chemicals (EDCs) and pharmaceuticals and personal care products (PPCPs) (Jones-Lepp and Stevens, 2007). Among these contaminants, antibiotics deserve a major concern because of their contribution to the risk of antibiotic resistance development as well as their adverse effects on the living organisms (Kim and Aga, 2007). Antibiotics have been detected at

high concentrations both in manure and in biosolids (Oncu and Balcioglu, 2011) and can exert high loads when these are applied as soil amendments. The presence of antibiotics in the soil constitutes a serious pollution potential by creating a risk for development of resistance in the local bacterial communities (Kummerer and Henninger, 2003). Antibiotic resistant bacteria or antibiotic resistance genes have been detected almost everywhere (Oncu and Balcioglu, 2011). These findings demonstrate the ineffectiveness of conventional biological processes. Therefore proper treatment of both waste sludge and manure aimed for soil amendment is required.
The chemical oxidation processes, ozone and Fenton technology have been utilized effectively for the destruction of antibiotics as well as resistance carrying Plasmid DNA both in water and manure (Oncu et al., 2011; Cengiz et al., 2010; Uslu and Balcioglu, 2009a, 2009b). Therefore, these processes can be proposed as promising technologies for the remediation of manure and sludge and for the improvement of their amendment properties.

Materials and Methods

Manure and waste activated sludge (WAS): The cow manure utilized in the study was obtained from an ecological farm in Turkey. Prior to utilization, the manure was dried at 70 ºC for 24 h and sieved (2 mm), and subsequently sterilized by autoclaving at 120 ºC for 15 minutes to prevent biological activity. The WAS was obtained from the recirculation line of an advanced biological wastewater treatment plant (WWTP). After collection, the WAS was concentrated up to 19.5±2.5 g/L total solids (TS) by settling and decanting the top liquid layer. The manure and WAS were stored at 4°C; the WAS was utilized within 1 week of collection. Then, the manure and WAS were artificially contaminated with oxytetracycline (OTC) to make final concentrations of 20 mg/kg manure and 20 mg/L sludge, respectively. Extraction by magnesium salt at pH 8 was applied both to manure and WAS prior to oxidation, in order to increase the oxidant-target contaminant contact as described previously (Uslu and Balcioglu, 2009a; 2009b).

Treatment of manure and WAS: While ozonation of manure (1 g) was carried out in a 10 mL Teflon tube, WAS (1.5 L, TS=2.5 g/L), ozonation was performed in a 2 L-cylindrical recycling glass. The effect of the oxidant dose and the pH were investigated in the ozonation experiments.
Fenton oxidation of both manure and WAS was carried out by the addition of Fe_2SO_4 and H_2O_2, sequentially. pH adjustment prior to the addition of oxidant was performed with H_2SO_4. Similar to the ozonation experiments, the manure oxidation was performed in 10 mL Teflon tubes. During the treatment, mixing of the manure and WAS (300mL) was provided in a thermostated shaker (Julabo SW 22) at 150 rpm and 25 ºC. While the treatment period was 24 h for the manure, the WAS was treated for 2h.

Analytical Methods: The antibiotics were recovered from two different waste matrices by ultrasonic assisted extraction followed by solid phase extraction and were analyzed by liquid chromatographic analysis. The antibiotic extraction and SPE purification procedures are described elsewhere (Uslu and Balcioglu, 2009a; 2009b). The subsequent analysis of the antimicrobials was carried out by HPLC (Agillent Technol. 1100 series) utilizing an YMC-Pack ODS-AQ column (3 µm, 50 x 4.0 mm). Acetonitrile (ACN, HPLC grade, Sigma Aldrich) and water (Milli-Q), both containing 0.1% formic acid (98%, Fluka) were used in gradient elution. OTC was detected at 360 nm with a diode array detector (DAD). The limits of detection (LOD) for OTC in manure and sludge were 0.01 mg/g and 0.22 mg/kg of dry weight, respectively.

The pH of the manure slurry and WAS were measured using a pH probe (WTW pH 330 pH meter). Soluble COD (SCOD) solid contents (TS and volatile solids (VS)) were determined in accordance with the standard methods (APHA, AWWA, WPCF, 2005). Determination of the organic carbon (OC) in the precipitated manure, the alkaline soluble organic carbon (sum of humic acid and fulvic acid carbon; C) the fulvic acid carbon (C_{FA}) content, the humic acid carbon (C_{HA}), and the TKN content of manure is described elsewhere (Uslu and Balcioglu, 2009b). The NH_4^+-N and NO_3^--N in manure were extracted with 2 N KCl and determined by the Nessler and Cd-reduction methods, respectively (Hach DR/2010 manual). The TKN and the total phosphorus (TP) contents of the

WAS were analyzed by the Digesdahl method provided by Hach (Hach DR/2010 manual). The NH_4^+-N in the liquid phase of the WAS was similarly determined by the Nessler method and the reactive phosphorus (PO_4^{3-}) was determined by ion chromatography (IC) (Dionex ICS-3000). Soluble contents of both manure and WAS were analyzed after filtration through cellulose acetate 0.45 μm filter (Sartorius, Germany). Hydrogen peroxide was determined by the iodometric method. The inlet and off-gas ozone concentrations were monitored with an online ozone analyzer (Fisher Ozotron 23).

Results and Discussion

The average characteristics of the manure and WAS are listed in Table 1.

Table 1. Characterization of WAS and manure

Manure		**WAS**	
Parameter	Value	Parameter	Value
pH	8.6	pH	6.5
Organic carbon (%)	26.5	TCOD (g/L)	10.7
Total nitrogen (%)	1.42	SCOD (mg/L)	130.0
C (Humic Acid) (%)	0.98	TKN (mg/L)	350
C (Fulvic Acid) (%)	1.15	TP (mg/L)	100
NH_4^+-N (mg/kg)	195	Alkalinity (g/L)	1.5
NO_3^- -N (mg/kg)	510		

In order to increase the effectiveness of the applied oxidation process for antibiotic destruction, both manure and WAS were subjected to a selective extraction pretreatment. The extraction efficiencies of OTC from manure and WAS with the optimum magnesium salt concentrations and after 30 min and 3 h of extraction periods, respectively are given in Table 2. 63.9% and 50% OTC extraction could be achieved by the pretreatment from manure and WAS, respectively as a result of complex formation between the magnesium cations and the OTC molecules (Tongaree et al., 2000).

High removal rates of OTC were achieved both from manure and WAS even by 15 min of ozonation. This is most probably the result of a reaction between the desorbed OTC to the liquid phase and the oxidant in aqueous phase of the slurry. The differences in the reaction conditions (volume, OTC concentration, oxidant dose, etc.) and the matrix components of the manure and WAS systems resulted in different removal rates in the early stages (i.e. 15 min) of ozonation; nevertheless, high removal rates were obtained in both matrices by 60 min of ozonation. It is obvious that a considerably high ozone dose is necessary for high OTC removal. But antibiotics are not the only pollutant in WAS that limits its disposal to land. Despite the increase in the SCOD by WAS solubilization at the higher ozone dose, the overall OTC removal reached 98.2%. It can be deduced that the amount of the solubilized sludge constituents in the final stages of ozonation have a minor effect and the OTC removal may become desorption-limited after consumption of the antibiotic present in the soluble phase. Complete removal of OTC could not be achieved both from manure and WAS. Higher oxidant doses may be required for the removal of the strongly bound residual antibiotic. During Fenton oxidation, remarkable degree of OTC removal from manure was obtained in 2 min of reaction as a result of the pretreatment. The removal was improved only by 13 % by extending the reaction period to 24 h. Comparative OTC removal from WAS was obtained in 2h of reaction, however with a relatively higher H_2O_2 dose from that utilized for manure. Again this may be the result of differences in the reaction conditions as well as the result of the effect of the different matrix components. The overall OTC removal degrees achieved in manure and WAS were lower than those obtained with ozonation. This can be due to fast consumption of the initially added oxidant in Fenton oxidation, as opposed to the continuous oxidant supply during ozonation.

Table 2. Ozone and Fenton oxidation of manure and WAS: Effect of oxidation on antibiotic removal, organic carbon and SCOD

Mg Pretreatment			
Manure Mg^{2+}=0.07g/g manure		**WAS Mg^{2+}=1.21 g/g TS**	
Pretreatment time	OTC in liquid phase	Pretreatment time	OTC in liquid phase
30 min	63.9 %	3 h	50.0 %
Ozone Oxidation			
Manure O_3=0.02 g/min, pH=10		**WAS O_3=0.35 g/min, pH=8**	
Reaction time	OTC removal	Reaction time	OTC removal
15 min	92.0 %	15 min	79.1 %
60 min	96.0 %	60 min	98.2 %
Reaction time	COD* solubilization	Reaction time	COD solubilization
15 min	32.4 % increase	15 min	44.7 % increase
60 min	36.5 % increase	60 min	336.3 % increase
OC	No change		
Fenton Oxidation			
Manure H_2O_2=44 g/kg manure Fe(g)/ H_2O_2 (g)=1.65, pH=2.6		**WAS H_2O_2=150 g/kg TS : Fe(g)/ H_2O_2(g)=1.3, pH=3**	
Reaction time	OTC removal	Reaction time	OTC removal
2 min	80.0 %	2 h	90.0 %
24 h	93.0 %		
Reaction time	COD solubilization	Reaction time	COD solubilization
2 min	50.2 % decrease	2 h	341.1 % increase
24 h	13.2 % decrease		
Reaction time	OC change		
2 min	4.5 % decrease		
24 h	4.5 % decrease		

*COD=SCOD

Table 2 also presents the variation of SCOD values by the application of oxidation processes to the manure and WAS. As can be seen from the table, oxidation resulted in COD solubilization. Higher SCOD release from the WAS was obtained by the Fenton treatment compared to ozonation. By applying high ozone doses continuously, simultaneous SCOD release and degradation could have taken place. Solubilization of manure did not improve remarkably by ozonation after 15 min, while the increase in the SCOD of WAS was more evident during prolonged ozonation, showing higher amount of solubilizable content. While high release of SCOD was obtained in the WAS by Fenton oxidation, the SCOD in manure decreased during the first 2 min, showing the reaction of the oxidant with the manure constituents. Thereafter, the SCOD increased as a result of organic matter release from manure, but the overall final value was lower than the initial. This difference in the SCOD behavior during Fenton oxidation of the two matrices can be due to their different textures. The OC content of the manure was not affected during ozonation, while decrease was observed during the Fenton process. It can be concluded that ozone reacts selectively with OTC rather than other manure constituents.

The effects of the pretreatment and ozonation on the soluble ammonia and the reactive phosphorus content of WAS, as well as the effect of ozonation on the mineral nitrogen content (N_M) of the manure are shown in Table 3. The fate of the nutrients in the manure was not evaluated during the pretreatment.

Table 3. Ozone oxidation of manure and WAS: Effect of oxidation on nutrient components

Mg Pretreatment			
		WAS Mg^{2+}=1.21 g/g TS	
		Pretreatment time	Change in PO_4^{3-}
		3h	87.2 % decrease
		Pretreatment time	Change in NH_4^+
		3h	96.1 % decrease
Ozone Oxidation			
Manure O_3=0.042 g/min, pH=10		**WAS O_3=0.35 g/min, pH=8**	
Reaction time	N_M solubilization	Reaction time	PO_4^{3-} solubilization*
15 min	from 1.2 to 1.4	15 min	78.3 % increase
60 min	from 1.2 to 1.8	60 min	241.7 % increase
		Reaction time	NH_4^+ solubilization*
		15 min	900 % increase
		60 min	3100 % increase

*Based on the differences between the values after pretreatment and the values after oxidation

The pretreatment of the WAS by Mg^{2+} resulted in a remarkable decrease in the reactive phosphorus concentration (from 47 mg/L to 6 mg/L) and ammonia concentration (from 2.6 mg/L to 0.1 mg/L), probably due to the formation of an insoluble magnesium ammonium complex as documented in previous studies (Ohtman et al., 2010). Further ozonation of the WAS on the other hand, resulted in the release of both NH_4^+ and PO_4^{3-} as a result of sludge solubilization. As a result of the combined treatment, the amount of the overall soluble PO_4^{3-} concentration (2.5 mg/L) of the WAS was remarkably lower than that of the initial concentration that was measured prior to pretreatment. Therefore, it can be concluded that the precipitation of the solubilized phosphate can increase the fertilizer value of the WAS. Similarly, the N_M of the treated manure, which is the sum of NH_3-N and NO_3^--N, represents the portion of manure that can be utilized by plants.

Conclusion

The chemical oxidation processes, ozonation and Fenton, constitute promising technologies for the removal of recalcitrant antibiotics from solid matrices such as manure and WAS and therefore can decrease the risk of antibiotic pollution resulting from utilization of manure and sludge as soil amendments. The application of a pretreatment prior to the oxidation processes provided important advantages such as increased oxidant contaminant contact and phosphorus recovery from the WAS. It is anticipated that both manure and sludge fertilizer values will be improved by the combined processes of pretreatment and ozonation or Fenton oxidation.

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Studies on Diffusion of Allelopathıc Compounds of Paddy Weeds and Their Allelopathıc Toxicity

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Abstract
Allelopathy can be defined as an important mechanism of plant interference mediated by the addition of plant-produced secondary products to the soil rhizosphere. Allelochemical substances released from the plant by four general routs like leachates, decomposition, volatilization and root exudation. In this experiment, the effect of volatile compounds of fresh plant part of paddy weeds viz. C*yperus difformis*, *Echinochloa crusgalli*, *Paspalum paspaloides* and *Sagittaria trifolia* in surface sterilized Desicator for 48 h on radicle and hypocotyl length of rice seedling and seed germination of Neda variety in soil and water were assayed. Results of volatilization bioassay of *Cyperus difformis* revealed that both radicle and hypocotyl length of paddy seedling as well as seed germination of test crop were significantly hampered by water kept with fresh plant materials of mentioned weed in airtight jar than soil which was not hampering on these parameters. Soil kept with fresh plant parts of the other weeds had no significant inhibition effect on radicle and hypocotyl length and seed germination except *Echinochola crusgalli* which significantly hampered on length of seedling about 37.2% only. In other method of study about water kept with plant material of these weeds was hampering on length of seedling in great extent. By preliminary phytochemical method, the secondary compound like terpenoids in root, stem, leaf and other parts of these weeds were detected. This study showed that the volatile compounds from studied weeds will be diffuse into distilled water more than soil solution.
Keywords: Weeds, volatile compounds, terpenoids, Soil and water

Introduction

Allelopathy can be defined as an important mechanism of plant interference mediated by the addition of plant-produced secondary products to the soil rhizosphere. Allelochemical substances released from the plant by four general routs like leachates, decomposition, volatilization and root exudation. Volatile toxins from the plants release into the atmosphere (Muller, 1966). Phenomenon of volatilization is too much significant under arid or semi- arid conditions than moister area. The compounds may be absorbed in vapour by surrounding plants be absorbed from condensate in dew or may reach the soil and be taken up by the roots (Rice, 1984 and Putnam, 1985).
In this experiment, the effect of volatile compounds of fresh plant part of paddy weeds viz. *Cyperus difformis* L., *Echinochloa crusgalli* (L.) P. Beauv., *Paspalum paspaloides* (Michx.) Scribner and *Sagittaria trifolia* L. in surface sterilized air tight glass jar (Desicator) for 48 h on radicle and hypocotyl length of rice seedling and seed germination of Neda variety in soil and water were assayed.

Material and methods

To test volatile components, are diffuse from the studied weed species, if any involved in the inhibitory effect on neighbouring crop plants, fresh weed species were collected from different farm of paddy fields. The samples were collected at full flowering stage. Then fresh plant parts were made clean with brush to remove soil and dust particles. From each plant species fresh parts (150 g) were kept in surface sterilized air tight glass jar for 48 h. Soil (150 g) and water (150 ml) were kept inside the glass jar separately in such a way that they are not indirect contact with plant materials, and the air inside only acts as a carrier for volatile substances (Avchar and Deokule, 2007). The soil and water, thus obtained were used for further bioassay using test crop. For laboratory bioassay, 10 healthy surface sterilized seeds of each variety were placed in sterilized petridish (10 cm diameter) containing a Whatman No.1 filter paper. 10 ml of the water obtained from glass jar was utilized to moisten the filter paper, and 10 g soil obtained from glass jar was used as a layer and made wet with 10 ml distilled water. Soil sample (free from growth of experimental weed species) from adjacent fields or sites in the same fields where the experimental

plants were not present was used as control soil (El Khatib, 1998) at the rate of 10 g per petridish. The soil was prepared with intensive attention of weeds through Rice Research Institute (RRI) of Amol (Iran). The triplicate of each dish were made under laboratory conditions (room temperaturemaximum temperature during mid day: 25 to 28^{o}C) for seven days (Maharjan et al. 2007). All the petridish were wrapped in brown paper to avoid direct light. The slight emergence of radicle was considered as a sign of germination. On 8th day germination percentage was determined and root and hypocotyl length in cm was measured.
Preliminary phytochemical method was used for detection and confirmation of Terpenoids in root, stem and leaf of studied weeds by using standard method of Trease and Evans,1989 and Harborne,1973 c.f. Edeoga et al. 2005. Student's t- test was used for comparison of two means.

Result and Discussion

Results of volatilization bioassay of *Cyperus difformis* (in Tables 1 and 2) revealed that radicle and hypocotyl length of paddy seedling as well as seed germination of test crop were significantly hampered (t- test, $p < 0.05$) by water kept with fresh plant material of above weed in airtight jar than soil which was not hampering on radicle and hypocotyl as well seed germination of test crop.

Table 1. Effects of volatilization bioassay from *Cyperus difformis* (water) on seedling growth and seed germination of Neda variety.

Type of bioassay	Radicle length (cm)	Hypocotyl length (cm)	Seedling growth (cm)	Seed germination (%)
Control	4.55±0.18	4.16±0.75	8.71±0.93	90.00±10.00
Treated water	1.10±0.78	0.58±0.39	1.68±1.16	33.33±15.27
p- value	0.013	0.014	0.012	0.019

Table 2. Effects of volatilization bioassay from *Cyperus difformis* (soil) on seedling growth and seed germination of Neda variety.

Type of bioassay	Radicle length (cm)	Hypocotyl length (cm)	Seedling growth (cm)	Seed germination (%)
Control	4.33±0.17	3.16±0.27	7.50±0.40	83.33±5.77
Treated water	4.14±1.00	2.69±0.38	6.83±1.35	76.67±15.27
p- value	0.759	0.161	0.464	0.520

Volatilize secondary compound like terpenoids in root, stem and leaf of studied weeds were detected. Perhaps this chemical more absorbed by water which is responsible for significant allelopathic effects.

In case of *Echinochloa crusgalli*, water kept with fresh plant parts in airtight jar significantly hampered on both radicle and hypocotyl length of test crop (t-test, $p < 0.05$). It was also showed that germination percentage of test crop was not significantly hampered from that collected water in airtight jar. In other method of study, soil kept with plant material in airtight jar significantly hampered on radicle length (t-test, $p < 0.05$) as compared with hypocotyl length of paddy seedling which was not affected. However, the negative impact of that collected soil on radicle length caused a decrease on seedling growth of test crop. It was also recorded that seed germination was not sensitive from that collected soil from airtight jar. However, seedling growth of test crop was more sensitive to water than soil about 48.22 and 37.2 % respectively (Tables 3 and 4).

Table 3. Effects of volatilization bioassay from *Echinochloa crusgalli* (water) on seedling growth and seed germination of Neda variety.

Type of bioassay	Radicle length (cm)	Hypocotyl length (cm)	Seedling growth (cm)	Seed germination (%)
Control	4.55±0.18	4.16±0.75	8.71±0.93	90.00±10.00
Treated water	2.62±0.57	1.89±0.96	4.51±1.53	80.00±10.00
p- value	0.018	0.046	0.029	0.292

Table 4. Effects of volatilization bioassay from *Echinochloa crusgalli* (soil) on seedling growth and seed germination of Neda variety.

Type of bioassay	Radicle length (cm)	Hypocotyl length (cm)	Seedling growth (cm)	Seed germination (%)
Control	4.33±0.17	3.16±0.27	7.50±0.40	83.33±5.77
Treated soil	2.70±0.52	2.01±0.89	4.71±1.32	73.33±15.27
p- value	0.020	0.109	0.039	0.352

Results of volatilization bioassay of *Paspalum paspaloides* showed in Tables (5 and 6) indicated that water kept with fresh plant parts airtight jar significantly affected on radicle (t- test, $p < 0.01$) as well as hypocotyl length (t- test, $p < 0.05$) of test crop. It was also recorded that seed germination was not remarkable affected. In case of soil kept with plant material in airtight jar had a weak effect on radicle and hypocotyl as well as percentage of germination of paddy seeds.

Table 5. Effects of volatilization bioassay from *Paspalum paspaloides* (water) on seedling growth and seed germination of Neda variety.

Type of bioassay	Radicle length (cm)	Hypocotyl length (cm)	Seedling growth (cm)	Seed germination (%)
Control	4.55±0.18	4.16±0.75	8.71±0.93	90.00±10.00
Treated water	2.41±0.07	1.72±0.56	4.13±0.62	63.33±11.55
p- value	0.009	0.024	0.014	0.052

Table 6. Effects of volatilization bioassay from *Paspalum paspaloides* (soil) on seedling growth and seed germination of Neda variety.

Type of bioassay	Radicle length (cm)	Hypocotyl length (cm)	Seedling growth (cm)	Seed germination (%)
Control	4.33±0.17	3.16±0.27	7.50±0.40	83.33±5.77
Treated soil	3.03±0.97	2.51±0.45	5.55±1.40	76.67±11.55
p- value	0.096	0.109	0.092	0.424

Data analysis of volatilization bioassay from fresh plant parts of *Sagittaria trifolia* (in Tables 7 and 8) revealed that radicle and hypocotyl length of paddy seedling exclusively was hampered (t-test, $p < 0.05$) by water kept with fresh plant parts in airtight jar than seed germination which was not significantly hampered. In other method of study about soil was not hampering on radicle and hypocotyl length of seedling as well as seed germination of test crop.

Table 7. Effects of volatilization bioassay from *Sagittaria trifolia* (water) on seedling growth and seed germination of Neda variety.

Type of bioassay	Radicle length (cm)	Hypocotyl length (cm)	Seedling growth (cm)	Seed germination (%)
Control	4.55±0.18	4.16±0.75	8.71±0.93	90.00±10.00
Treated water	2.95±0.71	2.22±0.15	5.17±0.86	70.00±10.00
p- value	0.034	0.026	0.022	0.082

Table 8. Effects of volatilization bioassay from *Sagittaria trifolia* (soil) on seedling growth and seed germination of Neda variety.

Type of bioassay	Radicle length (cm)	Hypocotyl length (cm)	Seedling growth (cm)	Seed germination (%)
Control	4.33±0.17	3.16±0.27	7.50±0.40	83.33±5.77
Treated soil	3.45±1.04	2.34±1.12	5.79±2.14	90.00±10.00
p- value	0.227	0.291	0.252	0.377

This study showed that the volatile compounds from studied weeds will be diffuse into distilled water more than soil solution.

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INVESTIGATION OF BIOSORPTION FOR ACETYLSALICYLIC ACID BY LIVE ACTIVATED SLUDGE

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Abstract
The biosorption of acetylsalicylic acid (ASA) from aquatic solution on live activated sludge was studied in a batch system. The acetylsalicylic acid biosorption was attained 10 minutes. The Langmuir, Freundlich and Tempkin isotherms models were applied to the experimental data and isotherm constants were calculated. Equilibrum data fitted better to the Freundlich model than the others models. Gibbs free energy values were calculated and it is concluded that the biosorption of acetylsalicylic acid from aquatic solution on live activated sludge was endotermic in nature. Octanol-water partition coefficient, $K_{ow,}$ and biosorption coefficient, K_d, which describes the solid liquid partitioning characteristics of a compound in biosorption mechanism were also calculated.
Keywords: Anti-Inflammatory Pharmaceuticals, Abiotic Loses, Isotherms, Activated Sludge.

1. Introduction
The release of pharmaceutical substances and their metabolites into the aquatic environment has become an increasing concern over recent years. Hundreds of tonnes of pharmaceuticals enter wastewater treatment plants each year. Previous studies show that, pharmaceuticals and their metabolites are poorly removed during wastewater treatment. As a result they are detectable in wastewater treatment plants effulents, rivers, lakes, seas and rarely in groundwater (Heberer, 2002, Calamari et al, 2003, Miao et al, 2002).
Acetylsalicylic acid known by trade name of Aspirin. Aspirin is used to relieve pain, fever, and inflammation in various conditions such as the flu, common cold, headache, migraines, nerve pain, toothache and muscle pain. More than 10.000 tons of aspirin are consumed in the United States each year (Insel et al, 1990). Below, the chemical structures of the acetylsalicylic acid is shown.

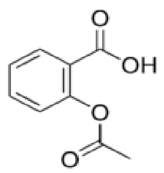

Fig. 1. Chemical structures to Acetylsalicylic acid

Acetylsalicylic acid is the salicylate ester of acetic acid. It is the pro-type of the salicylates and is a non-steroidal anti-inflammatory agent (NSAIA). The drug is hydrolysed in vivo to give salicylic acid in the stomach, which is its active form. Then salicylic acid is quickly absorbed and is responsible for the effects of aspirin (Sagar et al, 1999; Lutomsky et al, 1995). The purpose of this work is to determine adsorption properties of live activated sludge for the uptake of acetylsalicylic acid from aqueous solution. In this way, kinetic and equilibrium biosorption data were obtained and the effect of several sludge dose (0.5 g and 1.0 g) in the biosorption process was evaluated.

2. Materials and Methods
The activated sludge was collected from the full scale activated sludge plant of Pepsi Soft Drink Filling Industry, Adana, Turkey. The biosorbent was used on the same day as it was sampled. Total suspended solids (TSS) were measured by the standard gravimetric technique (Standard Methods, 1998). Test solutions containing acetylsalicylic acid were prepared by fresh stock acetylsalicylic acid solution which was obtained by dissolving weighed quantity of aspirin in methanol and distilled water. Some physical and chemical properties of acetylsalicylic acid used in this work are summarized in Table 1.

Table1. Some physical and chemical properties of acetylsalicylic acid

Drug class	Analgesics
CAS Number	50-78-2
Chemical name	Benzoic acid, 2-(acetyloxyl)-Salicylic acid acetate
Chemical Formula	$C_9H_8O_4$
Molecular weight (g/mol)	180.16
Solubility in water (mg/L)	3300
Melting point (^{0}C)	136
Plasma half life [h]	300–650 mg/day 3.1–3.2 sa 1 g/day 5 sa
UV Spectrums (Wavelenght) (λmax) nm	200
Solubility	Soluble in water
Physical appearances	White crystalline
Odor	Odorless
Metabolites	Salicylic acid

The sorption tests were conducted in a routine manner by a batch technique at 25 ^{0}C. The activated sludge (62.50 ml) was added to aqueous soluions (62.50 ml) of acetylsalicylic acid. Volume of final mix was adjusted to 125 ml containing 4000 mg/L activated sludge (0.5 g). Accordind to second study the activated sludge (125 ml) was added to aqueous (125 ml) of acetylsalicylic acid. Volume of final mix was adjusted to 250 ml containing 4000 mg/L activated sludge (1.0 g). The data for deriving the isotherms constant were obtained by using sludge (0.5 g and 1.0 g) and acetylsalicylic acid concentrations of 25, 50, 100 and 200 mg/L. The contact time was 160 min. Before analysis the samples were centrufuged at 6000 rpm for 20 min and the supernatant liquid was analysed for the remaining acetylsalicylic acid. All the experiments were carried out in duplicates. The final concentration of acetylsalicylic acid in solution was measured using an UV-Vis spectrophotometer Perkin Elmer at a wavelength of 200 nm. The amount of acetylsalicylic acid biosorbet onto activated sludge biosorbent, qe (mg g^{-1}), was calculated by a mass balance relationship as follows:

$$q_e=(C_0-C_e)\ \frac{V}{W}$$

where C_0 and C_e are the initial and equilibrium liquid-phase concentration of acetylsalicylic acid, respectively (mg l^{-1}), V the volume of the solution (l) and W is the dry weight (g) of activated sludge.

3. Results

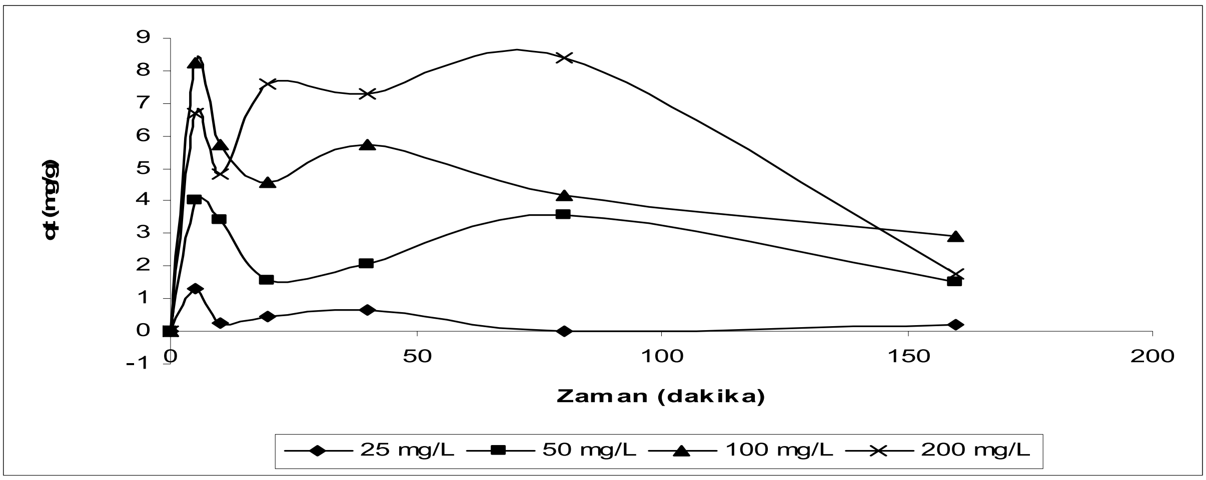

Figure 1. Changing of specific adsorption results for various acetylsalicylic acid concentration (0.5 g adsorbent)

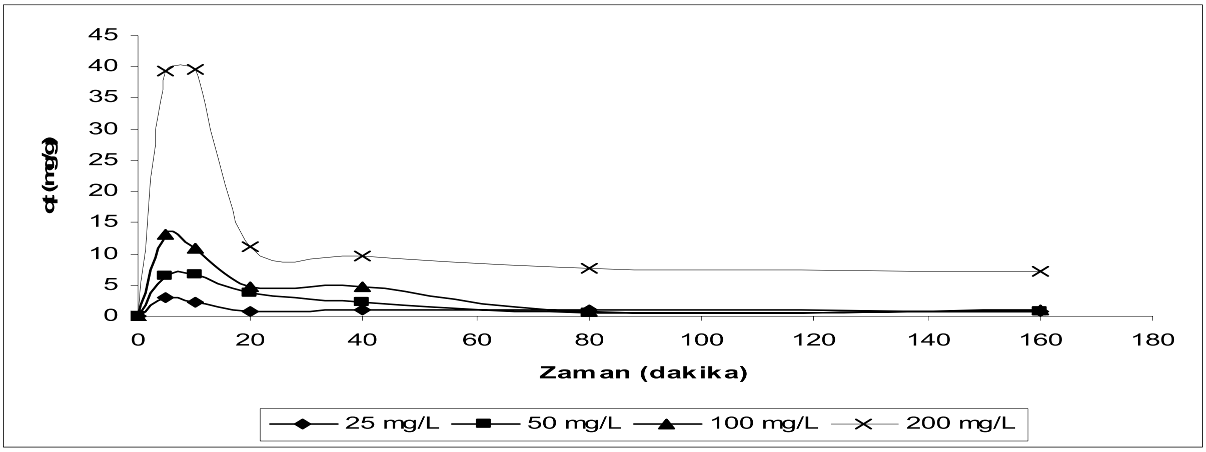

Figure 2. Changing of specific adsorption results for various acetylsalicylic acid concentration (1.0 g adsorbent)

Table 2. Applied isotherm models and their linear forms

Isotherm		Linear form	plot
Langmuir 1	$qe = \frac{q_m K_a C_e}{1+K_a C_e}$	$\frac{C_e}{q_e} = \frac{1}{q_m} C_e + \frac{1}{K_a q_m}$	$\frac{C_e}{q_e}$ vs. C_e
Langmuir 2		$\frac{1}{q_e} = \left(\frac{1}{K_a q_m}\right)\frac{1}{C_e} + \frac{1}{q_m}$	$\frac{1}{q_e}$ vs. $\frac{1}{C_e}$
Langmuir 3		$q_e = q_m - \left(\frac{1}{K_a}\right)\frac{q_e}{C_e}$	q_e vs. $\frac{q_e}{C_e}$
Langmuir 4		$\frac{q_e}{C_e} = K_a q_m - K_a q_e$	$\frac{q_e}{C_e}$ vs. q_e
Freundlich	$qe = K_f C_e^{1/n}$	$\log(q_e) = \log(K_f) + 1/n\log(C_e)$	$\log(q_e)$ vs. $\log(C_e)$
Tempkin	$q_e = \frac{RT}{b}\ln(K_t C_e)$	$q_e = B\ln K_T + B\ln C_e$	q_e vs. $\ln C_e$

Table 9 The Parameters Obtained From The Isotherm Models For acetylsalicylic acid Biosorption (0.5 g)

Isotherm	Parameters	Values	Equations
Freundlich	n	1.0285	y =0.9722x-1.7222
	K_f (mg/g)(L/mg)$^{1/n}$	0.0189	
	r	0.769	
Langmuir 1	q_m (mg/g)	5.461	y = 0.1831 x-54.715
	K_a (L/mg)	0.0182	
	r	0.3059	
Langmuir 2	q_m (mg/g)	-1.351	y =116.47x-0.74
	K_a (L/mg)	-0.0063	
	r	0.915	
Langmuir 3	q_m (mg/g)	0.5421	y =52.556x+0.5421
	K_a (L/mg)	-0.0190	
	r	0.616	
Langmuir 4	q_m (mg/g)	-1.194	y = 0.0072x+0.0086
	K_a (L/mg)	-0.0072	
	r	0.616	
Tempkin	B	0.852	y = 0.8523x-1.9904
	K_T	0.096	
	r	0.688	

Table 9 The Parameters Obtained From The Isotherm Models For acetylsalicylic acid Biosorption (1.0 g)

Isotherm	Parameters	Values	Equations
Freundlich	*n*	1.00715	y = 0.9929x-1.6798
	K_f (mg/g)(L/mg)$^{1/n}$	0.0209	
	r	0.799	
Langmuir 1	q_m (mg/g)	12.468	y =-0.0802 x+65.329
	K_a (L/mg)	0.0153	
	r	0.1503	
Langmuir 2	q_m (mg/g)	1.926	y =23.84x+0.5191
	K_a (L/mg)	0.021	
	r	0.680	
Langmuir 3	q_m (mg/g)	-1.5255	y =165.88x-1.5255
	K_a (L/mg)	-0.00602	
	r	0.713	
Langmuir 4	q_m (mg/g)	-5.258	y =0.0031x+0.0163
	K_a (L/mg)	-0.0031	
	r	0.713	
Tempkin	*B*	2.758	y = 2.7589x-9.237
	K_T	0.035	
	r	0.769	

Table Results from Langmuir Model to the equilibrium data of the adsorption of salicylic acid onto the adsorbents used (Otera, M., 2004)

Langmuir 1	Filtrasorb F400	Sephabeads SP207	Sephabeads SP206
q_m (mg/g)	351.0	81.6	45.2
K_a (L/mg)	1.85 10^{-5}	2.03 10^{-7}	1.27 10^{-8}

It is well known that Gibbs free energy would define system non spontaneity. Gibbs free energy (ΔG) can be calculated from the following equlations:

$$K_c=C_a/C_e$$

$$\Delta G = -RT\ln K^0_e$$

where K_c is the equilibrium constant, K^0_e is the thermodynamic equilibrium constant C_a is the equilibrium concentration in solution (mg/L) R is the universal gas constant 8.314 J/mol, and T is temperature (K). Thermodynamic equilibrium constant (K^0_e) can be calculated from the equilibrium constant by plotting equilibrium constant against initial ASA concentration (Aksu Z., 2002).

Table 3 Comparison of gibbs free energy values for various system

Sorbent	Drug	ΔG (kj/mol)	T (K)
Aktivated Sludge (0.5 g) (This work)	ASA	7.580	298
Activated Sludge (1.0 g) (This work)	ASA	18.380	298
Kaolin (0.25 g)[1]	Metmorfin HCl	3.43	310.50
Attapuligate (0.25 g)[1]	Metmorfin HCl	1.017	310.50

Table 3 shows the comparision of Gibbs free energy values adsorbents. The positive ΔG values confirm the nonspontaneous nature of adsorption process, there is some sort of interactions but weak interactions (Al-Bayati, 2010). Sorption ability can be estimated from the sorption coefficient (K_d) that mainly depends on the properties of the both acetylsalicylic acid and sludge. Following equation is given for the calculation of K_d; $K_d= q_e/C_e$ where; q_e is the equilibrium solid phase concentration (mg/g), and Ce is the equilibrium concentration of acetylsalicylic acid in solution after biosorption (mg/L) and Kd sorption coefficient (L/g). Several studies report expressions where K_d is estimated direcly from the octanol-water partition coefficient (K_{ow}). Octanol-water partitioning coefficient can be calculated using following equation. $K_{ow}= C_o/C_w$ where; C_o : concentration of compounds at octanol phase (mg/L), C_w : concentration of compounds at water phase (mg/L). The effectiveness of sorption can be stated using the octanol water partition coefficient (K_{ow}) (Jones et al 2007; Rogers, 1996).

1) If log K_{ow} is less than 2.5, the compound has a low adsorption potential.
2) If log K_{ow} is between 2.5 and 4 the compound has a medium adsorption potential
3) If log K_{ow} is greater than 4, the compound has a high adsorption potential
According to Table 9, ASA has low adsorption potential by live activated sludge.

Table 5. Experimentally obtained K_d values (0.5 g sorbent)

Initial concentrations (mg/L)	K_d (L kg^{-1})	K_d (average)
25	9.028	
50	32.114	
100	30.938	
200	9.003	20.271

Table 6. Experimentally obtained K_d values (1.0 g sorbent)

Initial concentrations (mg/L)	K_d (L kg^{-1})	K_d (average)
25	31.578	
50	15.184	
100	9.154	
200	38.551	23.617

Tablo 7 Some reported K_d values for various pharmaceuticals

Adsorban/Adsorbat	K_d (L kg^{-1})	Referances
Soil Ferralsol 12/ Salicilic Acid	26.04	Dubus et al, (2001)

Table 8 Equations developed based on relations between K_d and K_{ow}

Equations	References
$\log K_d = 0{,}58 \log K_{ow} + 1{,}14$	Dobbs et al., 1989
$K_d = 0{,}39 + 0{,}67\ K_{ow}$	Matter-Müller et al., 1980
$K_d = 0{,}33453 * K_{ow}$	Karickhoff. 1984
$K_d = 0{,}1435 * K_{ow}$	Lauridsen et al., 2000

Table 9 Values of K_{ow} and Log K_{ow}

System	LogKd=0.58LogKow+1.14		Kd=0.39+0.67 Kow		Kd=0.33453*Kow		Kd=0.1435*Kow	
	Kow	Log Kow	Kow	Log Kow	Kow	LogKow	Kow	LogKow
ASA-Activated Sludge (0.5 g)	0.481	-0.317	12.892	1.110	26.98	1.431	62.91	1.798
	4.285	0.632	47.349	1.675	95.99	1.982	223.7	2.349
	4.017	0.604	45.594	1.658	92.48	1.966	215.5	2.333
	0.478	-0.320	12.855	1.109	26.91	1.429	62.73	1.797
Average	**0.516**	**0.287**	**29.673**	**1.472**	**60.59**	**1.782**	**141.2**	**2.150**
ASA-Activated Sludge (1.0 g)	4.159	0.619	46.549	1.667	94.39	1.974	220.0	2.342
	1.177	0.071	22.080	1.344	45.38	1.656	105.8	2.024
	0.931	-0.0307	13.080	1.116	27.36	1.437	63.79	1.804
	5.874	0.769	56.956	1.755	115.2	2.061	268.6	2.429
Average	**2.523**	**0.402**	**34.667**	**1.539**	**70.59**	**1.848**	**164.5**	**2.216**

Table 10. Some reported Log K_{ow} values for pharmaceuticals in literature

Pharmaceuticals	Log K_{ow} value	References
Salicylic acid	2.26	Dubus et al, (2001)
Carbamazepine	2.25	Scheytt et al., (2005)

According to some Log Kow values for acidic pharmaceuticals by various researcher are given at Table 10. Values for salicylic acid and carbamazepine were found between 2.26 and 2.25 respectively. Hence acetil salicylic acid has low adsorption potential by activated sludge and also these were in good agreement reported at literature.

4.Discussion

From the work presented here following conclusions can be drawn;

- ASA was adsorbed by live activated sludge to some degree,
- Biosorption was fast completed with in 10 minutes
- Based on the K_d and K_{ow} values, it is possible to say that biosorption of ASA by live activated sludge was not high.

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The Effects of Cadmium Contaminations on the Properties of *Amaranthus retroflexusL.*

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Abstract
The aim of phytoremediation is remove heavy metals from environment or reduce pollution. It is a green revolution. In this research, Cadmium was added to soil as cadmium chloride in concentration (0, 20, 40, 60, 80, and 120). The plant was maintained in greenhouse under controlled circumstances. Then its effects were evaluated on Amaranthus retroflexus L. After harvest, cadmium content of the plant and soil was measured by ICP. Results of the experiment indicated that toxicity of cadmium was lead to reduce the plant growth in cadmium concentrations more than 10 mg/ kg soil. The Cd concentration was increased with cadmium contents in soil and the maximum accumulation occurred in the roots. High accumulation of Cd by this halophyte reveals that Amaranthus retroflexusL. can be considered as a suitable plant for phytoremediate Cd-contaminated soils, despite the fact that its biomass decreases by increasing soil cadmium.
Keywords: Amaranthus retroflexus , phytoremediation , Pollution , contaminated

Introduction
From a global perspective of climate, soil crust, the third major component of the human environment is considered today as a non-renewable resource is being destroyed. Accumulation of heavy metals in the soil of human concern in the present century is It can affect plant growth, health and environmental quality of agricultural products may have adverse effects (Ferguson, 1990).Human interference and moving of heavy metals in the geochemical cycles of these metals in the biosphere is an important process Release in the environment that they are faster than normal (Anthology, 1384).Biological accumulation of heavy metals in the food chain can be dangerous for human health (okrono, 2005). Heavy metals are usually two inhalation and feeding to humans, are entered. Today's world can be inspired by nature and its pristine inevitable failure of reforms in order to eliminate the dust pollution.Pollution refining technologies revolving around the plant, called "phytoremediation" or "green refinery" plants using And positive interaction and help with soil organisms Contaminants from soil and groundwater will be refined in situ. Cadmium is an element of environmental pollutants. Cadmium with the usual 0.6 to 1.1 mg. Kg is movable element in heavy soil, the element for humans and plants is not necessary, and it hurt for plant growth is not, but for humans and animals, very deadly. So be it from entering the environment and food chain to stop. Losses and damages resulting from the use of cadmium in humans with high blood pressure, lung cancer, kidney damage, nerve pain and heart disease is spread and ... . Has been associated .Phytoremediation is a technique economical, environmental, and scientific, which is very suitable for developing countries, unfortunately, despite these potential remains in some countries as a technology, not commercial use. The technology of green plants and its relationship with soil microorganisms are used to reduce pollution.
According to general principles of different methods of refining plant to be one of the best methods, is plant consolidation phytoextraction in this way the plant to absorb, concentrate and Sequestration of toxic metals from contaminated soils in populations of plant shoots and roots are used. To achieve a good result in phytoremediation techniques should be used simultaneously for several factors, including 1 - more than the desired metal accumulation of plants. 2 - High level of plant biomass. 3 - Transfer of metals from root to shoot is at a high level.
Cadmium uptake by plant roots to the environment and the chemical species of cadmium in the soil depends. In general, any linear relationship between concentration of cadmium in the body and the surrounding plant roots there.

Materials and Methods

We are a species of plants *Amarantus Retroflexus* that they are drought tolerant plants and halophytes as a suitable plant for phytoremediation cadmium are introduced. Therefore, some selected soil characteristics we consider the following table :(table 1)

Table 1 Analysis of the soil before testing and contaminated by cadmium salts.

EC of water Mmho /cm	Acidity Irrigation water	levels of cadmium in water	levels of cadmium in soil	Soil bulk density g/cm3	%CaCo3	CEC	% N	Organic carbon.	EC of soil Mmho /cm	Soil acidity	Soil
1.2	7.4	0.013	1.22	1.334	7.5	14	0.07	0.7	6.71	7.58	Sandy clay loam

EC = Electrical conductivity of water Mmho /cm (millimhos per centimeter)
Acidity Irrigation water = Adjusting the pH of Irrigation Water
Levels of cadmium in water = the initial concentration of cadmium in water Mg /liter (milligrams per liter)
Levels of cadmium in soil = the initial concentration of cadmium in soil Mg/ kg (milligrams per kg)
CEC = Caption exchange capacity (maximum quantity of total captions) (cmol+/kg)
% N =Percent OF Nitrogen
EC = Electrical conductivity of soil Mmho /cm (millimhos per centimeter)
Soil acidity = Adjusting the pH of Irrigation soil

In this method, phytoremediation of contaminated soil must be at least 2 months before planting. Certain amount ($Cdcl_2H_2o$) to help the spray to the soil and mix it thoroughly, we Permitted concentration of cadmium in the range of 0.6 to 5 mg kg. And contaminated samples with a concentration of zero, 20, 40, 60, 80, 120 mg. kg of cadmium should be considered for work. In this study, height 20 cm pots with three replicates for each concentration of use..The contaminated soil, especially the apparent mass density of the compacted soil in the pot, we have studied. *Amarantus Retroflexus* seeds after planting, the pots kept in controlled conditions after the completion of the course, we do take samples from various organs of the plant. To determine the pollutant concentrations in the ppb range, the washing with distilled water, measuring plant fresh weight, plant height, diameter and amount of chlorophyll a and chlorophyll b will have. The samples in special bags, dried in a dryer with a temperature of 70 ° C are. After grinding, the concentration of cadmium in plants and oxidation method with ICP-OES measurements can be obtained from plant samples. The data obtained in factorial experiments with randomized complete block design with software Spss done. To determine the amount of soluble cadmium in the soil (soil sample pots) to 10 g of soil sample, 25 mg solution of 0.0050 molar EDTA added, and 16 hours in centrifuges used to extract smooth and cadmium with atomic absorption measurements are. at the end of the data obtained from experiments, the *Amarantus Retroflexus* pigweed plants for the refining plant is referred to cadmium-contaminated soils.

Results

Analysis of physical and chemical soil showed that the salinity of soil saturation extract used in this experiment, 6.7 dS/ m which the amount of salt for culturing four species studied, the salty taste being the problem not the cause will not yield and electrical conductivity that is suitable for their cultivation. The soil PH test with 7.5 which is popular passion for plants. Sand, silt and clay, respectively, 50%, 24% and 26% was thus textured is a sandy clay loam. This soil is fairly light and allows proper root growth and distribution provides. The soil used in this study, soil is with low organic matter (0.7%).Drip irrigation soil moisture during the experiment by approximately "was kept constant and equal to field capacity. The initial concentration of cadmium in soil samples with 1.18 mg/ kg was compared to the allowable concentration of cadmium in the soil are allowed.

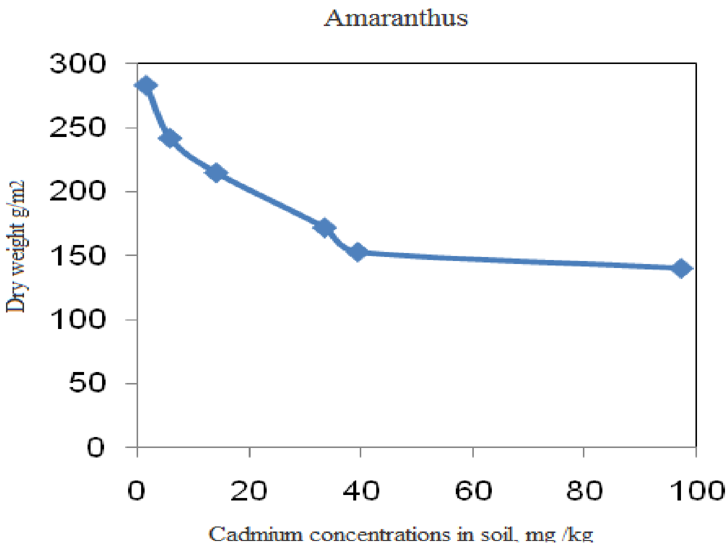

Figure 1. Linear chart the changes between *Amarantus Retroflexus* dry weight (g/m2) of soil cadmium concentrations in (mg/kg)

Cadmium concentration in soil, plant dry weight decreased to 39.475 up to concentrations of cadmium concentration in soil and plant dry weight is constant. (Figure1)

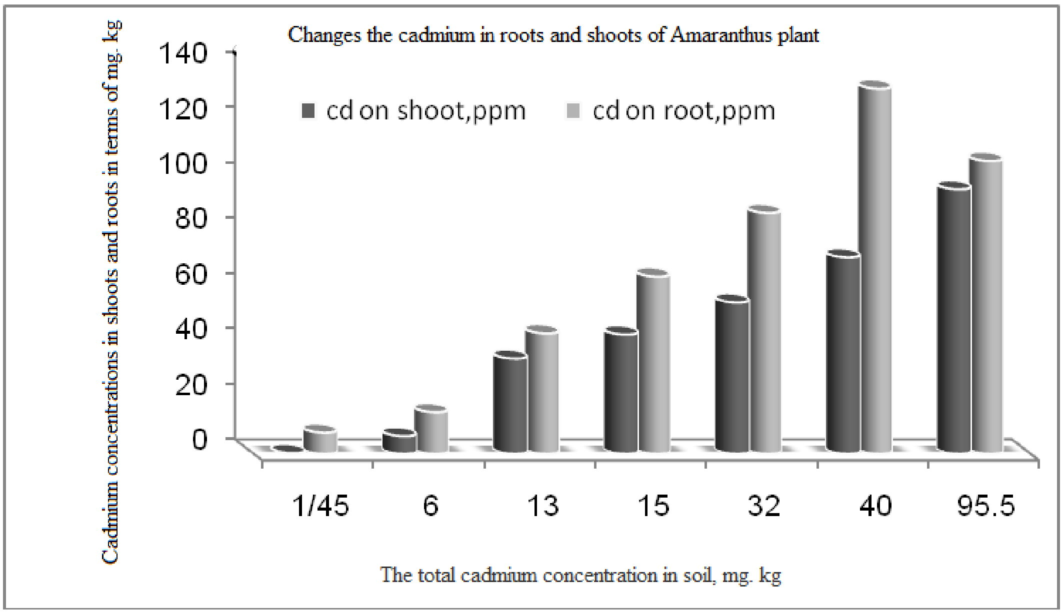

Figure 2. Bar graph of the changes in shoots and root cadmium concentrations in (mg.kg) of total cadmium concentration in (mg.kg) of soil

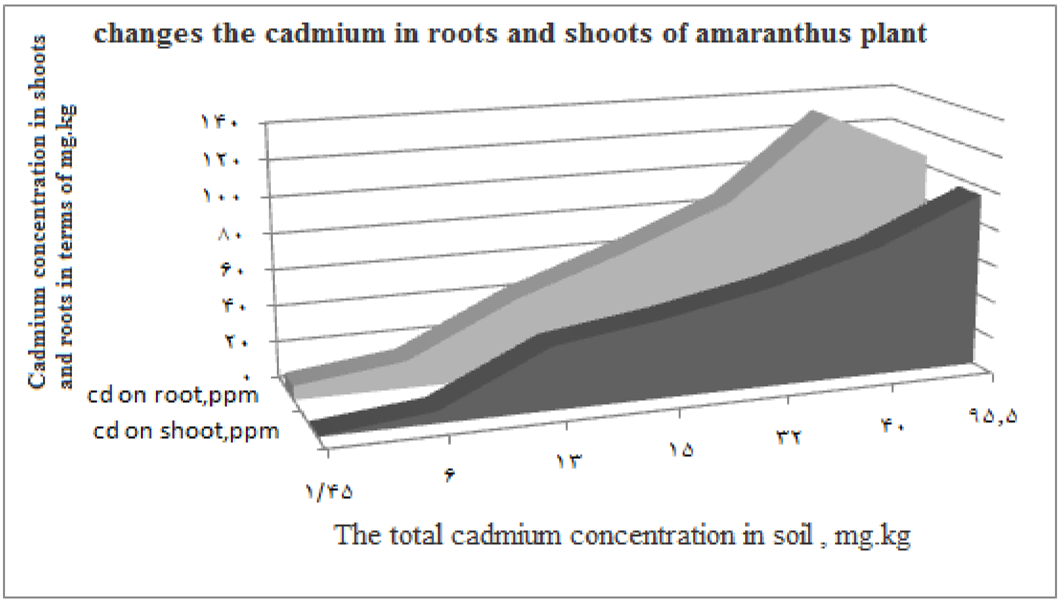

Figure 3. Area graph of the changes in shoot and root cadmium concentrations in (mg.kg) of total cadmium concentration in (mg.kg) of soil

Given the above graph it can be said with regard to increasing concentrations of cadmium in the soil, the concentration of cadmium is absorbed in the roots more than shoots. (Figure 2, Figure 3)

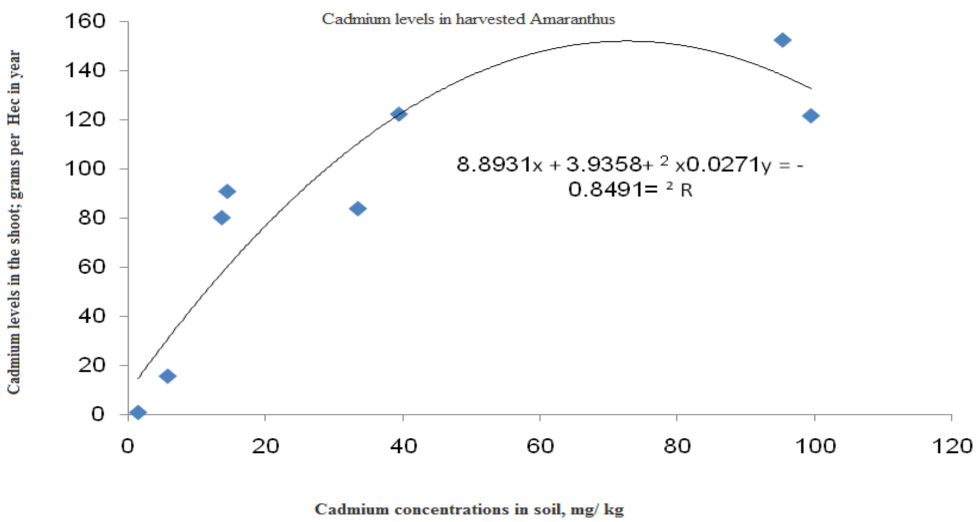

Figure 5. Linear chart changes in the amount of cadmium (g.hectare) of shoot cadmium concentration in soil (mg/ kg)

According to this chart due to the high concentration of cadmium in the soil, the amount of cadmium in shoot to concentrations ranging between" 70 to 80" increases the concentration, but will then decline. (Figure 5)

Acknowledgements
Praise God and thanks to his boundless grace believers who gave it to us so we can succeed in order to fix the problem of soil contamination problems and the promotion of agricultural knowledge and provide a healthy environment and with the coming freshness. Thanks to my parents who grew up and prospered View Post Originally I had helped my talent. In the end, I dedicate this paper to my homeland, Iran, and I thank my dear colleague Mrs. M. Mazhari doctor, engineer Mehdi Khosravi, which was my main incentive in this area. Finally, he expressed thanks and appreciation to my very own.

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Impact of Different Kind of Pig Manure on Organic Matter Functional Groups under Semi-arid Climate Conditions in Albacete Region SE Spain

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Abstract

Spain is the second largest pig producer in Europe, therefore managements of the waste is very important in terms of environmental pollution and safety. The nature and properties of animal manure change with species, age, weight, feeding diet e.g. physical-chemical process on these manures could change many properties. Annual application of pig manure to a farmer's field in southeastern Spain caused improved soil physical and chemical properties. The effects of manure addition on the chemical nature of soil organic matter functional groups of organic matter (OM) are investigated by solid-state ^{13}C nuclear magnetic resonance (NMR) spectroscopy. The objectives of this study are to clarify the effect of different kind of PS application on the changes of structural characteristics of organic matter using ^{13}C NMR, which provided new information for improving soil fertility by PS application. The ^{13}C NMR spectroscopy's results showed that all pig slurry application are decreased the carboxyl and increased aromatic groups of the OM. Raw pig slurry from liquid feeding diet increased alkyl group of OM and different feeding diet types differently affected on O-Alkyl group. Solid state ^{13}C-NMR spectra of OM of the thirteen surface soils, representing four different pig slurry applications and under similar vegetation display similar distribution of functional groups, so respectively alkyl>O-alkyl>aromatic>carboxyl. The influence of pig slurry is significant in terms of aromatic and phenolic groups.

Keywords: ^{13}C-NMR Spectrophotometry, Organic Matter, Pig Manure, SE Spain.

Introduction

Soil organic matter is very important in terms of physical, chemical and biological soil properties which, is very essential source of nutrients for plant and microorganism. Countries under the semi-arid Mediterranean climate zone, such as Spain, applied organic matter decomposes very rapidly in the soil with biological activities. Therefore, identification of chemical structure of organic matter is very important to management of the soil. Solid-state ^{13}C NMR spectroscopy technique allows obtaining information on the carbon components of the sample without any chemical or physical fractionation, and is well suited by characterization of natural organic matter (Preston, 1996).

Spain is second largest pig producer in the Europe, therefore managements of the waste is very important in terms of environment pollution and safety. This waste can be utilized as farmyard manure in agricultural production. For this reason application and management of animal wastes are becoming very important research topic.

The nature and properties of animal manure change with many factors such as the source, age, weight, feeding diet, e.g. Physical-chemical processes in these manures could change many properties. The objectives of this study are to clarify the effect of different kind of PS application on the structural characteristics using ^{13}C-NMR, to obtain new information for improving the soil fertility by PS application.

The ^{13}C-NMR spectra were divided into 7 chemical shift regions: alkyl-C (0–50 ppm); Methoxyl (50–60 ppm); O-alkyl C (60–98 ppm); di-O-alkyl (98–112 ppm); Aromatic C (112–145ppm); Phenolic C (145–163 ppm) and Carbonyl C (163–190 ppm). Areas of the chemical shift regions were measured by integration and were expressed as percentage of the total area (relative intensity). Sidebands contribution generated by the carbonyl region was corrected by multiplying the area under the peak to the low field sideband (215–230 ppm) by a factor of 2 and adding it to the carboxyl area (163–190 ppm). An area equivalent to the low field sideband was deducted from the 112–145 ppm area to correct the high field sideband contribution in this spectra range (Faz Cano et al., 2002).

Materials and methods

The study area, is located in the province of Albacete, in the town of Hellin, particularly in the hamlet of Cancarix (38°.45'13'', N latitude and 1°54'19'' W longitude), shown in Fig. 1. Climate is semiarid, with mean annual average rainfall of 376.7 mm and mean annual temperature of 20.6^{0}C. **.** The major soil type is Typical Entisol-Orthent-Xerorthent (Soil Survey Staff, 1999). The description of the study area and in-depth knowledge of it has identified the unit of production depending on the feeding diet. It contributes to the characterization of soil resources and the determination of the recommended doses of manure to apply.

Fig. 1. Location map of the study area.

The investigated organic matter complexes are obtained from the A horizon of thirteen soil samples, taken before and after application in non-irrigated field, planted with a continuous barley system. Fertilizer treatments are established in the field with three replicates. These are: application of raw and treated pig manure from conventional and liquid feeding and an unfertilized control.
The C fractions of bulk soils are examined by a solid-state CP/MAS ^{13}C NMR. These soil samples are air-dried and crushed to pass through the 2 mm sieve. Soil samples are sift with water to pass through the 0.05 mm sieve and dried in the oven at 40 °C and homogenized to remove sand from the soil. The soil samples are analysed using a spectrometer Varian Unity 300, located in the Support to Experimental Science (in SACE Lab.-Murcia) with silicon nitride rotors of 7 mm outer diameter and 5.6 mm internal diameter, sealed with Kel-F caps. 13C CPMAS measurements were carried out at 4 kHz rotational speed, with a frequency of 13 C sfrq = 75 420 MHz contact time p2 = 1.5 ms. The spectrum is acquired with a recycle time of 4s and pulse of 90 º pw = 6.7 ms. Spectral width sw = 50 000 Hz; scans nt = 60 000 and line broadening of 100 Hz.

Result and Discussions

Solid state CP/MAS 13C NMR spectra acquired for the loamy soil in sixteen application plot are given in Fig. 2. The results showed that mean average of alkyl groups have changed between 39.4% and 47.4%, methoxyl groups 5.7% and 7.8%, O-alkyl groups 20.5% and 15.9%, di-O-Alkyl groups 1.6% and 1.7%, carbonhydrate groups 24.1% and 27.3%, aromatic groups 15.3 %and 19.4%, phenolic groups 1.4% and 5.0%, aromatics 17.3 %and 24.8%, carboxyl groups 8.1% and 12.3% and A/OA have changed between 2.2 and 3.0.

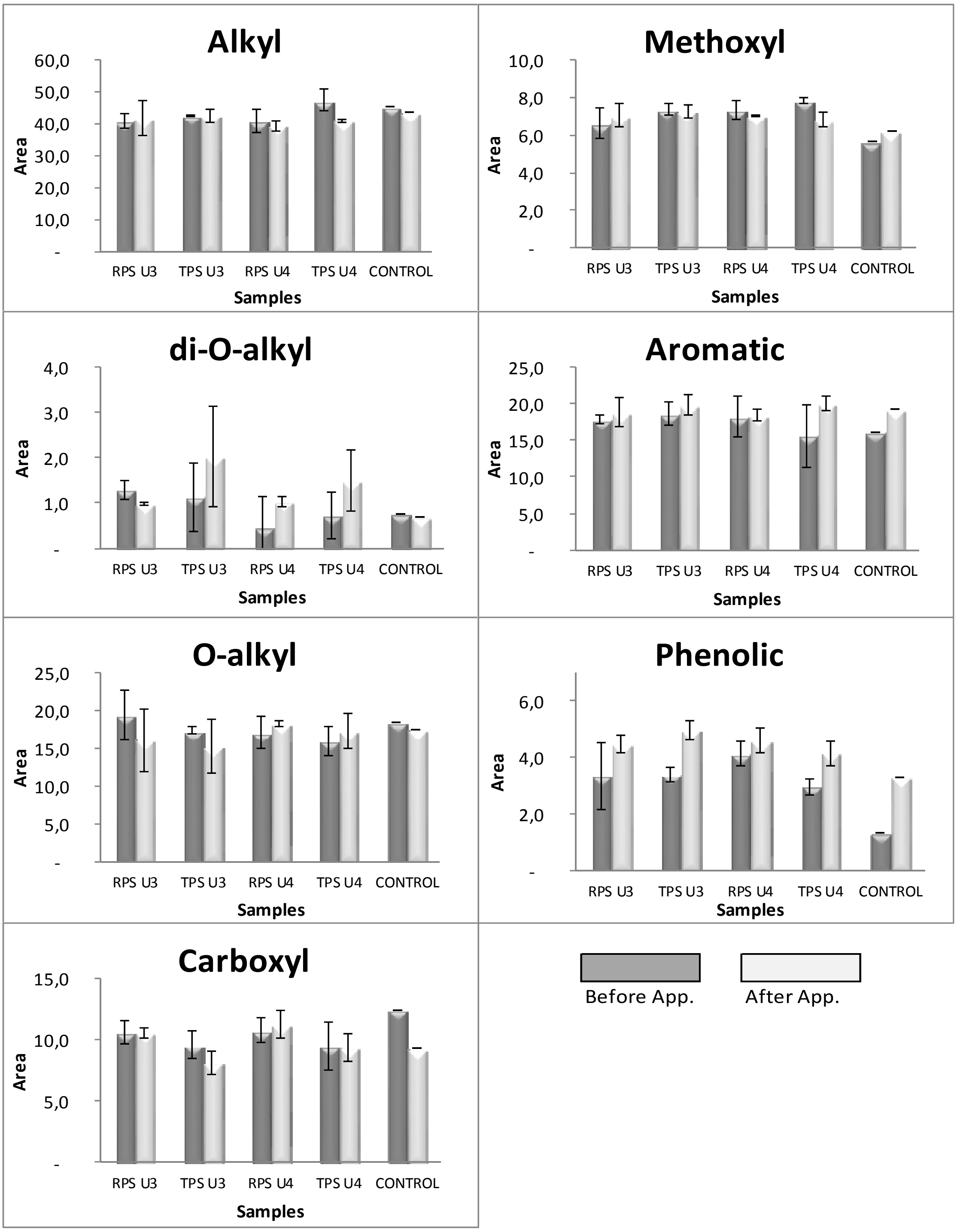

Fig. 2. Band assignments and relative distribution of C as percent reduction or increase of the main resonances ranges from initial (black) to final samples (gray) RPS (Raw Pig Slurry), TPS (Treated Pig Slurry), U3 (Unit 3) and U4 (Unit 4) the CPMAS 13C NMR spectra.

Amounts of O-alkyl groups were not different from the initial and final swine manure application in the soil on relative intensity of C region of O-alkyl group, according to paired t test. The percentage of O-alkyl group of the raw and treated pig slurry applications in unit 4 has caused an increase of this functional group in the soil. However application of PS to unit 3 decreased the percentage of O-alkyl group in the soil, (Fig. 2).

The O-alkyl-C content is lower, whereas the alkyl-C content is higher in the present study (Chen and Chiu, 2003). The alkyl carbon is a major contributor to the soil that does not contain high amounts of organic matter under the arid and semi-arid climate condition. These soils are rather dominated by alkyl carbon and accumulation of alkyl C may originate from a combination of microbial and plant sources (Shepherd et al., 2001; Reiderer et al., 1993). High amount of alkyl-C is likely associated with high bioavailability of the plant residues, which undergo fast decomposition of the O-alkyl-C, already at early stages of humification (Lehmann et al., 1997).

Research based on 13C NMR is shown that the importance of alkyl-C as a source of stable structures contributing to its formation rather than aromatic-C in the decomposition process of SOM and this aromatic-C contributing to the formation of recalcitrant humic substances (Almendros et al., 2000; Preston, 1996; Mahieu et al.,1999).

The feeding diet types are differently affected on properties of animal manure therefore, intensity of OM functional groups are changed with different treatments. The results confirmed that; alkyl and methoxyl-C groups were decreased with treated PS application. The O-alkyl-C groups were decreased with liquid feeding diets and increased with solid feeding diets. The di-O-alkyl-C groups were increased with all diet types except of RPS from liquid feeding diet. The aromatic-C gorups intensity were increased with TPS from solid feeding diets. The phenolic groups were increased with all PS applications. The carboxyl groups were increased RPS from solid feeding and decreased with TPS from liquid feeding diets (Fig. 2.).

Conclusions

The high amount of alkyl group confirms rapid mineralization of organic compound. Decrease of O-alkyl and di-O-alkyl groups' confirms the stage of decomposition of organic matter in the soil. Due to insufficient precipitation in Mediterranean climate conditions decomposition of organic matter is generally low. While the solid state ^{13}C-NMR spectra of organic matter of the 13 surface soils, representing four different pig slurry applications and under similar vegetation displayed similar sequence of functional groups: alkyl>O-alkyl>aromatic>carboxyl. The source of pig slurry applications did not affect the alkyl and O-alkyl functional groups statistically significantly. However applications significantly affected the aromatic and phenolic group of organic matter.

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Effects of Pig Slurry Application on Heavy Metal Concentration in Undisturbed Soil Column

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Abstract: The soils of SE Spain are often relatively infertile, low in organic matter, and highly susceptible to erosion. The application of livestock waste is therefore useful. As the second largest pig producer in the European Union, Spain generates $2x10^{10}$ kg yr^{-1} of pig slurry, over half of which is used directly as fertilizer. When pig slurry from intensive livestock operations are applied to agricultural soils at a high rate, large amounts of salts and heavy metals (HMs) are introduced into soils.
Using a column experiment, this study assessed the leaching potential of the downward movement of pig slurry, after an intensive application to the top 10 cm of silty loam textured soils, in 3 different doses: D_1 (170 kg N/ha), D_3 (540 kg N/ha) and unfertilized plots (C) served as controls. The possibility of HMs (Cu, Zn , Cr, Mn, Co, Ni, Mo, Pb and Cd) transfer from soil to ground-water was investigated through the analyses of solutions each week.

The results showed that concentrations of HMs in leachates were in the following order: Mn>Mo>Fe>Cu>Ni> Zn>Pb>Cr>Co>Cd. All the concentrations of HMs in the leachate samples are in permissible limits of Europe standard. High concentration of Cu and Zn in pig slurry causes higher leaching and uptake risk by plants in a silty loam soil.

Keywords: HMs, Leaching System, Undisturbed Soil Column.

Introduction

The recycling of pig slurry in agricultural soils is an alternative and valuable practice in countries such as Spain with significant European pig production (>2.5 million in 1998). Spain is the second pig producer in the Europe with 19% production power, with economic value of 3.7 billion € (Hansson, 2003). This is particularly the case since many regions are arid and are comprised of poor soils (‹1% organic matter). Recycling of organic matter applied to agricultural soils adds . When pig slurry from intensive livestock operations is applied to agricultural at a high rate, large amounts of heavy metals (HMs) are introduced into soils.
Heavy metals (HMs) are one of the most dangerous contaminants for environment and humans (Bradl, 2005). Once excessive HMs enter in the soil, water or air, it may become hazardous to human health through consumption of food crops cultivated in these contaminated environments (Zhao et al., 2002 and Wang et al., 2003). Food and Agriculture Organization (FAO) and Word Health Organization (WHO), United States Environment Protection Agency (US EPA) and other regulatory bodies of other countries strictly regulate the allowable concentrations or maximum permitted concentrations of toxic HMs in foodstuffs (FAO, 1984 and US EPA (United States Environmental Protection Agency, 2000).
The objective of this study were to: 1) determine HMs concentrations of the undisturbed soil columns, 2) compare with field condition, and 3) determine HMs transfer from soil to underground water of silty loam soils in Southeast of Spain.

Materials and Methods

Charactestics of the Study Area, Design and Sampling of Undisturbed Soil Columns

Study area is located in La Hoya near the Lorca in Southeast of Spain (Fig. 1). Climate is semiarid, with mean annual average rainfall of 300 mm and mean annual temperature of 18^0C and potential evapotranspiration is >900 mm yr^{-1} (López-Bermúdez et al., 2002).

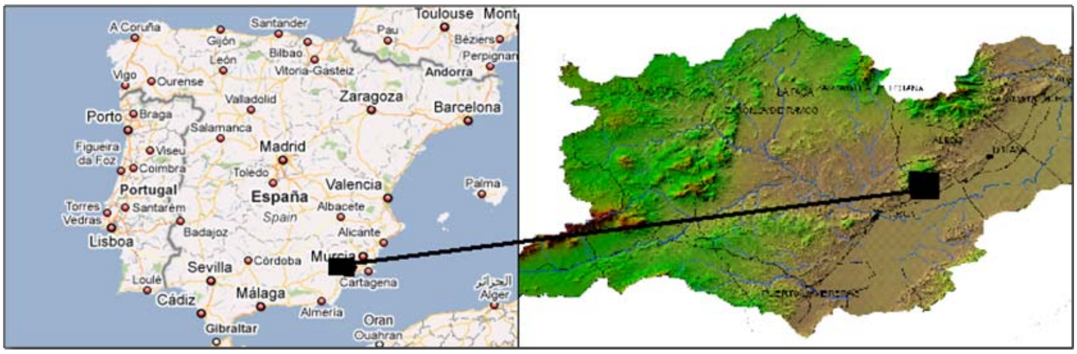

Fig. 1: Location of the area studied.

Nine undisturbed soil columns were taken from silty loam soils in Southeast of Spain. Soils were extracted using transparent metacrylate cylinder 60 cm long, 25 cm inside diameter, and 45 cm thick. Soil columns were taken from surface to a depth of app. 35-45 cm of three representative soils. Experimental design of column is given in Fig. 2. Sea sand free of salts was used to support material in columns. Simulated rainwater was given to columns by peristaltic pomp. Soil columns were amended with single and triple doses of pig manure applied in the surface 0 to 10 cm depth. The applied doses were calculated from the agronomic rate of N-requirement (170 kg N/ha/yr-Single Dose-D_1) (European Directive 91/676/CEE). Undisturbed soil columns were extracted using equipment designed by Mihaljevic et al., (2004), Ashworth and Alloway (2004), Doye & Duchesne (2003), Camobreco et al., (1996), and recommended by ISO/DIS 18772, (2006) and modified by Carmona and Faz (2004).

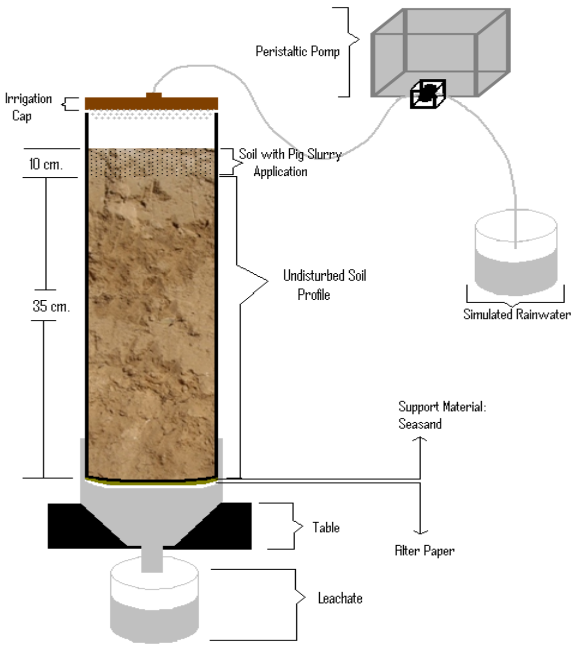

Fig. 2: Experimental design of columns

Leachings were carried out weekly using distilled water at a rate of 80 mL h^{-1} to simulate the monthly rainfall events in the area. Experiment was carried out for 12 weeks rainfall (W); three columns of them for single doses (D_1-Low Dose), three columns of them for triple doses (D_3- High Dose) and three columns of them for control (C). Leachates were collected each week, filtered with a Whatman nº 42 paper, and filtered liquid was refrigerated at 5 ºC while awaiting chemical analysis.

Some conditions of the laboratory are given in Table 1. First rainwater was applied in September, which is determined according to farmers, who apply pig slurry to field as the August. Temperature is changed between 23 and 27 ^{o}C, pressure is changed between 747 and 756.5 mmHg in the laboratory. There are no leachates in Week 11 which coincides with July.

Table 1: Quantity of leachates in each week and simulated months

Week	Temperature (ºC)			Pressure (mm Hg)			Simulated Months	Simulated Rainwater (ml.)
W0	23,6	±	0,5	748,5	±	0,4	Before application	-
W1	24,9	±	0,3	746,7	±	1,2	September	**1010,02**
W2	24,4	±	1,5	756,5	±	0,9	October	383,09
W3	24,2	±	0,6	749,1	±	3,5	November	462,95
W4	26,1	±	0,0	750,0	±	0,0	December	351,28
W5	25,6	±	1,3	748,0	±	1,8	January	530,81
W6	23,3	±	1,1	749,6	±	0,5	February	312,41
W7	26,4	±	2,1	749,8	±	1,8	March	541,41
W8	26,6	±	0,5	751,7	±	1,2	April	515,26
W9	25,7	±	1,5	750,3	±	0,5	May	650,96
W10	25,0	±	0,0	750,0	±	0,0	June	185,18
W11	26,1	±	2,5	748,8	±	1,3	July	**31,1**
W12	24,9	±	2,4	749,7	±	0,6	August	301,1

Chemical analysis

Selected chemical properties of soils and pig manure were determined following the routine methods for soil analyses. Chemical composition of leachates was determined following analytical techniques suggested in APHA (1998). pH and electrical conductivity (EC) were analysed in every day. Heavy metals were analysed each week in leachate at initial stage and final in the soil by an Agilent 7500a model of ICP-MS (Inductively Coupled Plasma Mass Spectrometer) and also atomic absorption spectrophotometer (AAnalyst 800, Perkin Elmer).

Results and Discussion

Soil Properties and Some Initial-Final Results of Soil Column

The soil results of studied area showed that; the study area had basic reaction, non-salty, very low contents of total nitrogen; high contents of phosphorus, low cation exchangeable capacity, and silty loam texture. Calcium carbonate contents were changed between 55 and 60 % and soil organic carbon were changed 13.08 and 14.31 g kg^{-1}. SOC was mineralized soils in the study area, quantities of carbon was similar to Mediterranean soils under arid-semiarid climate, which were less than 20 g kg^{-1}.The results of applied pig slurry showed that cations and phosphorus contents were high in applied pig slurry, the other parameters were normal. The dry matter content of the applied pig slurry was higher. Although Na and K concentration were very high, bioavalaible metal concentrations were not high.

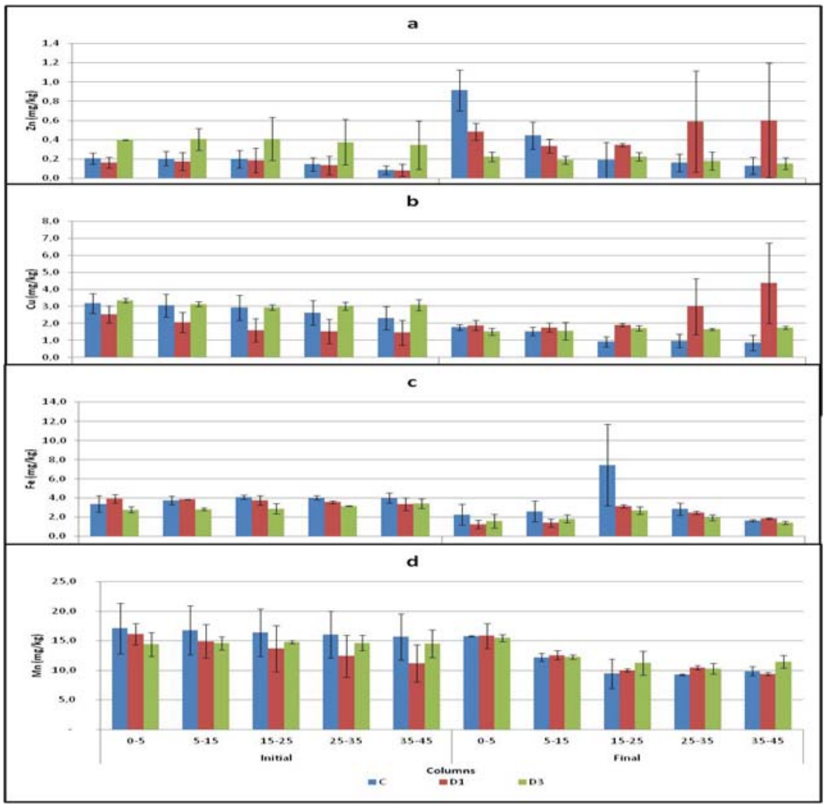

Fig. 3: Some heavy metals contents of initial and final stage of the soil columns

The results of initial and initial soil of columns showed that CEC in soil was decreased in control and D_1 application with depth, but D_3 application was stable with depth. Cu, Fe and Mn concentrations were decreased with all doses applications, because they were leached out. Zn concentrations were increased with D_1 application, but it was decreased with D_3 application (Fig. 3.).

The normal Cu content of agricultural soils is 5 to 50 mg kg^{-1}. Concentrations below 8 mg kg $^{-1}$ could indicate a deficiency for some crops, as Cu is an essential micronutrient (McBride, 1994; Kabata-Pendias and Pendias, 2001).In this study, soil samples were in the normal range of agricultural soils. Mean average of Zn for world-wide soils may be considered as 64 ppm. The Zn concentration in agricultural soils varies between 10 and 300 mg kg $^{-1}$. It is abundant in sedimentary materials and clayey soils (Kabata-Pendias and Pendias, 2001). In our study, soils contained less Zn than the maximum permissible concentration. Manganese mean calculated for world soils is 437 ppm, while for the U.S. soils the calculation is 495 ppm. In our study, Mn content of the soil was higher than permissible limits (Kabata-Pendias and Pendias, 1992).

Heavy Metals Contents of Leachate in Undisturbed Soil Columns

Copper (Cu): Cu contents were increased with application of pig slurry. According to limits prescribed by WHO (2008), it was found that the average value of copper in all leachates are much below the permissible limits.

Zinc (Zn): Zn contents were increased with application, of D_3. Zinc is an essential element for plant and human. The maximum allowable concentration of zinc in water is 15 ppm and 5 ppm respectively. According to WHO (2008), the average value of zinc in all leachate is below 100 ppb (0.1 ppm).

Manganese (Mn): Mn contents were decreased with application. High concentration was with D_3 application. Mn in leachate in the first four weeks was higher than other weeks. The maximum allowable concentration and permissible concentration of Mn in water 0.5 ppm and 0.05 ppm respectively according to WHO (2008). The most of the leachate analyses had less than 200 ppb (0.2 ppm), except the second week of study.

Iron (Fe): Fe contents were decreased with application. D_1 application had more Fe contents than the others. According to WHO (2008) the maximum allowable concentration in water in 1.0 ppm and 0.3 ppm respectively. Maximum concentration was 0.02 ppm in leachate with D_3 application. that the average value of iron in all leachates samples are much below the permissible limits.

Nickel (Ni): Ni contents were increased with slurry application. Especially with D1 application. Nickel concentration in Week 11 was higher than the others. The permissible concentration of nickel in groundwater is 0.02 ppm (WHO, 2008). D_1 and D_3 application were out of limit.

Molybdenum (Mo): Mo contents were increased at the beginning of the application. According to WHO (2008) the maximum allowable concentration in water in 70 ppb (0.07 ppm). Maximum concentration of Mo was 20 ppb with D_3 application in W2. All application doses were within the limit.

Lead (Pb): Pb contents were increased with especially D_3 application. This is a very toxic element, which accumulates in the skeletal structure of man and animal. The maximum permissible concentration of lead in water is 0.05 ppm (50ppb). According to WHO (2008) almost all the leachate samples had less than 50 ppb of lead.

Chromium (Cr): According to Cr results, Cr contents were similar control and D_1 application. Results of first six week were similar, but Cr contents were increased in the last six week with application especially with D_3 application. The maximum permissible limit of Cr in water according to WHO (2008) is 0.05 ppm. According to WHO almost all the leachate samples were below the limit.

Cobalt (Co): Co contents were higher at the beginning of the study especially in D_3 application. It was balanced with all application at the end of the study. Generally the natural content of Co in drinking water is very low ranging from 10 to 50 ppb except for the regions with substantial chromium deposits (Jayana et al., 2009). The average value of Co in all leachates samples are much below the permissible limits.

Cadmium (Cd): Cd contents in leachate were below the permissible limits, as they were lower than the permissible limit except of W1. Cd value is above the WHO (2008) recommended value (3µg/L).

Study has shown that heavy metals such as lead, cadmium, copper etc… were not excessive levels in groundwater due to D_1 pig slurry application.

Conclusions

The need for food production for growing human population increases the research interest in the area of soil and water quality. Specifically, the use of commercial fertilizers with high contents of metals created a very serious problem for agricultural soils. Toxic heavy metal enters the food chain from the soil through plant uptake. To avoid this, WHO has developed maximum tolerable limits of the HMs for agricultural soil and water. The heavy metals contents of the soil and leachate were below the allowable levels. We, therefore conclude that the quality of underground water is still high with pig slurry application. PS has high content of micronutrient and application of high doses can pollute the agricultural soils decrease the quality of crops. We concluded that the D_3 doses are not suitable especially when used for unfertilized soils that can cause reduction in plant. The benefits from the use of slurry depend on the proper use and management. Our results suggest that D_1 application to silty loam soils has positive effect on the soil and ground water.

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Potentially Toxic Elements of Volcanic Ash Soils in Middle Anatolia Region of Turkey

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Abstract
The objective of this study was to determine the variability and concentrations of the potentially toxic elements (PTE) such as Cd, Co, Cu, Cr, Mn, Zn, Pb and Ni in soils formed on volcanic parent materials (andesite, dasite, ignimbrite, volcanic ash and basalt) erupted from Erciyes strato volcano in middle Anatolia region of Turkey. This study was done based on 576 soil samples collected randomly from three different soil depths (0-30, 30-60 and 60-90 cm). The PTE concentrations of soils formed on ignimbrite and volcanic ash were higher than the soils formed on basalt, andesite and dasite. All the PTE concentrations decreased with increasing soil depth except for Cd. The Cd, Cr and Ni concentrations which increased with elevation while the Co, Cu, Mn, Pb and Zn concentrations decreased.
The high PTE concentrations in soils could be attributed to the volcanic parent materials. The greatest PTE variations in soils were determined by lithology structure related to the parent material and its composition. Agricultural practices did not affected to the PTE contents in soils. The PTE concentrations were greater at the uncultivated soils compared to the cultivated soils due to the high feldspars of uncultivated soils. Site specific soil management practices must be applied to soils in the study area because of the high PTE concentrations.
Key words: Potentially toxic elements, volcanic ash soils, soil parent materials, soil pollution, and spatial variability

Introduction

The soils formed on volcanic parent materials including high amounts of PTE are found in many regions of the world (Amaral *et al.,* 2006). The distribution and amount of PTE in soils depend on the nature of parent material, weathering processes, bio-cycling and addition from atmosphere and deposition from natural resources (Cortizas *et al.,* 2003).
These factors influence soil development and the mobility of specific elements, including PTE, in the soil system. Rock mineral weathering is the one of main natural sources of PTE to the soil system and metal concentrations in soil can generally be predicted from the element concentrations in the parent material (Palumbo *et al.,* 2000).
Volcanic rocks cover a significant part of Turkey. The majority of these rocks are located in the Volcanic Province of Cappadocia (VPC) (300x60 about 18 000 km^2). The soils located in this province were formed on volcanic parent materials of Neogene-Quaternary ages. Volcanic activity causes the release of PTE such as As, Hg, AI, Rb, Pb, Ni, Co, Cr, Mg, Cu, and Zn which in turn cause water and soil pollution. The majority of the volcanic ash soils have excellent properties for plant production (high water-retention capacity, high cation-exchange capacity and high organic matter content etc.). The aim of the present study was to determine the distribution and differences of PTE contents of Erciyes mount volcanic ash soils using GIS and remote sensing techniques.

Materials and Methods

This research was carried out on the soils formed on five different volcanic parent materials in VPC of Turkey. VPC covers about 18 000 km^2 and a part of this area (2400 km^2) was studied. The study area is generally located at the eastern of Erciyes strato volcano. There are five main volcanic parent materials in the study area. These parent materials are andesite, dasite, volcanic ash, basalt and ignimbrite.
In this research, stratified random sampling design was used. The study area was stratified by using the results of initial (field) surveys, archive LANDSAT-ETM+ images, a digital elevation model with 1/25000 scale, and digital soil maps. Sampling areas were randomly distributed within the established strata. Land use, land cover class and soil sampling sites were linked to their geographic references by using GPS. The all geo-referenced observations were compiled in a GIS

database. Soil samples were taken from 192 sampling points and three different depths (0-30 cm, 30-60 cm and 60-90 cm soil depths).

GPS and soil analysis data were compiled into the GIS database. Database was transferred to a GIS environment to produce surface layers of each variable by using Kriging (spherical variogram method). Next, correlations between these surface layers and SPOT-5 images were investigated.

In this study, one SPOT-5 image having 2.5 m resolution was utilized. Both supervised and unsupervised classifications were applied using ARC/GIS and ERDAS 8.7 software for GIS and remote sensing applications, respectively. Residual layers were created by using the maps produced by the model and the on-field observations. In this way, the differences between the predicted and observed values were mapped. Consequently, the determined models were tested by using these residual layers. The models were run in a GIS environment.

For soil samples, 1 g of dried samples was digested with 15 mL of HNO_3, H_2SO_4, and $HClO_4$ in 5:1:1 ratio at 80 ^oC until a transparent solution was obtained (Allen et al., 1986). The solution was filtered through Whatman No. 42 filter paper and the solution was diluted to 50 ml with distilled water. The concentrations of heavy metal in the filtrate were determined by using ICP emission spectrometry (Perkin-Elmer model 2100, USA).

Results and Discussions

PTE concentrations in soils developed on parent materials erupted from Erciyes Volcano were higher than standard limit values. For Cd, the minimum value was 0 ppm, the maximum value was 9358 ppm and the highest mean concentration was in the volcanic ash with 1778.7 ppm (Table 1). For Co, minimum value was 2.23, maximum value was 105.3 ppm and the highest mean concentration was with 43.43 ppm in the volcanic ash. For Cr, the minimum value was 1.86 ppm, the maximum value was 1853 ppm and the highest mean concentration was 95.08 ppm in the ignimbrite. For Cu, the minimum value was 11.09 ppm, the maximum value was 109.4 ppm and the highest mean concentration was in the volcanic ash with 52.82 ppm. For Mn, the minimum value was 23.71 ppm, the maximum value was 3988 ppm and the highest mean concentration was 1289.23 ppm in the volcanic ash. For Ni, the minimum value was 9.07 ppm, the maximum value was 8239 ppm and the highest mean concentration was 846.83 ppm in the ignimbrite (Table 1).

For Pb, the minimum value was 4.21 ppm, the maximum value was 242 ppm and the highest mean concentration was 39.90 ppm in the andesite. For Zn, the minimum value was 0 ppm; the maximum value was 279.08 ppm in the andesite.

In the andesite, Cd had the highest mean concentration, Co had lowest mean concentration. In the basalt, Mn had the highest mean concentration, Co lowest mean concentration (Table 1). In the dacite, Cd had highest mean concentration, Co lowest mean concentration (Table 1). In the ignimbrite, Mn had the highest mean concentration, Co lowest mean concentration. In the volcanic ash, Cd had the highest mean concentration, Pb lowest mean concentration. PTE distributions for the all parent materials were generally as follows;

Volcanic ash > ignimbrite > andesite > basalt > dasite, with the greatest shifts observed in relative amounts of Cd, Mn, Ni, and Zn among PTE.

The variations in PTE concentrations in the different soil depths were compared to the limit concentrations. The all PTE concentrations were higher than limit concentrations. Cd concentrations increased from the surface layer (0-30 cm soil depth) to the intermediate layer (30-60 cm soil depth) and to the deep layer (60-90 cm soil depth) (Table 2). The mean concentrations of Cd according to the different soil depths were 14.63 ppm, 1655.6 ppm and 2013.9 ppm, respectively (Table 2). The map of spatial distribution and variability of Cd were presented in Figure 3. Cd concentrations were generally higher with a maximum concentration ranges from 4700 ppm to 9400 ppm at the 30-60 cm soil depth in the parts of north and middle of the study area than the other parts.

Co concentrations did not show an important change from surface layer to the deeper layers. The high Co concentrations were found in the parts of northeastern and middle of the study area (Figure 3). Co concentration increased with the soil depth in the volcanic ash parent materials although Co concentration decreased with the soil depth in andesite, dasite and ignimbrite parent materials.

Cr concentrations generally showed a decrease from the surface to the deeper layers (74.18 ppm at the surface layer, 82.79 ppm for the intermediate layer and 64.57 ppm for the deep layer). The

highest Cr concentrations were found in the parts of north-west of the study area, with a maximum value 1900 ppm at the surface layer (Figure 3).
The amounts of Cu, Mn and Ni were found higher in the parts of North and middle of the study area than other parts except for Pb and Zn (Figure 3). The amounts of Pb and Zn were generally high in the the study area. The concentrations of PTE were appeared to be above the limit concentrations at the soil depths.
The data were analyzed using the Duncan test. The values of Zn, Ni, Mn and Cd were statistically significant at the different soil depths ($p<0.05$), except for Co, Pb, Cu and Cr (Table 2). Amaral et al. (2006) found that Cd and Co concentrations were increased with depth in volcanic soils of Portugal. The similar results for Cd and Co were reported by Palumbo et al. (2000) in various soil types developed from volcanic parent materials. They showed that Cd and Co concentrations increased with depth in andisols. Doelsch et al. (2006) reported that Cr, Cu, Ni and Zn concentrations were constant with soil depth, but Cd concentrations decreased from the surface to the intermediate layer and stayed constant in the deep layer.
The sampling locations in the study area were divided to the two different elevations from 1000 to 1500 m and from 1500 to 3000 m. The mean concentrations of Cd, Cr, Cu, Ni and Pb were statistically different between 1000-1500m and 1500-3000m elevations ($p<0.05$), although the concentrations of Co, Mn and Zn were not significant between elevations (Table 3). The amounts of Cd and Ni increased with the elevation at the parent materials while the amount of Pb decreased with elevation in the all parent materials. The amounts of Co and Mn decreased with elevation in andesite and volcanic ash while they increased in ignimbrite and dasite (Table 3). PTE concentrations in Andisols (Cd, Cu, Ni, Cr and Zn) were found higher at low altitude than that of high altitude (Doelsch et al., 2006). The soils at 1000-1500 m elevations are generally cultivated, but at 1500-3000 m elevations are not. The high concentrations of Cu, Mn, Pb and Zn in cultivated soils can be related to the agricultural practices such as fertilization.
Thus, high Cu and Zn concentrations in cultivated soils could be explained by the geologic origin rather than agricultural practices (Doelsch, 2006). Cr and Ni appeared to be high concentration particularly in the uncultivated soils. This suggested that geologic origin was prevalent, agricultural practices cannot be ruled out.
The high concentrations of Cr and Ni in the high elevations and uncultivated soils could be attributed to Feldspars (figure 4 and 5). Heavy metals such as Cr, Ni, Co, Cu and Pb are nearly always present in Plagioclase Feldspars (Huang, 1989).
This study presented the variability of potentially toxic elements in soils, formed on volcanic parent materials erupted by Erciyes strato volcano, according to the parent material, soil depth and elevation in the middle Anatolia region of Turkey. The PTE concentrations in soils were higher than standard values. This study demonstrated that PTE concentrations in soils were originated from volcanic parent materials and typically increased with increasing elevation and soil depth. The PTE contents of soils formed on ignimbrite and volcanic ash were higher than the soils formed on basalt, andesite and dasite. In addition, the PTE contents of soils in the north and middle regions of the study area found higher than the other areas because of the high elevations. These results indicated that the agricultural practices did not affect to the PTE concentrations. The high PTE concentrations in the uncultivated soils could be attributed to the mineralogical composition. Feldspars could account for the high PTE concentrations in high elevations. PTE in soils could affect the ecosystem and human health. The high PTE could be taken up by plants and interfere with the groundwater. Different soil management systems should be applied to the cultivated areas such as the site specific management practices and appropriate plants species with minimum PTE uptake.

Table 1 The results of Duncan analysis of PTE according to the parent materials

		Cd	Co	Cr	Cu	Mn	Ni	Pb	Zn
Parent material	Andesite	976,2^{b}	30,87^{c}	84,03^{a}	40,08^{b}	908,57^{c}	485,07^{a}	39,90^{a}	279,08^{a}
	Basalt	651,0^{c}	31,12^{c}	39,43^{a}	40,33^{b}	997,97^{c}	93,42^{b}	37,23^{a}	218,66^{a}
	Dasite	694,5^{c}	16,27^{d}	49,86^{a}	30,37^{b}	332,69^{b}	405,13^{a}	31,63^{a}	193,99^{b}
	İgnimbrite	1750,8^{a}	38,22^{b}	95,08^{a}	43,36^{b}	1247,36^{a}	846,83^{c}	36,57^{a}	255,55^{a}
	Volcanic ash	1778,7^{a}	43,43^{a}	83,63^{a}	52,82^{a}	1289,23^{a}	611,84^{a}	37,18^{a}	276,78^{c}

Table 2. The results of Duncan analysis of PTE according to the soil depths

	Potentionally toxic elements							
Soil depth (cm)	Cd	Co	Cr	Cu	Mn	Ni	Pb	Zn
0-30	14.63^{a}	34.11^{a}	74.18^{a}	46.97^{a}	1070.67^{a}	538.60^{a}	42.85^{a}	512.46^{a}
30-60	1655.60^{b}	31.09^{a}	82.79^{a}	39.19^{a}	862.27^{b}	447.00^{b}	35.83^{a}	126.44^{b}
60-90	2013.9^{b}	31.02^{a}	64.57^{a}	36.42^{a}	817.08^{b}	477.52^{a}	34.06^{a}	119.57^{b}

Table 3. The results of Duncan analysis of PTE according to the elevation

	Potentionally toxic elements							
Elevation (m)	Cd	Co	Cr	Cu	Mn	Ni	Pb	Zn
1000-1500	14.60^{a}	35.86^{a}	74.43^{b}	54.47^{a}	1132.18^{a}	454.49^{b}	49.78^{a}	585.21^{a}
1500-3000	18.98^{b}	31.15^{a}	131.81^{a}	36.82^{b}	987.22^{a}	652.78^{a}	33.46^{b}	413.74^{a}

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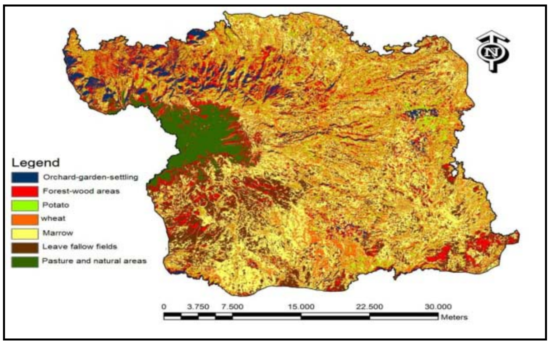

Fig. 1. The land use map of the study area.

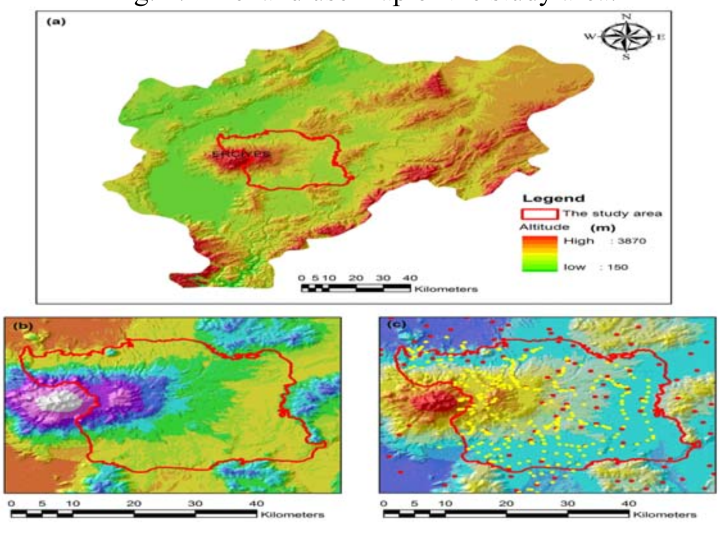

Fig. 2 .The location of the study area (a), the map of DEM (b), the points of soil sampling (c).

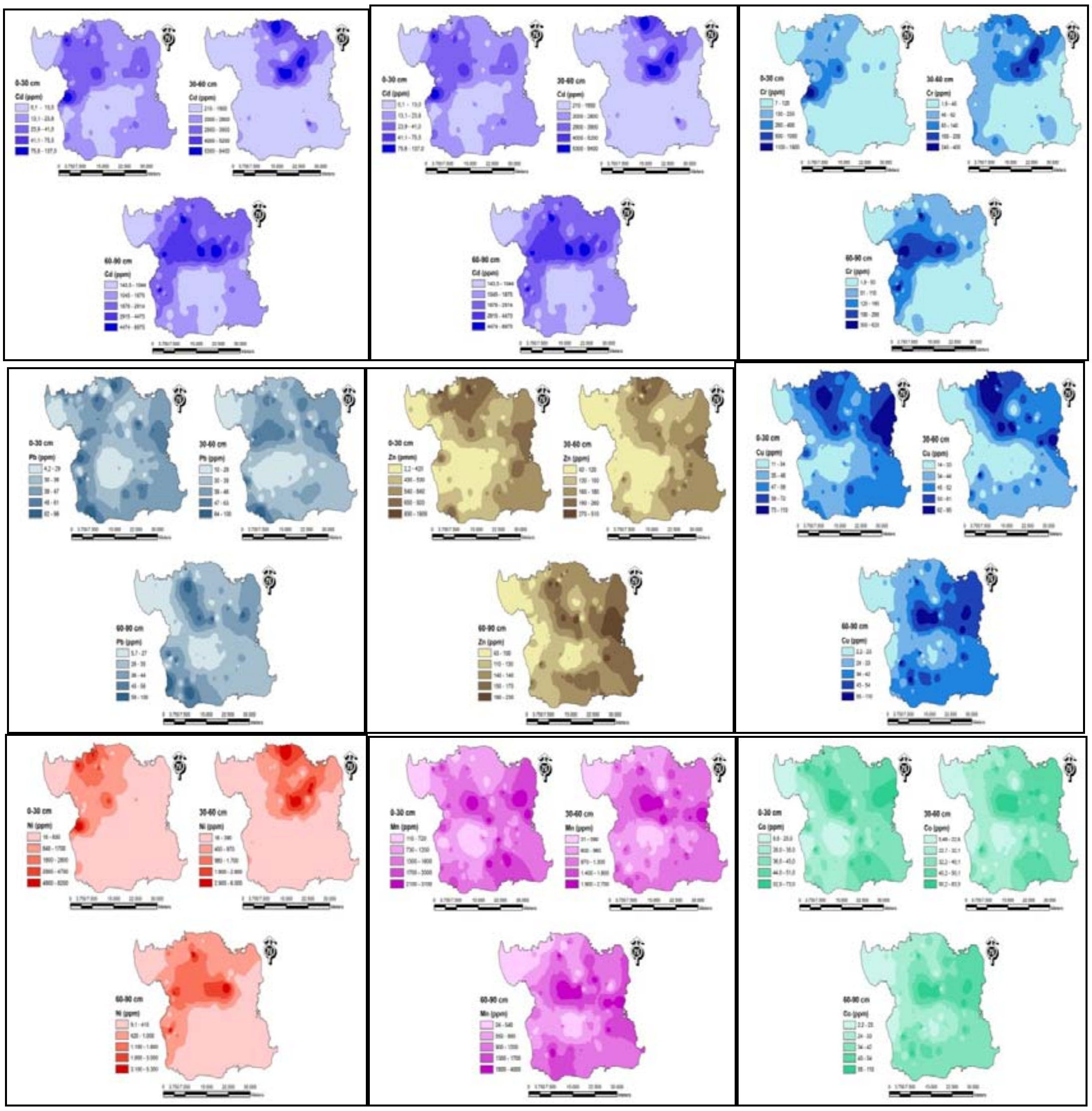

Fig 3. Spatial variability of potantially toxic elements.

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Effects of inoculation of *fluorescent pseudomonads* with ACCD activity on plant growth and Zn uptake by barley and canola

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Abstract

Metal-resistant PGPR play an important role in the successful survival and growth of plants in contaminated soils by alleviating the metal toxicity and supplying the plant with nutrients. Soil used in this study was artificially polluted with Zn (250 and 500 mg L^{-1}). Three isolated strains included *Pseudomonas potida* strain 11 (*P.p.*11*)*, *Pseudomonas potida* strain 4 (*P.p.*4*)*, *Pseudomonas fluorescens* strain 169 (*P.f.*169*)* showing Zn-resistance were investigated for their effects on the soil Zn solubilization and promotion of plant growth and Zn uptake by canola and barley in a pot experiment. The bacteria produced IAA (indole acetic acid), siderophore and 1-aminocyclopropane-1-carboxylate deaminase (ACCD). When strains added to the Zn-amended soils, bacteria increased Zn extraction from the soils. Inoculation with the isolates was found to increase shoot dry matter of canola and barley in the Zn-contaminated pots. In Zn treatment, *P.p.*11 inhanced TF (translocation factor) values in canola, while all of the three strains decreased it in barley. Value of TF indicated that inoculated canola with *P.p.*11 had abilities of phytoextraction and translocation of Zn in the contaminated soil compared to the non-inoculated control respectively. The present observation showed that bacterial strains used in this study protect the plants against the inhibitory effects of Zn probably due to production of IAA, siderophore and ACCD activity.

Keywords: Metal-resistant, *Pseudomonas*, ACCD activity, translocation factor, contamination.

Introduction

The threat of heavy metal pollution to public health and wildlife has led to an increased interest in developing systems that can remove or neutralize heavy metal toxic effects in soil, sediments and wastewater (Valls and Lorenzo, 2002). In order to eliminate or control the pollutants in soils, physical, chemical, and biological methods have been employed. Bioremediation is the application of biological processes for the clean up of hazardous chemicals present in the environment (Gianfreda and Rao, 2004).

A number of plants which can tolerate and accumulate high concentration of metals were defined as hyperaccumulators. Ideal hyperaccumulators for bioremediation require the characteristics of rapid growth and a high amount of biomass (Nie et al., 2002). But in fact, many hyperaccumulators are slow in growth and inhibited in the presence of high concentration of heavy metals.

Plant growth-promoting rhizobacteria (PGPR) are bacteria capable of promoting plant growth by colonizing the plant root (Kloepper and Schroth, 1978). PGPR have a positive effect on plant growth by producing indole aceti cacid and siderophores and by consuming amino-cyclopropane carboxylic acid (ACC), the immediate precursor to ethylene, through synthesis of 1-aminocyclopropane-1-carboxylate deaminase (ACC deaminase) to decrease the ethylene production in stressed plants (Reed and Glick, 2005).

Due to the sensitivity and the sequestration ability of the microbial communities to heavy metals, microbes have been used for bioremediation (Hallberg and Johnson, 2005). Soil microorganisms are known to affect the metal mobility and availability to the plant, through acidification, and redox changes or by producing iron chelators and siderophores for ensuring the iron availability, and/or mobilizing the metal phosphates (Abou-Shanab et al., 2003). The presence of rhizosphere bacteria increased concentration of Zn in Thlaspi *caerulescens* (Whiting et al., 2001) and Ni in *Alyssum murale* (Abou-Shanab et al., 2003). In addition, plant growth-promoting bacteria can improve plant growth, plant nutrition, plant competitiveness and responses to external stress factors (Egamberdiyeva and Hflich, 2004).Therefore, the application of heavy metal-solubilizing microorganisms is a promising approach for increasing heavy metal bioavailability in soils.

The recent researches of PGPR on the remediation of contaminated soils show a brilliant prospect for the successive studies. For example, the application of *Azotobacter chroococcum* HKN-5 can stimulate plant growth and protect *Brassica juncea* from Zn and Pb toxicity (Wu et al., 2006b).

Also the application of some rhizobacteria can increase uptake of Ni from soils by changing its phase (Abou-Shanab et al., 2006). The major objectives of the present study were to evaluate the potentiality of *P. putida* and *P. fluorescens*, with ACCD activity, to plant growth promotion and Zn uptake in barley and canola in order to improve the efficiency of phytoremediation of Zn-contaminated soils.

Materials and Methods

Preparation of bacterial inoculum

Three strains *Pseudomonas potida* strain 11 *(P.p.*11*)*, *Pseudomonas potida* strain 4 (*P.p.*4) and *Pseudomonas fluorescence* strain 169 (*P.f.*169) were isolated from the roots of wheat plants in the land of Iran. In particular they utilized ACC as the sole nitrogen source and produced indole acetic acid (IAA) and siderophores. The characteristics of bacterian strains as shown in table 1.

Table 1. Characteristics of bacterial strains

Bacterial strains	ACCD activity	IAA production ($mg\ kg^{-1}$)	Siderophor e production (mm)
P.putida strain11	+	7.67	1.6
P.putida strain 4	+	9.6	1.9
P.fluorescens strain169	+	5.8	1.8

P. : Pseudomonas

TSB (Tryptone Soya Broth) medium was used as liquid media in which the bacterial strains were grown. This medium contained different concenterations of Zn (0, 30, 60 mg L^{-1}). The content of light absorption at 405 nm wavelength was determined. The bacteria showed higher resistance to Zn toxicity was selected for further study. The bacteria were grown in TSB medium and incubated at 30 ^{o}c on the rotary shaker at 250 rpm for 24 h. Cells in the exponential phase were collected by centrifugation at 1500 g for 10 min, washed twice with sterile distilled water and recentrifuged. Bacterial inocula were prepared by resuspending pelleted cells in sterile distilled water to get an inoculum density of Ca 10^5 colony forming units (CFU) mL^- (Sheng et al., 2008).

Soil (characterisation, sampling and treatment of soils)

Characteristics of the soil used are described in table 2. The soil was classified Fine Loamy Mixed Typic haplocalcid. The soil was sampled from noncontaminated agricultural fields, air dried and passed through a 2 mm (10 mesh) sieve. Total concentration of Zn initially in the soil measured with an atomic absorption spectrometer (Perkin Elmer 2380; Carter, 1993). To prepare the contaminated soils different concenterations of $ZnSo_4$ (0, 250, 500 mgL^-) were sprayed to the soil. The soil samples contaminated with Zn were wetted for 2 weeks with deionized water to maintain the moisture content at 60% water-holding capacity (WHC) of the soil to enable the metal to reach an assumed steady state as far as possible when added to the samples.

Pot experiment

The experiment consistsed of three levels of Zn and two treatments of control and bacterial inoculation and two plants with three replicates. The pots were conducted in a factorial arrangement based on completely randomized block design. A mixture of 5 kg (dry weight) of artificially contaminated soil and sand was put into plastic pots. The seed of canola (Sarigol CV.) and barley (Karondarkavir CV.) were surface-sterilized with a solution of 1.5% (v/v) sodium hypochlorite for 10 min and washed with sterile water. Six steriled seeds of barley and canola were sown in each pot at a depth of approximately 2 cm below the soil surface.

Table 2. Charactristics of the soil used for isolation of rhizobacteria

Soil name	pH	$Caco_3$	EC	OM	P[a]	K[a]	Total content of elements (mg kg^{-1})			
		(%)	(ds m^{-1})	*(%)*			Mn	Cu	Fe	Zn
Typic haplocalcid	7.8	37.5	5	0.85	15	125	354.5	25	185.5	149.5

[a] The datas are given as Phosphorous and Potassium concentration is available content (mg kg^{-1}).

Twenty five mL^- of the bacterial suspension was added to every seed on each pot for the inoculation treatment while the uninoculated control treatment received nothing. The pots were placed in a greenhouse with a 25 ^oc temperature and were watered with deionized one to maintain the moisture content at approximately 60% water-holding capacity (WHC) of the soil.

Analysis of biomass and contents of Zinc

At the end of experiment, plant samples were collected, divided into above-aground shoots and roots and washed three times with deionized water. To determine the dry weight, shoots and roots oven-dried separately at 75 ^oC and were ground to 0.5 mm for analysis. Then 0.2 g of shoots and 0.1 g of roots were digested with the H_2So_4/H_2O_2 method (Harmon and Lajtha, 1999). The digested solution was diluted with deionized water to a volume of 50 m L^- in a flask. The Zn concentration was measured with an AAS.

Two uptake amounts (UA) in aerial and root, and translocation factor (TF) index as defined in Equ.(1) and (2) which were computed from the treatment concentrations will be used to discuss the results from this study.

UA=concentration * biomass (1

TF= the ratio of metal concentration in shoots to that in roots (2

Data analysis

Mean of datas were compared by LSD test at the 5% level. F-values and significance levels of the data were analyzed using General Linear Model (GLM) in SAS.

Results and Discussion

Isolation and selection of metal-resistant bacteria

In order to ensure the success of inoculation by these strains, the resistance to metals of Zn was tested on TSB medium containing different metal concentrations. All the three strains *P.p.*11, *P.p.*4 and *P.f.*169 showed higher resistance when the concentration of Zn in the medium reached 60 mg L^{-1} (table 3), so these three strains were used for future experiments.

Plant growth promotion

Zn tretments decreased root and shoot growth of canola and barley. Significant increases of shoot dry weight of canola were observed when the seeds were inoculated with *P.p.*11 at concentration of 250 and 500 mg Zn kg^{-1}. In barley among the treatments shoot dry weight increased significantly when the soil was inoculated with *P.p.*4 at the Zn concentration of 250 mg kg^{-1} and with *P.p.*11 at the Zn concentration of 500 mg kg^{-1} compared to the soil without inoculation (table 4). In both canola and barley the maximum mean shoot biomass were obtained in the pots were inoculated with beneficial bacteria special with *P.p.*11.

This approach was in agreement with previous observation that inoculation of rhizosphere with PGPRs increased plant biomass in *Brassica juncea* grown on contaminated soils (Belimov et al., 2001). The study demonstrated that *P.p.*11 could facilitate barley and canola growth. Bacteria that could produce IAA and siderophores are capable of stimulating plant growth and helping plants acquire sufficient iron for optimal growth (Rajkumar et al., 2006). Moreover, PGPRs have been reported to influence plant biomass production by lowing plant ethylene synthesis through 1-aminocyclopropane-1-carboxylic acid (ACC) deaminase activity (Glick et al., 1998).

The content of shoot dry matter in the canola with *P.p.*11 treatment was the highest (6.28 g pot^{-1}) ($p<0.05$). There were no obvious differences in root dry weight of canola and barley between the the inoculated plants and the plants without inoculation (table 4).

Metal uptake

There was no significant difference in shoot Zn uptake of barley and canola between bacterial inoculation and without bacterial inoculation. Significant increases ($p < 0.05$) of root Zn uptake of canola and barley were observed when the soil was inoculated with *P.f.*169 at the Zn level of 250 mg kg^{-1} and with all the three strains at the Zn level of 500 mg kg^{-1} compared to the without bacterium-inoculated soil, respectively (table 5).

Table 3. Bacterial growth in different concentrations of heavy metals (light absorption at 405 nm wavelength)

Bacterial strains	**Zn(mg kg^{-1})**		**blank**
	30	***60***	
P.putida **strain11**	2.74	0.024	2.63
P.putida **strain 4**	2.74	0.013	2.88
P.fluorescens **strain169**	2.70	0.02	2.70

P. : Pseudomonas

Table 4. The effect of different bacterial strains on the dray weight (g pot^{-1}) of canola and barley, at different concentration of heavy metals (mg kg^{-1})

plant	**Bacterial strains**	**Zn (mg kg^{-1})**					
		0		**250**		**500**	
		Root	**Shoot**	**Root**	**Shoot**	**Root**	**Shoot**
canola	***P. p. 4***	0.55a	4.37a	0.51b	3.90b	0.42a	3.99b
	P. p. 11	0.58a	5.08a	0.58ab	6.28a	0.48a	5.27a
	P. f. 169	0.55a	4.96a	0.65a	4.71b	0.47a	3.91b
	Control	0.65a	4.58a	0.55ab	4.58b	0.51a	4.32b
barley	***P. p. 4***	0.37a	3.33a	0.27a	3.37a	0.26a	2.18ab
	P. p. 11	0.42a	2.91a	0.30a	1.94b	0.24a	2.35a
	P. f. 169	0.38a	3.08a	0.35a	2.00b	0.26a	1.63ab
	Control	0.45a	3.21a	0.37a	1.99b	0.23a	1.42b

P. f.: Pseudomonas fluorescens, *P. p.*: Pseudomonas putida

The maximum Zn uptake in the above-ground tissues were obtained in canola compared to barley due to the greater biomass. The content of Zn uptake in the canola with *P.p.*11 bacterial treatment were the highest (table 5).

These results showed that inoculation with the bacteria could effectively increase the available Zn in rhizosphere soils. Some studies have demonstrated that heavy metal-resistant bacteria can inhance metal uptake by hyperaccumulator plants (Whiting et al., 2001; De Souza et al., 1999).

Kumar et al. (2008) recorded that the addition of NBRI K28 and its mutant to *B. juncea* increased the level of metal accumulation. Burd et al. (2000) have also recorded similar observations upon inoculation with *K. ascorbata* under Ni, Pb and Zn stress. Similarly appreciable amount of Cr and Zn were accumulated in roots as compared to shoot system as reported earlier by Rajkumar et al. (2006) and Burd et al. (2000), however in this work, it was found that the inoculated shoot systems accumulated considerably more zinc as compared to root systems. This can be attributed to more translocation of metals from the under ground part to the aerial part of the plant. As a consequence, the phytoextraction efficiency of canola was increased with *P.p.*11 bacterial treatment due to the increase in total metal uptake in the harvestable aboveground biomass although it was not significant (table 5).

Translocation factor (TF) index of plants in Zn treatments

TF values can describe movement and distribution of heavy metals in plants. TF values were studies in each treatment. The values of TF showed that significantly there was more Zn moved into aerial of canola inoculated with *P.p.*11 in the treatment of Zn500 mg kg^{-1} than barley and TF of plants tended to decrease with the increasing of Zn (fig. 1). The results showed that inoculation canola with *P.p.*4 and inoculation barley with three strains decreased significantly TF index at the concentration of 500 mg kg^{-1} Zn(fig. 1). It was apparent that canola had better ability of bioaccumulating Zn in contaminated soil than barley.

The abilities of enrichment by plants were different among species. The level of heavy metals in plants might be affected by several physiological factors of plants, including heavy metals uptake from the solution, xylem translocation from root to shoot and sequestration of them (in subcellar compartments or as organic complexes) (Hart et al., 1998).

5. Influence of bacterial inoculation on shoot and root metal uptake (mg pot^{-1}) of canola and barley, at different concentration of Zn (mg kg^{-1})

plant	Bacterial strains	Zn (mg kg^{-1})					
		0		250		500	
		Root	Shoot	Root	Shoot	Root	*Shoot*
canola	*P. p. 4*	0.032a	0.168bc	0.052c	1.664a	0.097a	1.84a
	P. p. 11	0.03a	0.255a	0.065b	2.364a	0.075a	2.46a
	P. f. 169	0.046a	0.205ab	0.101a	1.950a	0.092a	1.929a
	Control	0.064a	0.137c	0.057bc	1.958a	0.089a	1.836a
barley	*P. p. 4*	0.027a	0.103a	0.081ab	1.494a	0.126a	1.35a
	P. p. 11	0.039a	0.097a	0.064b	0.724b	0.118a	1.35a
	P. f. 169	0.03a	0.094a	0.096a	0.817b	0.111a	0.951a
	Control	0.032a	0.101a	0.083ab	0.919ab	0.072b	0.878a

P. f.: Pseudomonas fluorescens, *P. p.*: Pseudomonas putida

Bioaccumulation depends not only on the characteristics of the organism itself, but also on the characteristics of the substance and the environment factors (Niu Zhin-Xin et al., 2007). One recent study of heavy metals translocation into peanut fruits provided evidence that accumulation occurred predominantly via the phloem (Popelka et al.,1996).

In a word, the strong Zn accumulation in the shoot indicated that inoculation canola with *P.p.*11, that has ACCD activity, is potentially useful for remedying Zn-contaminated sites.

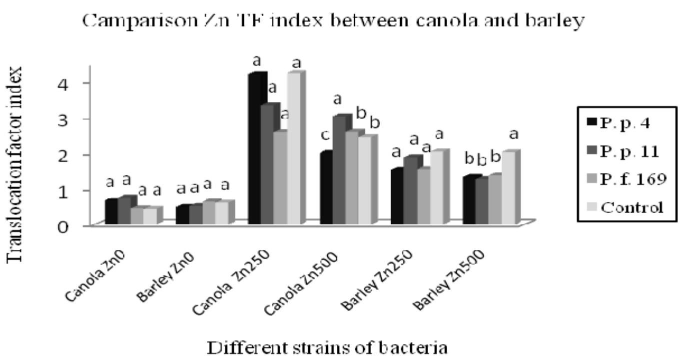

Figure 1. Comparison zinc TF index between canola and barley at different concentration of Zn

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Long Term Effect of Metal Pollution in the Catchment Area of Tisza River

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Abstract
In January and March 2000 two tailings dam failures occurred in the upper Tisza catchment area near Baia Mare and Baia Borsa (Romania). These accidents focused attention on the metal pollution of the Tisza catchment area, and the short term effects of them were studied by many researchers. The aim of this study was to evaluate the long term effects of these pollutions by determining the Lakanen-Erviö extractable easily available metal contents of samples collected in 2011 from floodplains and pastures along the Tisza (Tivadar, Vásárosnamény, Rakamaz, Tiszacsege), and comparing them to our earlier results. Cu and Zn contents were measured by Optima 3300 DV ICP-OES (Perkin-Elmer). The measurement of Pb and Cd was conducted by QZ 939 GF-AAS (Unicam) in 2000 and by an X7 ICP-MS (Thermo Fisher) in 2011.
We found that the Cd, Zn and Pb contents of the pasture near Vásárosnamény exceed limit values and natural background values. In addition, during a 11 year period the easily available Cd, Zn and Pb contents increased significantly, suggesting that the hazard of this pollution should not be neglected.

Keywords: Tisza River, heavy metal pollution, Lakanen-Erviö extraction

Introduction
The Hungarian section of the Tisza River is highly influenced by metal pollution sourced from mining activity in Romania. Mining in Maramures County (former Máramaros) traditionally exploits host ores of base metals (Cu, Zn and Pb) and precious metals (Au and Ag). Besides, metal pollution has also a long history in the upper Tisza catchment (Nguyen et al., 2009). Nowadays the processing of old tailings pond material by using cyanide to recover Ag and Au is spreading in this region. Mining activities that use dangerous and toxic chemicals such as cyanide can be serious sources of contamination. Besides, wastewater may contain heavy metals associated with fine-grained sediments. However, metal concentration of river water is not remarkable 30 km downstream from the point sources (Macklin et al., 2003), sediment-associated metals are dispersed much greater distances.
In January and March 2000 two tailings dam failures occurred in Baia Mare (Nagybánya) and Baia Borsa (Borsabánya) and resulted cyanide and metal pollution in the Lápos - Szamos - Tisza and metal pollution in the Visó - Tisza river systems, respectively (UNEP, 2000).
The short term effects of the pollution events were studied by many researchers, and water and sediment of the Lápos-Szamos-Tisza and Visó-Tisza river systems were found to be contaminated by Cu, Zn, Pb and Cd (Bird et al., 2003; Brewer et al. 2003; Győri et al., 2003a, Macklin et al. 2003; Osán et al., 2002; Wehland et al., 2002).
The mining accidents were followed by floods; therefore the metal pollution of the floodplains were also observable (Kraft et al., 2006; Győri et al., 2003b). Deposition of contaminated sediment on floodplains during flood events and the mobilization of the pollutants may increase the plant available metal content of the upper soil layer.
The mobility and phytoavailability of metals depend on their chemical forms (Kabata-Pendias and Pendias, 2001). Hence, the river sediment was measured by sequential extraction procedures (SEP). Bird et al. (2003) by the BCR (Community Bureau of Reference) SEP found that a remarkable rate of Cd and Zn contents of polluted sediments (Lápos, Szamos and Tisza Rivers) were in exchangeable form. Sulfide fraction of Cu and Zn were significant in the Lápos River but this phase decreased by the increasing distance from the source of pollutants. Non-destructive speciation of metals showed that high amounts of Zn and Cu contents of sediment particles and

suspended particles from the main bed of the Tisza near to Tivadar exist in sulfide form (Osán et al., 2002). Kraft et al. (2006) found that by SEP in the floodplains of the Szamos and upper Tisza, Cd and Zn could be found in an easily available form.

The aim of this study was to evaluate the long term effects of these pollutions on the phytoavailable metal content of soils by Lakanen-Erviö extraction of samples collected in 2011 from floodplains and pastures along the Tisza (Tivadar, Vásárosnamény, Rakamaz, Tiszacsege), and comparing these results to our earlier ones.

Materials and Methods

Soil samples were collected in April 2011 by deep drilling with a Nordmeyer drill (Nordmeyer Holland, Overveen, The Netherlands). We sampled the 300 cm deep soil layer in three replications. Sampling sites are represented in Table 1.

Table 1
Sampling sites

Sampling sites	Geographical	River km	Type of samples	Additional information
Tivadar	N 48° 04' 00.6" E 22° 31' 04.8"	709	active floodplain	affected by the 2nd pollution event
Vásárosnamény	N 48° 07' 46.5" E 22° 19' 39.5"	683	pasture	affected by the 1st and 2nd pollution events
Rakamaz	N 48° 07' 43.8" E 21° 26' 28.7"	543	pasture	affected by the 1st and 2nd pollution events
Tiszacsege	N 47° 42' 59.9" E 20° 57' 08.7"	455	active floodplain	affected by the 1st and 2nd pollution events 8 years ago the area was refilled with soil

Soil samples were air dried and sieved (<2mm) for further analysis. The extraction of the easily available metal content was conducted according to Lakanen and Erviö (1871). Cu, Zn contents were measured by an Optima 3300 DV ICP-OES (Perkin-Elmer). The measurement of Pb and Cd was conducted by a QZ 939 GF-AAS (Unicam) in 2000 and by an X7 ICP-MS (Thermo Fisher) in 2011.

All statistical analyses were performed with SPSS (version 13). Significant differences between the metal contents of the soils in 2000 and 2011 were examined by nonparametric, two related samples test (Wilcoxon test).

Results

Lakanen-Erviö extractable Zn, Pb and Cu contents of the studied soils (100 cm deep soil profile) and the limit values (Kádár, 1998) are presented in Fig. 1. According to the easily mobilizable metal contents the Tivadar floodplain considered to be unpolluted. The Zn contents of Vásárosnamény, Rakamaz and Tiszacsege and the Pb content of Vásárosnamény exceed the limit values.

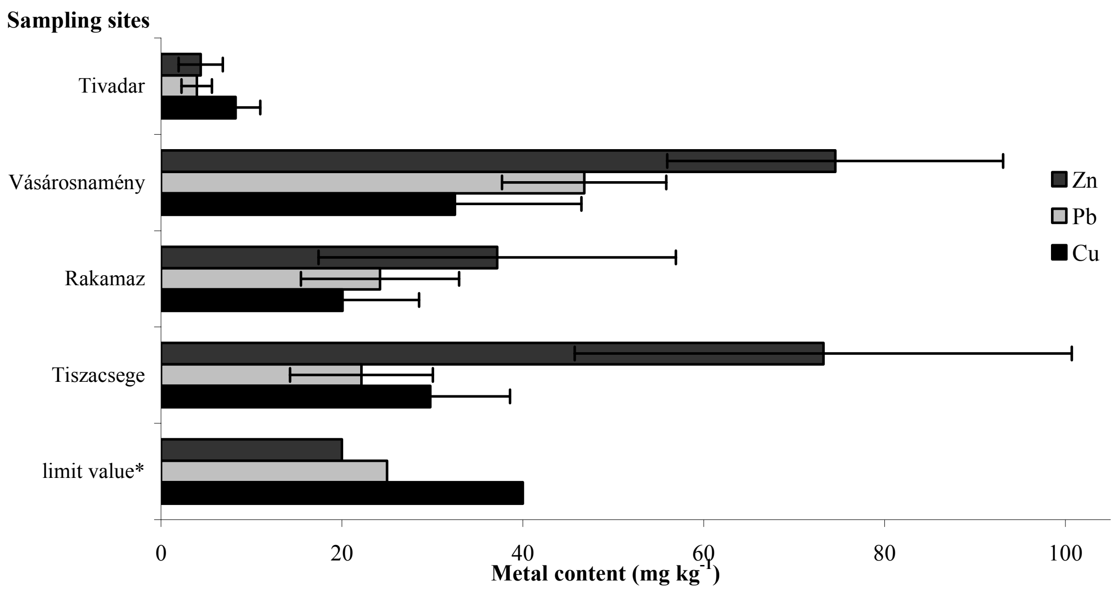

Figure 1. Available Zn, Pb and Cu contents (mg kg^{-1}) of the 1 m deep soil profile (2011)
*Proposed temporary limit value of available metal content for soil (Kádár, 1998)

Cd contents of the sampling sites (Fig. 2) were remarkable in Vásárosnamény, Rakamaz and Tiszacsege. The average Cd content of unpolluted reference soils (Meers et al., 2007) is also presented in Fig. 2. The Cd content of Vásárosnamény was nearly 6 times higher than that of unpolluted soils.

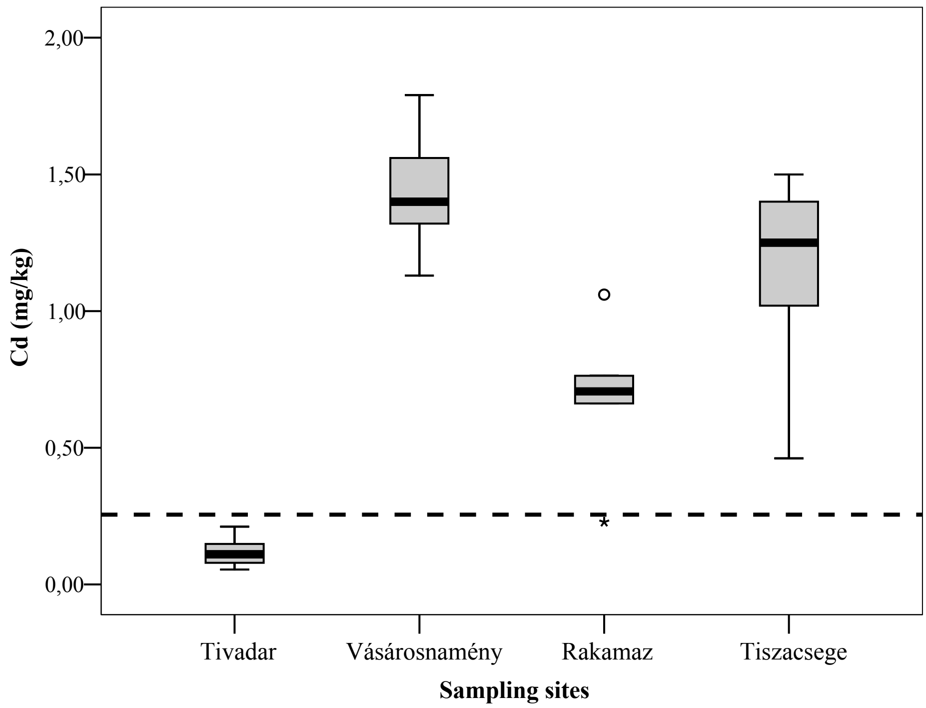

Figure 2. Box-plot of Cd contents (mg kg^{-1}) of the 1 m deep soil profile. Cd level (mg kg^{-1}) in unpolluted reference soils is indicated according to Meers et al. (2007) (0.26 ± 0.04 in soils with pH≤7 and 0.25 ± 0.10 in soils with pH≥7)

* extreme outlier
° outlier

Our results proved that the pasture near Vásárosnamény is significantly polluted with Zn, Cd and Pb. Hence, we compared the metal contents of the 300 cm deep soil profile to our earlier results (Győri et al., 2003b) (Table 2).

Table 2. Changing of the Lakanen-Erviö extractable metal content of the 300 cm deep soil layer of a pasture near Vásárosnamény (2000 and 2011)

Year	**2000**			**2011**		
Depth (cm)	Zn	Cd	Pb	Zn	Cd	Pb
0-10	60	0.75	42	106	1.79	47
10-30	37	0.58	30	72	1.56	42
30-50	34	0.48	28	68	1.38	44
50-70	31	0.45	30	53	1.13	36
70-90	41	0.59	41	68	1.32	51
90-110	40	0.53	38	80	1.42	61
110-140	34	0.39	26	71	1.31	48
140-170	26	0.22	15	43	0.66	12
170-200	19	0.04	7	35	0.48	10
200-230	26	0.20	15	42	0.53	11
230-260	28	0.25	18	80	1.36	38
260-300	17	0.08	11	30	0.56	14

Significant increases were observable ($P \leq 0.05$) in the case of Zn, Cd and Pb contents. The increase is remarkable in the upper 150 cm. A second peak is observable in the curves in 250 cm depth that may refer to a previous major pollution event.

Discussion

Our results proved that the sampling sites affected by the first pollution event (Baia Mare, January 2000) are significantly polluted. Zn, Pb and Cd contents of Vásárosnamény, Rakamaz and Tiszacsege were remarkable.

Farsang et al. (2009) studied the health risk of metals in the topsoil of a Fluvisol soil (pH_{H2O} 7.7, loamy texture) located near the River Tisza. They found moderate risk (if the proportion of vegetables grown in the studied soil is extremely increased in the consumption) when the Lakanen-Erviö extractable Cd, Cu and Zn contents were 0.41, 27.1 and 53.1, respectively. Cd and Zn contents of the studied soils in Vásárosnamény and Tiszacsege exceed these values, which refers to the significant hazard of the pollution.

Sharma et al. (2009) studied the easily mobilizable (Lakanen-Erviö extraction) Cd, Cu, Pb and Zn contents of high, medium and low contamination areas (due to mining activities). Comparing our measurements to these results it can be concluded that the Cd, Cu and Zn contents of the studied soils in Vásárosnamény, Rakamaz and Tiszacsege exceed the average Cd, Cu and Zn contents (0.87, 12 and 26 mg kg^{-1}, respectively) of the low contamination area.

The effect of the second pollution (Baia Borsa, March 2000) on the available metal contents of the Tivadar floodplain was not detectable, which is in accordance with the results of Szabó et al. (2008).

We found that during the 11 year period the easily available Cd, Zn and Pb contents of Vásárosnamény floodplain increased significantly. This may caused by the periodical flood events and the mobilization of the pollutants. Further investigations (solid state partitioning of metals by sequential extraction) are required to find an explanation of this increase.

Acknowledgements

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Groundwater Quality of Assini and Iria Valleys in Peloponnese Region, Greece

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Abstract
The degradation of groundwater quality is mainly related to the intensification of agriculture, the use of fertilizers and the overexploitation of groundwater aquifers which in coastal areas leads to sea water intrusion. An assessment of groundwater quality was conducted in Assini and Iria valleys. Groundwater samples was collected in the beginning (May) and in the end (October) of the irrigation season and subjected to chemical analyses for the main anions and cations. Groundwater was classified using the Piper diagram. Chloride and E.C. (electrical conductivity) contour maps of the regions were obtained in order to evaluate the extent of sea water intrusion. The main cultivated crops in the regions are irrigated citrus and high amounts of nitrogen fertilizers are used. Nitrate concentration of groundwater was found often to exceed the value of 50 mg/l. A comparison was made with the situation that was prevailing in the region eight years ago. The suitability of groundwater for irrigation was evaluated.
Keywords: nitrate, sea water intrusion, fertilizer movement, nitrate

1. Introduction

The quality of groundwater is influenced by natural and anthropogenic factors including climate, geology, land use, irrigation practices. Groundwater in coastal areas is vulnerable to salinization by the intrusion of seawater. Especially in the Mediterranean countries degradation of groundwater quality is a common problem due to multiple pressures on the aquifers, excessive pumping in relation to low natural recharge, return flow from irrigation water with intense use of agrochemicals, leakage from urban areas, land fills, septic tanks (Barraque, 1998; Fornes et al., 2005). Irrigated agriculture is the main consumer of water in the Mediterranean and its expansion in the last half of the century has been remarkable. In coastal areas urbanization and tourism place also pressures on groundwater resources leading to unsustainable overexploitation (Iglesias et al., 2007). In Greece many coastal aquifers are affected by salinization (Petalas, et al. 2009, Voudouris and Daskalaki, 1998; Stamatis and Voudouris, 2003, Lambrakis, 1998).
The purpose of this study is to determine the chemical characteristics of groundwater in two coastal aquifers in Greece, to examine the processes and chemical reactions involved and also to evaluate suitability of groundwater for irrigation and domestic use.

1.1. Study area

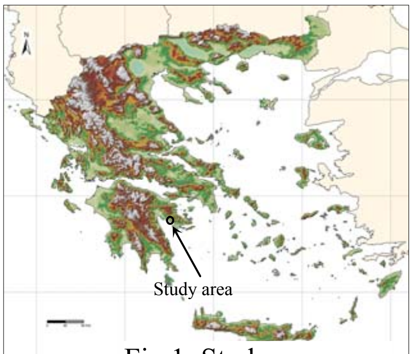

Fig.1: Study area

Assini and Iria valleys are situated southeast of the city of Nafplio and belong to Argolis Prefecture (Figure 1). Iria valley is a coastal plain open towards the Argolis gulf. It is shaped as an isosceles triangle with a direction from east to west the base of which forms the coastal zone. It is run through by Karnazeikos torrent by the erosive action of which it was formed. The watershed of the torrent covers an area of around 200 km^2 and Iria valley, of an area of about 3500 ha, lies in the end part of the watershed close to the sea. The plain lies between the mountainous complexes of Mavrovouni to the north and Didymon to the south. The flat area of the valley lies between 0 and 20 m, the area near the village of Karnezaika extends between 20 to 40 m. In the mountainous area of the watershed elevations more than 800m are encountered. The area falls into the geotectonic zone of Eastern Greece. The limestone formations of "Pantokrator" is surrounded the valley. The more recent deposits of sand and clay of Quaternary covering the lower area is of small or moderate permeability and in some areas they become more coarse and of greater permeability (Figure 2). Upstream of the plain and mainly in the bed of Karnazeikos river coarse grained materials are encountered mainly boulders of different sizes and high permeability. This zone of coarse grained materials is the main recharging zone of the aquifer of the plain. Across the plain a gravel layer has been created and in some areas the depth of this layer is less than 1 m from the soil surface creating soils with moderate depth. Groundwater was the only source of irrigation water and the aquifer of the plain is seriously degraded quantitatively and qualitatively by the intensive and long overexploitation of groundwater.

The Assini valley is situated between Argolis plain and Iria valley. The plain is crossed by a small ephemeral river (Dafnopotamos). Outcrops of the several formations encountered in the wider area are presented on the geological map. Quaternary alluvial deposits are encountered throughout the Assini- Drepano plain overlying the flysch formation (Figure 2). Within the alluvial sediments an unconfined aquifer is developed, its base is the interface between flysch and alluvium deposits ranging in depth up to 50 m.

The mean annual precipitation in the Iria and Assini valleys is 411.68 mm and 494.6 mm respectively, 85% of which occurring mainly from October to April.

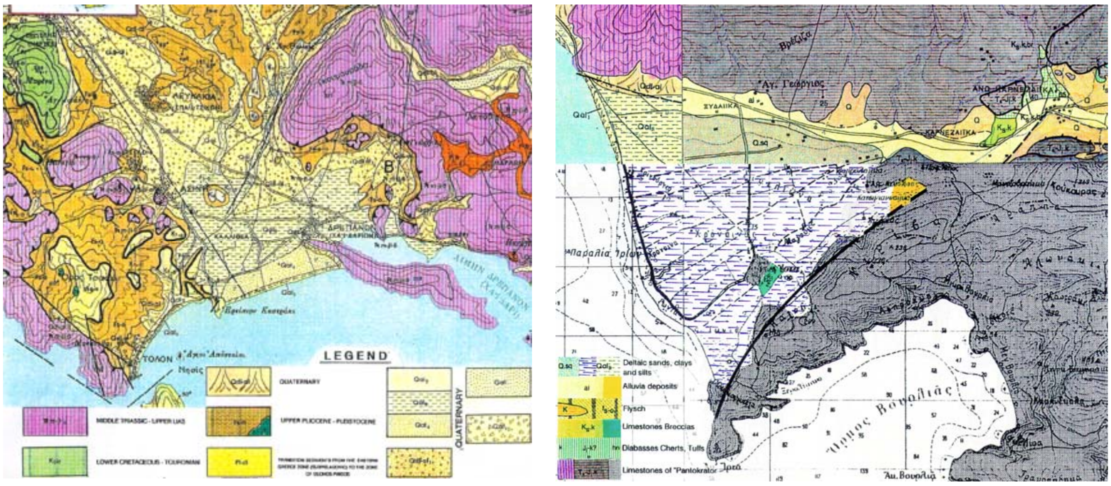

Fig. 2: Geological Map of Assini (left) and Iria (right)

Irrigation agriculture has been expanded in these areas during the last 40 years and the main crops cultivated in Iria regions are vegetables, especially the salt tolerant artichokes that have replaced the cultivation of citrus. A small proportion of the total cultivated area is covered by citrus and olive trees. In Assini citrus trees are the main cultivation, covering 95% of the total cultivated area.

2. Materials and methods

For the assessment of groundwater quality water samples were collected from 40 boreholes in Iria valley and from 25 boreholes in Assini. Groundwater samples were collected in June and October 2009 after the end of the rainy season and of irrigation period respectively. In all collected samples EC, pH and the main anions and cations were determined. Ca^{2+} and Mg^{2+} concentrations were measured by the EDTA titrimetric method, K^+ and Na^+ by flame photometer, CO_3^{2-} and HCO_3^- by titration, SO_4^{2-} by the turbidimetric method, NO_3^- by the cadmium reduction method and Cl^- by potentiometric titration with silver nitrate (APHA, 1989).

3. Results and Discussion

In Table 1 the maximum and minimum values of E.C., pH and ion concentrations of groundwater samples in May and October 2009 are presented for Iria and Assini valleys. Also for Iria valley the values of ion concentrations prevailing in October 2004 are shown. In Iria groundwater was seriously affected by sea water intrusion, EC values varied between 0.737 dS/m and 8.48 dS/m with a mean value of 3.68 dS/m in October 2009. In the majority of water samples (56%) EC values were greater than 3 dS/m. Cl^- dominates the anionic composition of water samples, its values fluctuated from 1.36 meq/l to 70.36 meq/l and with 36% of the samples presented values greater than 30 meq/l. The dominant cations are Mg^{2+} and Na^+ with mean values of 14.01 meq/l and 13.85 meq/l respectively followed by Ca^{2+} with a mean value of 8.24 meq/l.

Table 1: Hydrochemical data of groundwater in the study areas (min and max values).

	IRIA			ASSINI-DREPANON	
	Oct 2009	**May 2009**	**Oct 2004**	**Oct 2009**	**May2009**
E.C. (dS/m)	8.48 - 0.74	7.43 - 0.72	11.28 - 1.46	3.76 - 1.72	3.92 - 0.48
Ca^{2+} (meq/l)	14.9 - 2.2	15.6 - 1.8	35.4 - 4.0	16.9 - 8.5	15.9 - 3.1
Mg^{2+}(meq/l)	40.5 - 3.5	33.6 - 2.8	61.0 - 7.6	17.3 - 5.1	14.3 - 1.2
Na^+ (meq/l)	42.78 - 1.27	41.48 - 1.15	47.92 - 0.81	10.16 - 3.33	11.21 - 0.27
HCO_3^-(meq/l)	11.0 - 1.2	9.6 - 2.0	13.2 - 3.2	5.2 - 1.8	5.2 - 1.4
Cl^- (meq/l)	70.36 - 1.36	57.50 - 1.32	99.2 - 7.60	21.31 - 9.67	21.50 - 0.92
SO_4^{2-}(meq/l)	12.80 - 0.84	11.22 - 0.01	9.38 - 1.06	8.32 - 2.01	11.60 - 0.46
NO_3^- (mg/l)	192.44 - 0.62	235.75 - 17.16	317.6 - 6.57	430.1 - 18.75	365.50 - 7.66
SAR	19.73 - 0.66	18.02 - 0.65	16.22 - 0.27	3.17 - 0.91	3.36 - 0.23

In Assini valley EC values of groundwater are lower than in Iria, varied from 1.72 to 3.76 dS/m with a mean value of 2.70 dS/m in October 2009. The dominant cations of groundwater in Assini are Ca^{2+} and Mg^{2+} followed by Na^+. The dominant anion is Cl^- with a mean concentration of 14.77 meq/l, its concentration varied from 9.67 meq/l to 21.31 meq/l. In Figures 3 and 4 the spatial distribution of EC values and Cl^- concentration of groundwater are presented for October 2009 in Iria and Assini regions respectively.

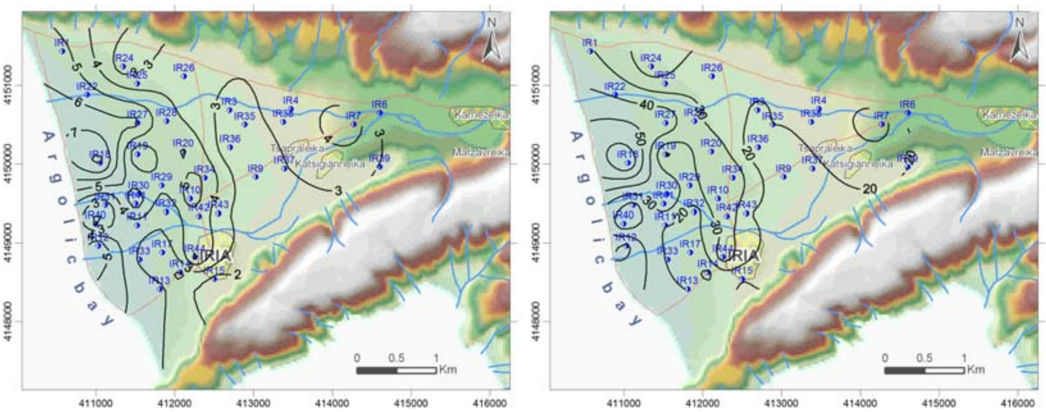

Fig. 3: EC (left) and Cl (right) contours in Iria region for October 2009.

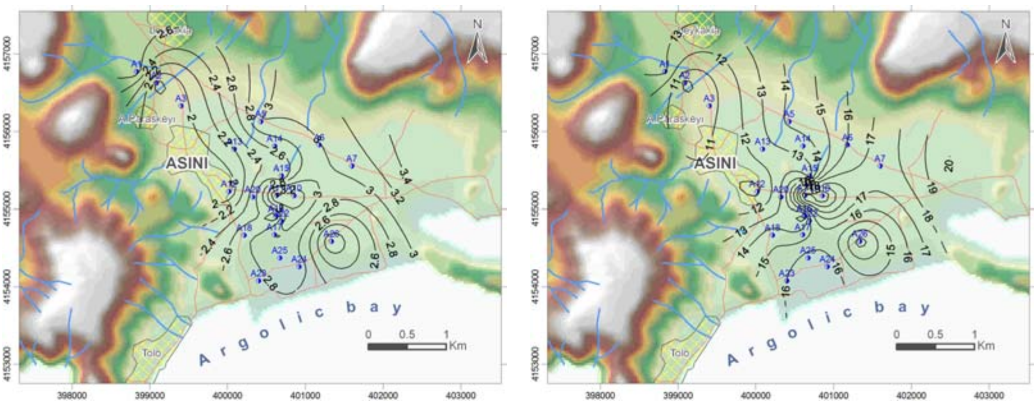

Fig. 4: EC (left) and Cl (right) contours in Assini region for October 2009.

EC and chloride contours are clear evidence of saline intrusion in the two regions since higher values are observed in the coastal zones in the Gulf of Argolis. The area by the sea is consisted of highly permeable materials allowing the intrusion of sea water and the deterioration of groundwater quality. The phenomenon is more pronounced in Iria valley where also in the centre of the valley, high values of EC and Cl^- are prevailing. Groundwater quality in Iria valley was more degraded some years ago (Poulovassilis et al., 1994) as it can be seen from the range of ion concentrations in Table 1, but the transfer of water from Kiveri springs as an additional irrigation source relieved to some extent the pressure in the aquifer from the intense agricultural practices in the region and there is an obvious amelioration of groundwater quality.

In order to examine the relationship between the different ionic species correlations between major ions were performed using Speerman's coefficient analysis. Relative strong correlations, significant at p=0.05 level, were found between EC and Cl^- (r=0.098), Mg^{2+} (0.885), SO_4^{2-} (0.838), Na^+(0.776) and Ca^{2+} (0.72) for Iria groundwater samples in October, implying that groundwater chemistry was mainly controlled by these ions. The EC values in Assini valley are strongly correlated with Cl^- (0.933), Ca^{2+} (0.832) and SO_4^{2-} (0.789). To classify the groundwater and to identify the hydrochemical processes the Piper diagram is used (Figure 5) (Piper, 1944).

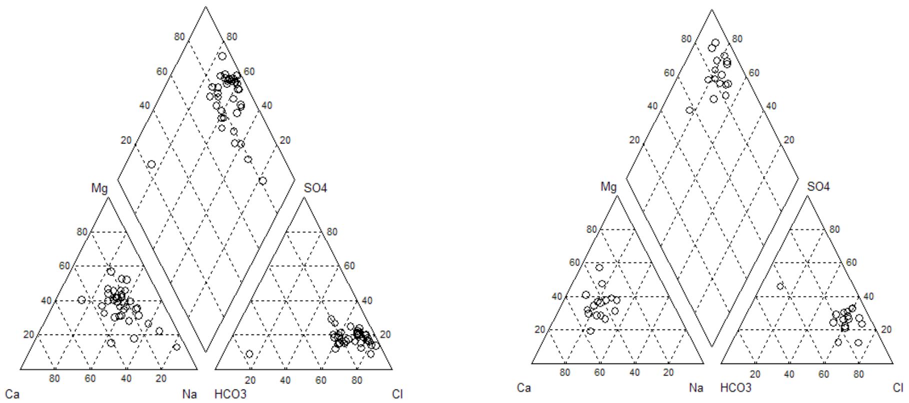

Fig 5: Piper diagram for groundwater in Iria (left) and Assini (right) regions

The most obvious indication of seawater intrusion is the high increase in Cl concentration. The dominance of chloride ions in relation to carbonate ions indicates secondary pollution from sea water intrusion (Todd, 1980). All samples (except one) in Iria had values of the ratio Cl^-/HCO_3^- much greater than 1 and 60% of the samples have values greater than 5 reaching also very high values greater than 15, thus indicating significant saltwater encroachment. In Assini region all samples present values of Cl^-/HCO_3^- higher than 1 and 40% of samples have values between 5 and 10. The existence of Ca-Cl or Mg-Cl or Ca-Cl, SO_4 facies, as the water types of the study areas, in a coastal aquifer indicates the existence of inverse ion exchange (Appelo and Postma, 1994; Lloyd and Heathcote, 1985). The Na^+/Cl^- ratio is also used is an indicator of seawater intrusion. The Na^+/Cl^- ratio in all samples is significantly lower (0.57 in average) compared to the seawater ratio (0.858) indicating reverse ion exchange processes.

Nitrate contamination of groundwater has become an issue of growing concern since intense agricultural practices; high rates of nitrogen fertilizers generally result in low N use efficiency and high N loss through leaching rending the water of the aquifer unsuitable for drinking (Keeney, 1986). Nitrate concentration ranged from 17.66 to 430.1 mg/l in Assini region and from 0.62 to 235.62 mg/l in Iria region during the two sampling periods. The distribution of NO_3^- for the two study regions is shown in Figure 6.

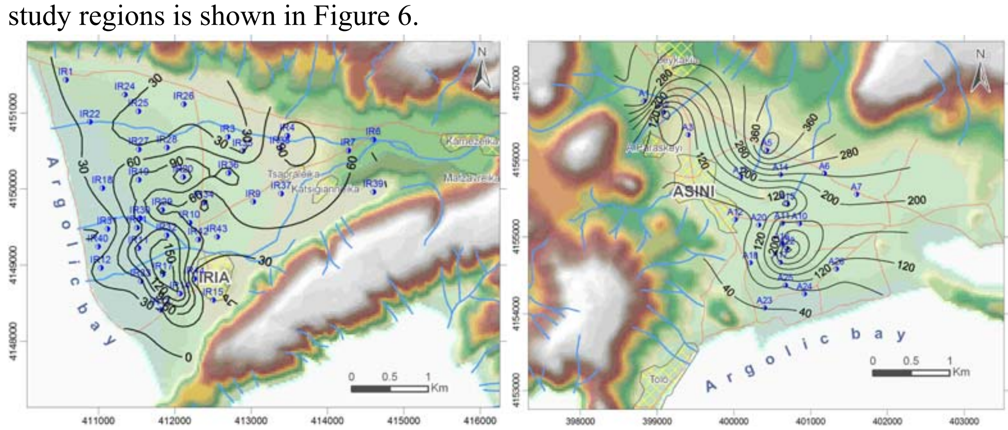

Fig. 6: NO_3^- contours in Iria (left) and Assini (right) regions in October 2009.

In October 2009, nitrate concentration of groundwater exceeded the limit of 50 mg/l set for drinking water (EC, 1998; WHO, 2006) in 42% and 80% of the water samples for Iria and Assini respectively. Groundwater in the two regions is heavily contaminated by nitrates since it is a common agricultural practice the addition of large amounts of nitrogen fertilizer, in an effort to counter the adverse effects of irrigation water salinity on crop yields. Shallow unconfined aquifers as the aquifers in Iria and Assini valley present a high contamination risk of nitrates. Additionally the increased rates of leaching imposed to tackle with the soil salinity and the existence of gravel layer across the valley especially in Iria enhance the leaching and the transfer of NO_3^- to groundwater.
Different schemes for the evaluation of water for irrigation have been proposed (USSL, 1954, Ayers and Westcot, 1988). In Figure 7 the classification of groundwater for irrigation purpose is shown according to USSL, 1954 based on EC and SAR (sodium adsorption ratio) values. SAR (Sodium adsorption ratio) is used to evaluate the sodicity hazard of irrigation water. Excess exchangeable sodium in soils can lead to soil swelling and/or dispersion causing water infiltration, aeration and root penetration problems.

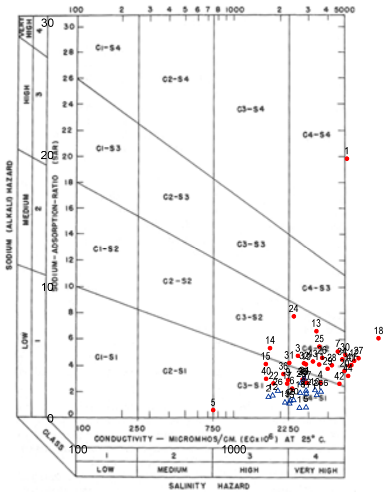

Fig. 7: USSL diagram for irrigation water quality classification
(USSL, 1954) (red circles: Iria, blue triangles- Assini).

From the evaluation of water quality for irrigation according to USSL, 1954 it is obvious that the quality of groundwater is deteriorated and its use for irrigation is severely restricted. Most of the samples are characterized as C4S1 for Assini and C4S2 for Iria regions (Figure 7) presenting a severe danger to develop high levels of soil salinity and important reduction in crop yields. The adverse effects of salts on plants are generally related to the decrease of the osmotic potential of the soil solution reducing the availability of water to plants and thus affecting plant growth and its productivity (Shainberg and Shalhevet, 1984, Tanji, 1990). Excessive concentrations of Cl^- and Na^+ cause also specific toxicities to the crops. Citrus trees are sensitive to salinity and the use of groundwater for the irrigation of citrus trees leads to reduced yields (Maas, 1993). The low to medium levels of SAR value in the majority of samples indicate that soil alkalization problems of dispersion or reduction of the infiltration rate in the soil could not be expected by the use of these waters.

4. Conclusions

Groundwater quality in both areas due to over-pumping and intrusion of sea has been seriously degraded. The higher values of EC and Cl^- are prevailing in the coastal zones but also extent to the central areas of the regions. Cl^- dominates the anionic composition of water samples and is strongly correlated with EC values. EC is also positively correlated with Mg^{2+}, SO_4^{2-}, Na^+ and Ca^{2+}. Ionic ratios and water hydrochemistry show that groundwater degradation is due to extensive seawater intrusion. Hydrochemical facies of groundwater and the low ratio of Na/Cl prevailing in the area reflect reverse ion exchange processes. Groundwater in both regions is also heavily contaminated by nitrates due to intense use of fertilizers.

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Effects of Tillage, Phosphorus Rate, and Irrigation Efficiency on Phosphorus Runoff Losses in a Furrow Irrigated Cotton Field.

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Abstract

Phosphorus (P) additions are vital for profitable crop and livestock production. However, excess losses of P from agricultural lands to watersheds can contribute to eutrophication problems. Therefore, implementation of management practices that minimize erosion and runoff plays a crucial role in P transport. This field study was conducted to investigate the effects of P rates (0, 90 and 135 kg ha^{-1}), tillages (conventional-till (CT), and Chisel-till (ChT)), and irrigation efficiencies (40% and 60%) on P losses (total P (TP), Bioavailable P (BAP), and water soluble P (WSP)) in runoff water from furrow irrigated cotton fields at two different locations (Eyyubiye and Koruklu). Sediment losses, runoff volume and P were summed over the growing season. Significant interaction between tillage and irrigation efficiency for runoff volume in 2009 were found in both locations. Runoff volume was generally highest with low irrigation efficiency (40%) and ChT. Similar interaction was also found for sediment losses in 2009 at Koruklu location as well as in 2010 at Eyyubiye location. Generally the highest sediment losses were with CT and lower irrigation efficiency, whereas the lowest ones were with ChT and higher irrigation efficiency (60%). Phosphorus losses significantly increased with increasing P rate and decreasing irrigation efficiency. For example, P losses in 2010 for Eyyubiye location were ranged from 132.4 to 1298.6 g ha^{-1} for TP, 60.4 to 293.2 g ha^{-1} for BAP, 83.7 to 716.2 g ha^{-1} for WSP. The highest P losses were with low irrigation efficiency and CT, whereas the lowest ones were with high irrigation efficiency and ChT. Overall, the results suggested that increasing P rate increased P losses but no effects on sediment losses and runoff volume which were accelerated by CT and low irrigation efficiency.

Keywords: P losses, P runoff, Eutrophication, Tillage, P transport.

METHODOLOGY FOR THE DETECTION OF CONTAMINATION BY HYDROCARBONS AND FURTHER SOIL SAMPLING FOR VOLATILE AND SEMI-VOLATILE ORGANIC ENRICHMENT IN FORMER PETROL STATIONS, SE SPAIN

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Abstract: The optimal detection and quantification of contamination plumes in soil and groundwater by petroleum organic compounds, gasoline and diesel, is critical for the reclamation of hydrocarbons contaminated soil at petrol stations. Through this study it has been achieved a sampling stage optimization in these scenarios by means of the location of potential contamination areas before sampling with the application of the 2D electrical resistivity tomography method, a geophysical non destructive technique based on resistivity measurements in soils. After the detection of hydrocarbons contaminated areas, boreholes with continuous coring were performed in a petrol station located in Murcia Region (Spain). The drillholes reached depths down to 10 m and soil samples were taken from each meter of the drilling. The optimization in the soil samples handling and storage, for both volatile and semi-volatile organic compounds determinations, was achieved by designing a soil sampler to minimize volatilization losses and in order to avoid the manual contact with the environmental samples during the sampling. The preservation of soil samples was performed according to Europe regulations and US Environmental Protection Agency recommendations into two kinds of glass vials. Moreover, it has been taken into account the determination techniques to quantify the hydrocarbon pollution based on Gas Chromatography with different detectors and headspace technique to reach a liquid-gas equilibrium for volatile analyses.

Keywords: petroleum contaminated soils, petrol stations, soil sampling, electrical resistivity tomography method.

Introduction

Soil pollution due to petroleum products: gasoline, diesel and heavy oils as well as the possible scope of the groundwater bodies is one of the major currently environmental issues for great concern due to the toxicity of some volatile organic compounds (VOCs) and semi-volatile organic compounds present in these fuels. A soil contamination could be produced by petroleum products spill and leaks related to activities of refinement and fuel dispensing at petrol stations. Although pipelines and tanks are designed to avoid this kind of accidents, the large amount of fuel dispensed at petrol stations during the years which are operating may cause a very important damage in the surrounding area.

Electrical resistivity tomography is increasingly being used at sites contaminated by petroleum organic compounds, as an aid in the characterization and monitoring of these sites (Atekwana *et al.*, 2000). Hydrocarbons are excellent insulators and exhibit very high values of electrical resistivity(Delaney *et al.*, 2001). Geophysical techniques can be also used to monitor remedial actions conducted on the environmental sites (Park, 2001).

Some authors have studied different electric responses in the subsurface for the presence of organic substances and they have been proposed several models to interpret the tomographic profiles: an insulating layer model that assumes that the presence of hydrocarbons, usually with a hydrophobic character, in the subsurface can be inferred because of a lower electrical conductivity than the surrounding zone (King and Olhoeft, 1989; Daniels *et al.,* 1992; Endres y Greenhouse, 1996; Campbell *et al.*, 1996). This model attributes a low resistivity to mineral weathering resulting from microbial redox processes in the subsoil. A newly proposed

geoelectrical model for hydrocarbon contaminated sites predicts high conductivities coincident with the contaminated zone as opposed to the traditionally accepted low conductivity (Werkema *et al.,* 2003).
Environmental contamination by diesel and gasoline can be quantified by DROs (Diesel Range Organics) and GROs (Gasoline Range Organics) analysis. The main problem to determine volatile and semivolatile organic compounds in the environmental soil samples is the losses by volatilization of target analytes during the sampling stage and storage. Most determination methods are designed with dry soil samples previously.
In order to minimize losses of analytes and optimize the sampling design, a combined geophysical and geochemical methodology has been designed in this study for former petrol stations.

Materials and Methods

A. ERT 2D at petrol stations

An environmental monitoring was carried out by applying the non destructive geophysical technique: Electrical Resistivity Tomography 2D (ERT) in a petrol station located in the semiarid Murcia Region (SE Spain) (Fig. 1.). The application of electrical tomography technique involves performing variable-length profiles as a function of electrode gap (Bernard, 2003, Martínez-Pagán, 2006). Thus, the survey design requires covering the zone where fuel storage tanks are and optimizing the resolution of the shallow layers by the application of different measurement configurations. In order to minimize the error and to obtain good results, the dipole-dipole and Wenner-Schulmberger configurations were tested in the study areas. After tuning, it was performed a dipole-dipole configuration for measurements due to good results in all petrol stations with a electrode gap of 2 meters, getting a depth of investigation of 12 meters. Dipole-Dipole device is very sensitive to horizontal resistivity changes, but relatively insensitive to vertical changes, so it is useful in vertical structures such as buried walls, pits, and contaminant plumes, but relatively poor in horizontal structures such as sedimentary layers (Lopez *et al.* 2009).

Figure 1. “Petronor” Petrol Station location

Placement of three tomography profiles at petrol station was performed according to the location of the filling mouths in the underground storage tanks (USTs) and to cover the largest possible area (Fig.2). A SYSCAL R1 geophysical resistivimeter by IRIS Instruments (France) has been used at petrol station. After performing field tomographic profiles we proceeded to download the experimental data on a laptop through the PC connection with ProSYS II software. It was made a filtered process and elimination of outliers to obtain a lower error than the permissible for obtaining the best quality results.

Similarly, we proceeded to perform the topographic corrections in those profiles where it was necessary. For the correct interpretation of the results, it was performed a investment process with RES2DINV software, a process based on several iterations comparing the measured results with those calculated by the software configurations taking into account the device used: dipole-dipole. A new implementation of the least squares technique based on quasi-Newton optimization (Loke and Barker 1996) is used for the inversion process in RES2DINV software. As a result of the geoelectric data treatment process, it is obtained a pseudosection representing soil resistivity contours as a function of the depth and length of the profile drawn. The isovalue outlines are defined by a colour scale.

The main purpose of the ERT surveys is a design optimization of the soil sampling at different depths for the physico-chemical characterization and the identification of organic pollutants due to the anthropogenic activity in located areas. The sampling was carried out by mechanical drillings.

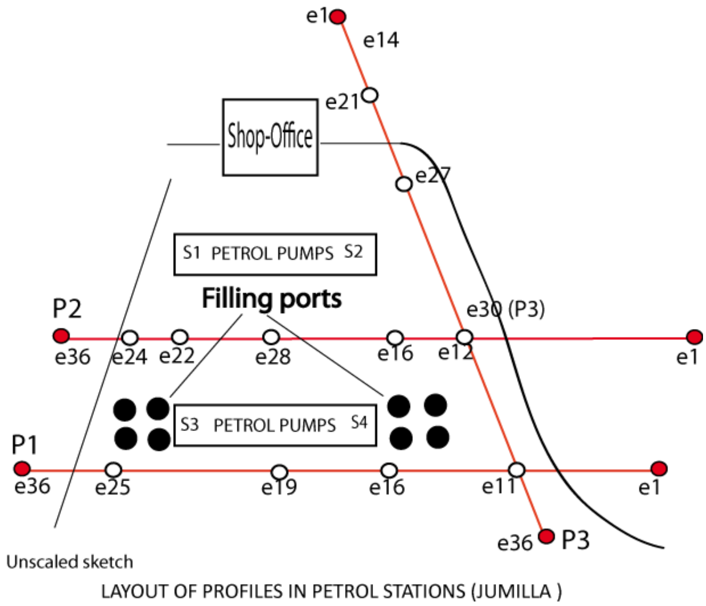

Figure 2. Layout of ERT profiles in the selected petrol station for electrical measurements

B. Handling and Storage: soil samples for VOCs and semi-volatile determinations

In this paper, it's proposed a general method for the sampling and preparation of VOCs and semi-volatile in soil and sediment for further determinations by gas chromatography (GC). Soil samples were taken each meter by mechanical drillings in the selected zones by ERT results and stored into two kind of glass vials at the same time of sampling. For VOCs analysis (GROs), a soil sampler has been designed for this purpose in order not to handling the contaminated soil (Fig. 3). This soil sampler provides different quantities of soil to put in a headspace glass vial containing 10 mL of modified matrix solution (MMS) (Fig. 4.) in order to minimize volatilization losses, until the introduction in a Headspace device previous GC/MS determination (HS-GC/MS) for VOCs. Three samples into headspace vials have been taken from each meter depth. The MMS for soil samples was KCl 250 g/L (Serrano and Gallego, 2006). Headspace vials containing soil samples must be stored at 4ºC in the field. This methodology is applicable to a wide range of organic compounds which have a high volatility sufficiently to be effectively removed from soil samples using an equilibrium headspace procedure.

Figure 3. Soil sampler designed for collection of environmental samples without handling

For semi-volatile analysis (DROs), a composed soil sample from each meter is stored into a 40 mL amber glass vial without headspace, keeping the vial completely full (Fig. 4.). Amber vials containing soil samples must be stored at 4ºC in the field for analytes determination by gas chromatography-flame ionization detector (GC/FID) without previous drying samples.

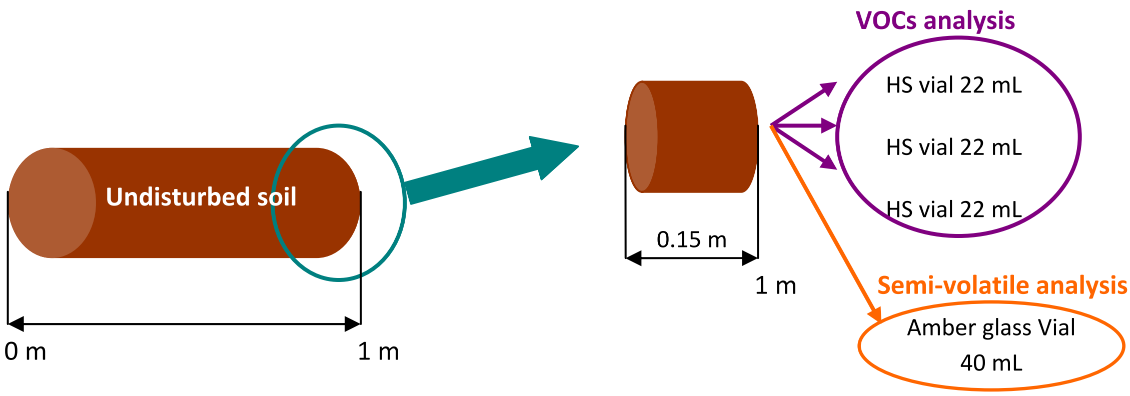

Figure 4. Sampling methodology and handling for hydrocarbons analysis

We have followed the US Environmental Protection Agency recommendations (US EPA 5000) for the cleaning of sampling glass containers and glass material. For semi-volatile analysis, the glass material is rinsed with distilled water-MilliQ® water-acetone twice while for VOCs it is rinsed with distilled water-MilliQ® water-dichloromethane before using them to store samples.

Results and Discussion

Results from the ERT 2D application at the petrol station are shown in Fig. 5. The geoelectrical pseudosection for ERT profile 1 in wet season shows several resistive regions called A, B, C, D, E and F. The A and C regions have moderate electrical resistivity values between 80 Ω·m to 600 Ω·m due to the compressed natural soil influence out of the asphalted zone. On the other hand, D and E regions show resistivity values above 500 Ω·m, getting even 2000 Ω·m. These regions are located between 4 to 5 m depth matching up with the UST filling mouths positions in the surface. These resistivity anomalies could be caused by the UST positions. The F region is a non natural anomaly produced during the data processing step and it can't be related to the presence of any hydrocarbons leak in the subsoil.

In the electrical pseudosection from ERT profile number 2, it must be highlighted the F anomaly with values above 2000 Ω·m because of its size and large expanse. This anomaly is situated between electrodes 14 to 24 where filling mouths and auxiliary services, like piping, are located. A moderate resistivity layer (H) with values between 10 Ω·m to 80 Ω·m is located above F anomaly. The insulating layer model (Sauck, 1998; Atekwana, 2000) assumes that the presence of hydrocarbons, usually hydrophobic, in the subsoil can be inferred due to contaminated groundwater and lower electric permittivity than its surroundings, so, F region could be related to the presence of petroleum organic pollutants in the subsoil.

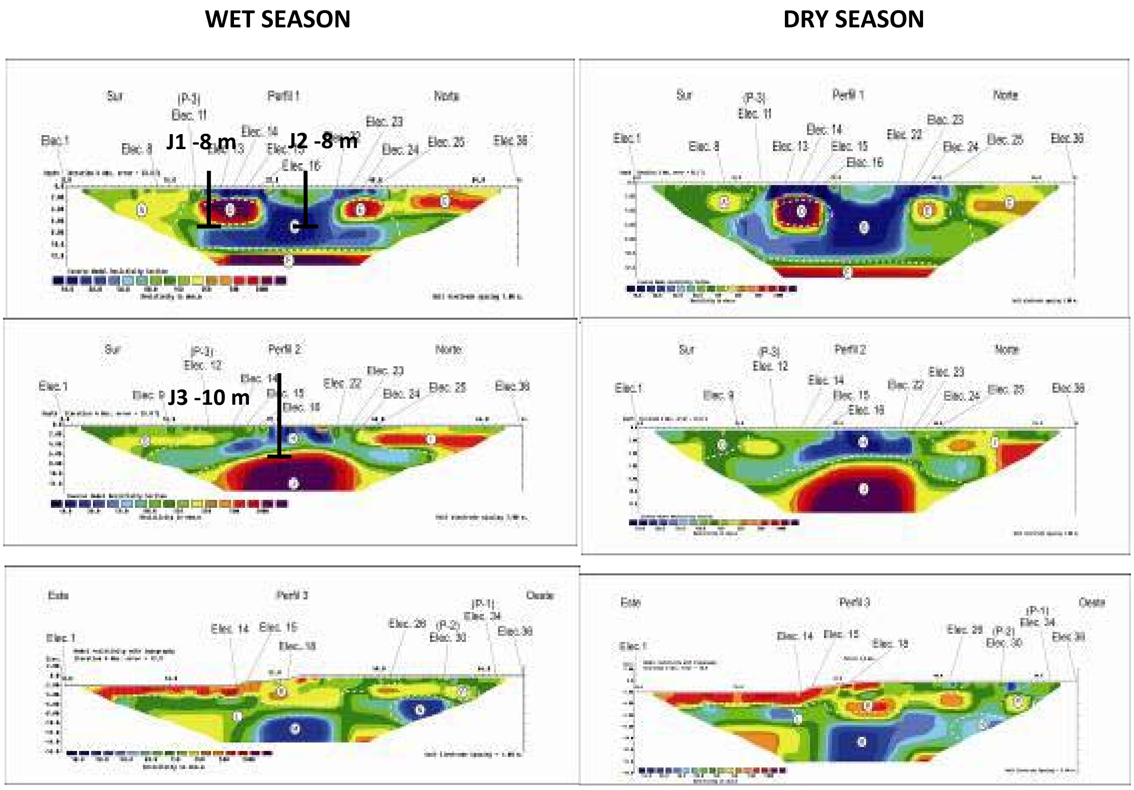

Figure 5. ERT processed pseudosections from “Petronor” petrol station in wet and dry seasons and selected areas for sampling.

Data measurements from ERT profile number 3 have given a pseudosection with two main moderately resistivity anomalies, M and N whose resistivity values are below 50 Ω·m that could be related to old fuel spills. K and L layers, with values between 80 Ω·m and 500 Ω·m, could be produced by different geologic materials and slightly porous but not by the presence of hydrocarbons in this zone.

Following the geophysical interpretation, the sampling design stage should be consistent with these results. Thus, the anomalies B, H and J from ERT profiles 1 and 2 were selected as potential hydrocarbons contaminated areas for sampling, reaching 8 m depth. In this way, if the results of hydrocarbons determinations are negative in the selected soil samples for both drillings, it could rule out the existence of important fuel contamination plumes in the subsoil of the petrol station.

For each mechanical drillhole (J1, J2 and J3), soil samples were taken as it is defined in Fig. 6, using the appropriate containers and the soil sampler designed for that purpose. Subsequently, the GROs and DROs analysis in the environmental samples were carried out by HS-GC/MS and GC / FID analytical techniques, respectively, confirming the existence of some petroleum hydrocarbons in the subsoil below 50 mg/kg.

In short, combining the geophysical technique Electrical Resistivity Tomography with a specific sampling design methodology is presented as an useful tool for the location of USTs and the detection of areas with anomalous values which could indicate the existence of hydrocarbons in the subsoil due to leakage at petrol stations.

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Pollution of Irrigated Arable Land by Heavy Metals

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Abstract

The production of environmentally friendly products in Kazakhstan is an important task of present period. Raw cotton is the main raw material for the textile and food industries. Cotton seeds are used for production of cottonseed oil, which is widely used along with sunflower oil. The quality of produced oil depends on the ecological condition of arable land under cotton. In this connection, we carried out studies on contamination of arable land with heavy metals. Scheme of survey was based on cartographic basis of the farm. Land plot is divided into elementary plots with area of seven hectares. Of each plot the combined soil sample of twenty cm layer was taken. In soil samples taken by atomic absorption method, the content of mobile forms of zinc, copper, lead, cadmium and nickel was identified. According to the obtained results of chemical analysis of soil, the colored cartograms of contamination of arable land were made. Experimental data have shown that a priority pollutant of cotton arable land are lead, nickel and cadmium with a twofold excess of the maximum permissible concentration in the soil. The content of mobile zinc and copper in arable land is insignificant and do not exceed the maximum permissible concentration. In cotton plants, taken from highly contaminated sites, the highest content of lead, nickel and cadmium in stems and bolls is observed, from which these elements can penetrate to cotton seeds. Every year samples of irrigated water were taken and analyzed. Analyses showed that the source of contamination of arable land can be irrigated water that contains lead, nickel and cadmium. In order to exclude contamination of arable land with heavy metals and production of environmentally friendly products it is necessary to improve collector-drainage systems on irrigated gray soils subjected to secondary salinization and conduct monitoring of pollution of irrigation water.

Key words: Soil, environment, pollution, heavy metals, cartogram.

High Arsenic Water Problems in Emet-Kütahya, Turkey: A Review

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Abstract
The largest colemanite reserves occur around the Emet Town which is located in the Midwest of the Anatolia. Two groundwater (drilled) sources (Malı 1 and Malı 2) were supplied to the town for drinking about 20 years ago. Later, mean values of Arsenic contents of Malı 1 and Malı 2 water samples were reported as 448 ppb. and 633 ppb., respectively. The water sources contained much higher levels of arsenic than maximum contaminant level (MCL) set as 50 ppb. in Turkey. Hence, use of these waters was prohibited in Oct. 2001. MCL was lowered to 10 ppb in 2005. A good quality water was supplied to the town center about four years ago. It was reported that geochemical examinations of the Emet borate deposits contained As and S bearing minerals such as: Realgar (AsS) and Orpiment (As_2S_3) as scattered nodules in the colemanite formations. High levels of Arsenic in the groundwaters were considered to be associated with the dissolution of these minerals occurring in the study area. Some skin disorders seen at the inhabitants of İğdeköy Village are linked to the high arsenic contents of drinking waters. Detailed Arsenic analyses of the soil, plant and irrigation water samples are suggested for the region.
Keywords: Arsenic, Waters, Borates, Emet, Kütahya.

Introduction
Since arsenic poisoning via groundwater has become a worldwide problem, a rapid growth in research on arsenic occurence and behaviour has happenned over the last decades. Today, environmental arsenic problems have been very well documented in several publications (Smedley and Kinniburgh, 2004; Smedley, 2006; Ravenscroft et al. 2009; Atabey, 2009). Arsenic is ubiquitous element detected in low concentrations in virtually all environment. Although most environmmental problems are result of mobilization under natural conditions, anthropogenic impacts have been significant in places due to activities such as mining, fosil fuel combustion, wood preservation, use of arsenical pesticides and arsenic based additivites in livestock and poultry feeds. Arsenic occurs as a major constituent in more than 200 minerals including elemental arsenic, arsenides, sulfides, oxides, arsenates and arsenites. Among the most common occurrences in ore zones are arsenian pyrite ($Fe(S,As)_2$), arsenopyrite (FeAsS), realgar (AsS), and orpiment (As_2S_3). Most groundwaters are usually low or very low having arsenic concentrations below the WHO provisional gudieline value for drinking water of 10 ppb. However, in some circumstances they can reach far in excess of these statuary drinking-water limits and thus cause a potentially threat to human health. Groundwaters are generally more vulnerable to As contamination than surface waters because of the interaction of groundwater with aquifer minerals and the increased potential in aquifers for the generation of the physicochemical conditions favorable for As release. Indeed, the majority of the world's reconised As-related health problems are linked with long-term use of high arsenic drinking waters. Some of the best documented and most severe cases of As-contaminated grounwater ocur in aquifers from Bangladesh, China, India, Nepal, Argentina and Mexico. It was also emhasised that 35 million people were drinking groundwater with As concentrations above 50 ppb. And up to 57 millions were drinking water with greater than 10 ppb. (Smedley, 2006). Recognized health problems resulting from chronic exposure to Arsenic in drinking water consists mainly of skin disorders, notably pigmentation changes (melanosis) and keratosis, although skin cancer has also been identified (Smedley and Kinniburgh, 2004; Smedley, 2006). This paper is mainly aimed to review the high arsenic containing water problems, its relation to arsenic bearing minerals in boron mines and health aspects in Turkey.

Arsenic Studies in Turkey
1-Arsenic concetrations in waters and its relatonship to sulfide minerals: The largest colemanite reserves occur around the Emet Town which is located in the midwest of the Anatolia. Two groundwater (drilled) sources (Malı 1 and Malı2) were supplied to the town (population of 20.000) about 20 years ago by the Bank of Provinces (Emet-Kütahya İçme Suyu Tesisi, 2001).

Table.1. Arsenic contents of the groundwaters (Malı 1 – Malı 2) Arsenic Levels, (ppb.) (Emet-Kütahya İçme Suyu Tesisi, 2001)

Source	Sample Size	Minimum	Maximum	Median	MCL
Malı 1	(n=8)	150	634	448	50 (1984)
Malı 2	(n=9)	48	633	384	10 (2005)

The above results indicated that these two groundwater sources contained much higher levels of arsenic than Maximum Contaminant Level (MCL) (50 ppb.) set in Turkey. Hence, use of these waters was prohibited on Oct. 2001. In 2005 the MCL was lowered to 10 ppb. which was set as 50 ppb. in 1984. It was reported by Helvacı and Firman (1976), Helvacı (1984) and Helvacı and Orti (1998) that geochemical examinations of the Emet borate deposits contained As and S bearing minerals such as; realgar (AsS) and orpiment (As_2S_3) as scattered nodules in the colemanite formations. Consequently high levels of Arsenic in the natural waters were considered to be associated with the dissolution of these minerals occurring in the Emet water catchment area (Oruç, 2004; Doğan and Doğan, 2007). A good quality water have supplied to the town about four years ago. The hydrochemical study of the area surrounding the Hisarcık (Emet-Kütahya) colemanite mine showed extremely high Arsenic contamination in groundwater between 0,07 to 7,754 ppm. and the Neogene borate-bearing clay unit that contains some arsenic minerals such as realgar (AsS) and orpiment (As_2S_3) in colemanite ($Ca_2B_6O_{11}.5H_2O$) nodules were responsible for arsenic contamination in the study area (Çolak et al., 2003).

Çöl and Çöl (2004) reported about the arsenic concentrations in the surface, well, and drinking waters of the Hisarcık-Turkey, ranging from no detectable amounts to 3,00 ppm. (mean: 0,46 ± 0,07 Std).

According to Doğan and Doğan (2007), groundwaters in Emet showed the highest total arsenic concentrations varying from 8,9 to 10,7 ppm. The level of arsenic in the groundwater in the area was related to the dissolution of sufide minerals, dissolution or desorption from iron oxyhydroxides and/or up flow of geothermal water. It was also indicated that arsenic in groundwater also effects ecology. For example only Juriperus oxycedrus and J. varoxycedrus types of vegetation were observed in locations with the highest levels of arsenic in the region. Branches and roots of these plants enriched in arsenic.

Gemici et al., (2008), studied the environmental impacts of borate mines in Bigadiç District which are the largest colemanite and ulexite deposits in the world. They indicated that arsenic was the major pollutant in the groundwater samples, concentrations varying from 33 ppb. to 911 ppb.

Geochemical study of Emet-Hisarcık Boron Basin revealed that average arsenic contents of seven drinking water samples was 0,17 (± 0,257) ppm., (Özkül et al., 2008).

Gündüz et al., (2010) indicated that in 27 wells drilled in the superficial aquifer of the Smav Basin has yielded an average As concentration of 99,1 ppb. with a maximum of 561,5 ppb. It was also noticed that death statistics from 1995 to period 2005 period collected from the study area has revealed increased gastrointestinal cancers above Turkish average. They are some cities in Turkey that drinking waters contained higher amounts of arsenic than permitted level of 10 ppb. It worths to mention that three natural water sources supplying drinking water to the inhabitants of Geater Municipality of İzmir were found to contain As levels 59 ppb. in Göksu, 32 ppb. in Sarıkız, and 13 ppb. in Halkapınar springs until 2008 (Başkan and Pala, 2009). Nowadays, following the treatment of these water sources with very high costs, arsenic levels of drinking waters are reported to be around to 5 ppb. in İzmir Metropolitan, as reported by the Greater Municipality web. sites. On the otherhand, disposal of large amounts of arsenic sludge poses another environmental probems.

2-Health Aspects: Arsenic-related skin lesions in a palm and sole of an inhabitant of İğdeköy Village in Emet was first reported by Bariş (2003). It was indicated that people of Dulkadir Village of Tavşanlı-Kütahya (located up of the Ag mining) where the water contained As as 0,3-0,5 ppm. had fewer lesions than those of from İğdeköy people at Emet with As as 8,9-9,3 ppm. arsenic in waters. Among the inhabitants of İğdeköy 30% (33/97) had arsenic-related skin disorders compared to 5,35% (3/56) of Dulkadir inhabitants (Doğan et.al., 2005). After studying on the blood and hair samples of 30 inhabitans of İğdeköy-Emet Şardaş (2009) indicated that exposure to arsenic in drinking water causes serious DNA damage thus leading to carcinogenesis. The recomended levels of arsenic by the WHO are 0,01 ppm. for drinking waters, 0,01-1,0 ppm. in air, and 2-23 ppb. in blood, however, determined values in this research were found as 1,70 ppm. for drinking water, 89,09 ± 0,64 ppm. in hair and 155,42 ± 3,59 ppb. in blood.

Concluding Remarks

About 40 years ago, several university members have indicated the occurence of arsenic containing sulfide minerals such as realgar and orpiment in borate deposits at Emet environs. Presumably, since due attention has not been given to the possibility of arsenic contamination of groundwaters in this water catchment area, two groundwaters (drilled) were brought to the Emet Town Center. These drillings were shotdown after detailed arsenic analyses of the waters, indicating about ten times higher levels of arsenic than official limits (50ppb.) used at that time. Since investment and running costs of arsenic removal are very high especially for small municipalities, mixing possibilities of good quality water with high arsenic waters should also be considered in endemic sites. Disposal of sludge loaded with very high amounts of arsenic can pose another environmental problem that must bu considered throughly in the arsenic treatment project planning.

Arsenic analyses of groundwater, surface and drinking waters, soil and plants are suggested in addition to Emet-Hisarcık-Kütahya, and also in other borate mining areas such as Kırka-Eskişehir, Bigadiç-Balıkesir. Healths of inhabitants of these areas should also be screened. Turkey takes place on Alp-Himalayan geogenic belt with high tectonic activities triggering hot waters with elevated mineral contents. Uncontrolled discharge of wastes of geothermal fluid can contribute arsenic and boron contamination of surface waters in goethermal areas, notably in B. Menderes and Simav Basins which have very fertile soils. Also water, soil and plants around smelting sites such as in Murgul-Artvin should be carefully analyzed for arsenic and other elements.

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Implementation of Nitrates Directive in Turkey

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Abstract
This work is based on the Technical Assistance project for the Implementation of the Nitrates Directive in Turkey*. The nitrates content of waters in Turkey is, in general lower in comparison to several southern and central EU countries. However, it has been observed from available data that the pressure from N fertilization and livestock manure on water quality in certain provinces is rather high. The monitoring net work consists of 1630 monitoring water stations in the whole territory of Turkey, among them 919 for surface and 711 groundwater stations. Elaboration of data received from the Ministry of Food, Agriculture and Livestock (MFAL) indicate high variation of nitrates in both surface and ground waters. Nitrates Vulnerable Zones (NVZs) and Potentially Nitrates Vulnerable Zones (PNVZs) were dessignated, Codes for Good Agricultural Practices (COGAPs) are under preparation, and a rational nitrogen fertilization plan for the main crops will be finalized by the end of July 2012. Fertilization plans take into consideration the following factors: N crops requirements, expected yield, soil texture, mineralized nitrogen, quantity of inorganic nitrogen in the root zone before sowing date, nitrates inputs from irrigation, inorganic fertilizers and manure, soil slope and nitrates leaching. Thematic maps for the entire country were compiled, presenting spatial and temporal variation of nitrates at river basin level and also at province level. Mean values of nitrates in groundwaters higher than 50 mg/l were observed in 5 provinces and those higher than 15 mg/l in surface waters were recorded in 13 provinces. The monitoring network is suggested to be more representative and effective, and the density of sampling points (especially for groundwaters) should be based on hydro lithological characteristics, geology, geomorphology, soil properties, land use and water exploitaion for agriculture. Improvements in the nitrates monitoring network, will assist the competent authorities of the Ministry of Food, Agriculture and Livestock to implement the requirementd of DIR, 91/676/EEC in order to mitigate pollution caused by nitrates from agricultural activities.

Introduction
Nitrates in surface and groundwater vary greatly across European Countries, and this variation mainly reflects climatic conditions, population distribution and agricultural activities. The Mediterranean regime has a warmer climate than that of the northern Europe which allows more intensive cropping and exploitation of the cultivated land. In addition, the population density in northern European countries is lower and the geological conditions are generally not suitable for intensive cultivation, while the overall level of nitrates pollution is low.
On the way to prepare the accession to the European Union, environment is regarded as one of the most important topics in Turkey. In 1991, the Council Directive 91/676/EEC on the protection of waters against pollution caused by nitrates from agricultural sources was entered into force for the EU members. In the context of adopting and implementing this Directive, the Regulation on protection of the water against nitrate pollution caused by agricultural resources was put into force in Turkey in February 2004 (Official Gazette 18/02/2004, numbered 25377). In this framework, applying Codes for Good Agricultural Practices in the Nitrates Vulnerable Zones, one of the aims is to improve water and soil quality as well as to prevent water and soil pollution caused by nitrates. Another important element is the improvement of the national monitoring network related to ground and surface waters and the provision of information to the policy makers and to the

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public in order to prevent further pollution by nitrates. The implementation process requires a number of technical challenges in Turkey having one of the largest surface areas as a candidate among EU member countries. Many of the Turkish river basins are international and shared by Middle East countries. This situation has an impact on establishing rational water management plans. Furthermore, Action Plans and Codes for Good Agricultural Practices have to be established, soon after the designation of the NVZs. It should be stressed that there is an increased concern from central and provincial authorities for the implementation of nitrates directive which is a challenge for scientists involved in this initiative.

Based on the assessment of the Nitrates Regulation of Turkey it can be argued that the national regulation is a useful starting point for the forthcoming secondary legislation, i.e. establishment of action programme(s) and codes of good agricultural practices. The establishment of a monitoring system from MFAL was a rather good starting point for a rough identification of polluted (or potentially affected) waters in Turkey according to the Nitrates Directive and for the delineation of nitrate vulnerable zones at acceptable certainty.

General information on regions and provinces are available regarding climate, agricultural regions, land use, irrigated areas, areas of major animal and crop production, total use of inorganic fertilizers and animal manure, pressure from agriculture on the environment and areas with intensive farming.

A harmonized and appropriate methodology for assessing the eutrophic status of surface waters is required to be developed. Coordination is needed in terms of covering the different legal obligations (e.g. UWWTD, WFD, Nitrates Directive etc.) and in terms of the distributed responsibilities for these obligations (e.g. Ministry of Food, Agriculture and Livestock, Ministry of Environment and Urbanisation and, the Ministry of Forestry and Water Affairs).

The T.A. project financed by the EU, is implemented by Vakakis International S.A. (GR), and is under the supervision of the MFAL as the main administrative body for nitrate pollution prevention. After the re-structuring, the General Directorate of Agricultural Reforms (GDAR) is the main implementing body. However, limited information is available on the eutrophic status of surface waters which linked to the identification of sensitive zones under the EU Urban Waste Water directive 91/271/EC as published in the Turkish OJ 27271 in June 2009 (Declaration MoEF “Urban Waste Water Treatment Regulation – Sensitive and Less Sensitive Areas Declaration”).

Materials and Methods

Nitrates concentration from 1630 monitoring stations (919 surface and 711 ground water) for the period 2008-2011 that are sampled on average at quarterly intervals or less, were provided by MFAL, and appropriately transfered into an SQL Server, to create a functional database. A set of statistical parameters were calculated for each monitoring point and for every group of monitoring points (i.e. surface, groundwater, river basin level, etc). For those catchments which did not have any water quality monitoring points it was not possible to employ any statistical analysis. The non-parametric method was used to calculate the 90^{th} percentile (P90) nitrate concentration at each monitoring point and whether this exceeded 37.5 mg/l and 15.0 mg/l for ground and surface waters, respectively. On the basis of these threshold values (TV) the NVZ’s and potential NVZ’s were identified, setting as inclusion criteria the exceedence of 20% and 5% of the monitoring points per river basin (or province), respectively.

This Project adopted a threshold value (TV) of 50 mg/l for groundwaters, that is the official Maximum Admissible Concentration (MAC) for potable use in EU, and further considers the 75% of this concentration value, i.e. 37.5 mg/l as the actual criterion upon which pollution is documented. Hence, sustained concentration trends > 37.5 mg/l were looked for. Efforts in the assessment of groundwater quality status, hence the key criterion to designate NVZs, were concentrated in the plains, along the coastal fertile regions, along the Syrian border and the basins in Anatolia. Overall, most regions suggested low impact by nitrates pollution.

Results (preliminary) and Discussion

In areas with increased use of synthetic fertilisers, problems with N pollution of water resources were recorded. The quantities of applied inorganic nitrogen fertilisers varies among provinces depending on crops, and local conditions (Fig. 1). Manure application to land is a common practice in many parts of Turkey and proper management is the key to its efficient utilization as a nutrient resource.

Fig. 1 Provinces with a mean nitrogen application of inorganic fertilizers + manure >170 kg N/ha

Surface waters were classified into the following categories: rivers, lakes, transitional and coastal. In addition, maps with mean nitrate concentration were compiled for surface waters at river basin level (Fig. 2).

Fig. 2 Surface water monitoring stations for nitrates in river basins (2008-2011, n=919 stations)

Mean NO_3^- content in surface monitoring stations in Turkey for the period 2008-2011 (N=919 stations) is 2.94 mg/l), while the respective content in groundwaters (N=711 stations) is 11.68 mg/l. The areas which have been affected by nitrates were calculated. The percentage of Nitrates Vulnerable Zones and the Potential Nitrates Vulnerable Zones for surface and groundwater bodies (Fig. 3) is presented in Table 1.

Table 1 Areas affected by nitrates

Water bodies	NVZs %	Area (km^2)	PNVZs %	Area (km^2)	Total area (km^2)
Surface	3.66	28.569	12.09	94.372	122.941
Ground	5.57	43.478	6.50	50.738	94.216
Total	9.23	72.047	18.59	145.110	217.157

Compiled by Vakakis Consortium,2012

Fig. 3 Map of NVZs and PNVZs at river basin level (n=1630 stations)

Statistical analysis performed in 8,810 water samples (period 2008-2011), indicated that nitrates in surface waters were higher than 15 mg/l in 16.4% of the monitoring stations. In groundwaters, average content between 25 and 50 mg/l was recorded in 6.0 %, while higher than 50 mg/l was observed in 3.6% of the stations. Mean values of nitrates in groundwaters higher than 50 mg/l were observed in 5 provinces and those higher than 15 mg/l in surface waters were recorded in 13 provinces. Water quality has deteriorated in most of the inland water resources mainly due to the lack of appropriate pollution control mechanisms, coupled with failure to enforce efficient legislation (Baltacı et al., 2008). The distribution of nitrate content varies significantly among river basins and this may be attributed to factors such as farming intensity, different fertilization and irrigation practices, livestock density, hydrogeology, soil inclination, soil and climatic factors (Karyotis et al., 2011).

To mitigate nitrates pollution in Turkey, a rational fertilization plan for the main crops in each region is under preparation. The maximum allowable quantity of N fertilization takes into consideration the following factors: N crops requirements, soil texture, mineralized nitrogen, quantity of inorganic nitrogen in the root zone before sowing date, nitrates inputs from irrigation waters, soil slope and nitrates leaching. The groundwater monitoring stations are relatively low and rather evenly distributed over Turkey due to high variability of geomorphology, topography and relief. In view of the relatively low density and uneven distribution of monitoring stations, and taking into consideration its importance for the designation of NVZs, it is suggested once again the increasing of monitoring stations in areas which are influenced greatly by agricultural practices.

The monitoring network is focused on large rivers and there are very few monitoring sites in small rivers. The relatively low density and uneven distribution of monitoring stations for small rivers and streams, taking into account the possibility that these surface waters may be affected by nitrates from agricultural sources, requires a re consideration of the distribution of monitoring stations for surface waters, especially in areas with intensively cultivated land. Sampling a larger number of monitoring sites (wells, boreholes) and incorporating consistent field and analytical data would make trend detection more feasible.

Conclusions

The level of nitrates content in waters in Turkey is in general lower in comparison to EU countries. This may be attributed mainly to the fact that agriculture in Turkey is in general less intensified. However, in certain provinces the pressure from N fertilization and livestock manure on water quality is high. Improvements in the nitrates monitoring network, and the use of hydrogeological knowledge and data, will assist MFAL to identify areas vulnerable and/or polluted by nitrates. It must be underlined that new and more accurate fertilization plans for the main crops in Turkey will assist authorities and public to mitigate the nitrates problem originated from agriculture. The introduction of COGAPs and Action Plans will facilitate proper measures and policies to be applied according to crops requirements and regional edapho-climatic conditions.

With regard to fertilization use in Turkey, from the existing data it can be concluded the following:
- there is a general lower level of nitrogen use across the country in comparison to EU countries;
- fertilization is restricted mainly on arable land and does not cover the extent of the whole agricultural land;
- no systematic handling of manure management is developed, hence once manure is placed on site it is used regardless its usefulness at the particular period of the year.

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SOIL AND WATER POLLUTION

POSTER PRESENTATIONS

Photocatalytic Deterioration of Organic Polutants in Water by UV/ZnO System

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Abstract
In the setting of the waters purification, some molecules appear recalcitrant to the traditional treatments. The exploitation of the properties of some catalysts permits to amplify the oxidization performances with ultraviolet radiance and to damage this pollution by a non biological way. This study was conducted to investigate the effect of a photocatalysis oxidation system for organic pollutants treatment. Oxidative degradation of Tylosin by hydroxyl radicals (OH) was studied in aqueous medium using suspended forms of ZnO and UV light. The results improve that the treatment was affected by many factors such as flow of solution, initial pollutant concentration and catalyst concentration. The reaction kinetics followed an apparent first-order reaction and the removed ratio of Tylosin was 97 % in less than 60 minutes.
Keywords: TOA (Advanced Technique oxidization), oxidization, photocatalysis, ZnO, UV, Tylosin,

Application SWAP Agrohydrological Model to Predict Crop Yield, Soil Water and Solute Transport with shallow groundwater condition

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Abstract

In this study, the effects of different quality and quantity levels of irrigation water on crop yield, soil water and solute transport simulated using SWAP model with shallow groundwater levels condition. Six main crops of the Rudasht region of Esfahan in IRAN such as Wheat, Barley, Onion, Sunflower and Sugar beet were selected. Field experiments were conducted with effect of saline water with different irrigation managements on crop yield, crop yield components and soil salinity at Research Station of Drainage and Soil Reclamation of Rudasht. By using measured parameters for each crop such as meteorological data, irrigation event data, soil profile data, time series of groundwater level, solute transport parameters of soil and time series of crop input parameters, SWAP model that linked with PEST model, was calibrated by inverse modeling. Model calibration was consisted of hydraulic parameters of soil layers, solute transport parameters, and finally crop parameters and crop yield. The objective functions were to minimize the difference between simulated and measured values of soil moisture, saturation extract of saline soil, crop yield and biomass, respectively for each optimization. In order to evaluate SWAP model and its validity, statistical indices such as coefficient of determination, root mean square error, modeling efficiency and coefficient of residual mass were calculated. Result of model analysis showed that with precise calibration, SWAP model is able to predict soil water and solute transport, water table balance and crop yield with high degree of accuracy.

Keywords: crop yield, solute transport, production function, salinity, inverse modeling, SWAP model

Introduction

One of the most crucial worldwide matters of irrigated agriculture is the soil and water salinity. In most arid and semi-arid regions, water deficit makes the quality of water poorer. As for the determination and analysis of different managements on brine irrigation, field tests have been useful, while involving severe restrictions such as limited reliability of the tests in real conditions, shortness of test duration for the determination of some parameters like salinity, and insufficiency of the scenarios being surveyed in field tests(Russo et al., 1986). Simulating models can be considered as a development of field tests to overcome these restrictions. The accuracy of these models relies mainly on the accuracy of the input data and in case of fine calibration; they can be employed to simulate various scenarios of irrigation management with no temporal or spatial restrictions existing in field tests or being costly and time consuming (Mostafazadeh-fard et al., 2009). SWAP, an agro-hydrologic model, is one of crop yield prediction models. This is a comprehensive model for the study of the movement of water, salts and heat in saturated and unsaturated soils comprised of a number of sub-models to simulate water flow and salts and heat transfer in soil, evaporation from soil, plant transpiration, irrigation management and crop growth and yield (Kroes and Van Dam, 2008). Many studies have been carried out to simulate and predict the crop yield and generally they have brought about satisfying results in comparison with the field tests. Bastiaansesen et al., (1996) employed SWAP to study the relationships of the quantity and quality of irrigation water, relative yield and improvement of irrigation management in arid regions. Droogers et al., (2000) also used SWAP to study the effects of qualitative and quantitative changes of water on the soil salinity and cotton crop yield in Rudasht in Isfahan province of Iran. Vazifedoust et al., (2008) made use of SWAP in water deficit conditions in order to increase water productivity for wheat, sunflower, sorghum and sugar beet in Borkhar irrigation network in Isfahan province of Iran. Having calibrated the model, he managed to optimize the water productivity by altering irrigation depth and interval. This study aims to calibrate SWAP for regional crops, predict the crop yield under different irrigation water qualities and quantities, study the movement of water and salts in the soil and set production functions for optimized irrigation scheduling. Six crops including wheat, barley, cotton, sugar beet, onion and sunflower were considered to be the main productions of Rudasht irrigation network in this study. Field tests were conducted on each of the mentioned crops in Rudasht Research Center comprising the effects of different managements of brine irrigation on crop yield, crop yield components and soil salinity. On the basis of the field tests, SWAP was calibrated for each crop using reverse modeling method in different irrigation water qualities and quantities. On this basis, each crop yield and salinity and moisture of the soil profile was estimated using the model. The production-water-salinity functions were derived from the calibrated model for irrigation scheduling.

Materials and Methods

Ruddasht drainage and land reclamation research center is situated in eastern Isfahan, at 52^o E, 32.5^o N and 1500 m altitude, having an arid climate with 120 mm mean annual precipitation. In order to determine the effects of different managements of irrigation water salinity and volume on crop yield and soil salinity and moisture, field tests were conducted in this research center. The tests were done on wheat, barley, cotton, sugar beet, onion and sunflower as the main regional products during several years. Tests on wheat were performed during 2005-2006, on sugar beet during 2007, on onion during 2006-2007, and on cotton, barley and sunflower alternatively during 1996-1998 with together crop rotation. The following treatments were applied to the five crops of wheat, barley, cotton, sugar beet and sunflower: three treatments of irrigation water quality Q_1, Q_2 and Q_3 (2, 8 and 12ds/m EC's respectively), two treatments of leaching LR_0 and LR_1 (no leaching and leaching according to 75% of crop yield), two treatments of irrigation management GU (application of fresh water with 2 ds/m EC until the plant stability and then application of the three mentioned salinities until the end of growing season) and GQ (application of different water qualities during the whole growing season) in four repetitions as a full random block test. As for onion, four levels of salinity (1.5, 3, 5 and 7 ds/m) and approximately invariable irrigation water depth (65 mm each time) were applied. Table 1 shows the measured parameters for each of the crops. The daily meteorological data for each crop was acquired from the Rudasht Climatologic Station. For cotton, barley and sunflower, soil parameters were acquired from the samples taken from the three depths of 0-20, 20-40 and 40-60 cm; for other crops it is 0-30, 30-60 and 60-90 cm.

Table 1. data measured or collection for each crop to simulating

Parameter	Measurement Method	No. of Samplings
Soil Texture	USDA Method	Once
Bulk Density of Soil	Core Sampler	Once
Hydraulic Conductivity in Saturated Soil	Falling Head Method	Once
Soil Water Content	Weight Method	Before Irrigation events
Saturated Extract Salinity	EC Meter	3-7 times
PWP & FC Water Content	Pressure Plates	Once
Irrigation Water Volume (Depth)	Volumetric Counter Gauge	Each Irrigation
Plant Height	Field Measurement	5-6 times
Leaf Area Index	LAI Meter	5-6 times
Root Depth	Field Measurement	5-6 times

SWAP simulating model

Movement of water in soil profile is modeled in SWAP based on the Richards Equation and solved by the numerical method of Finite Differences considering definite initial and boundary conditions.

$$\frac{\partial \theta}{\partial t} = C_w(h)\frac{\partial h}{\partial z} = \frac{\partial}{\partial z}\left[K(h)(\frac{\partial h}{\partial z}+1)\right] - S(z) \quad (1)$$

Where: θ; the soil water content (cm^3 cm^{-3}), C_w; soil water capacity (cm^{-1}), h; water head in soil (cm), K; hydraulic conductivity (cm d^{-1}), S; water volume adsorbed by the root in unit volume of soil in unit time (cm^3 cm^{-3} s^{-1}) and z; depth of soil (cm). The relationships between the soil water content, hydraulic head and hydraulic coefficients in water content function and unsaturated hydraulic conductivity show that these functions have to be determined in order to determine each layer of soil. Soil hydraulic functions are defined as the relationships among hydraulic conductivity (K), soil water content (θ) and soil water pressure head. In SWAP, the analytical functions presented by Van Genuchten (1980) are used for the definition of the soil moisture characteristic curve and the capillary model of Mualem (1976) for unsaturated water conductivity curve.

$$\theta(h)=\theta_{res}+\frac{\theta_{sat}-\theta_{res}}{\left[1+|\alpha h|^{n}\right]^{n-1/n}} \qquad (2)$$

Where: θ_{res}; the moisture residue ($cm^3\ cm^{-3}$), θ_{sat}; saturated moisture ($cm^3\ cm^{-3}$), α (cm^{-1}) and n; empirical shape factors.

$$K(\theta)=K_{sat}S_e^{\lambda}\left[1-\left(1-S_e^{n/n-1}\right)^{n-1/n}\right]^2 \qquad (3)$$

Where: K_{sat} is the hydraulic conductivity of saturated soil ($cm\ d^{-1}$), $S_e=(\theta-\theta_{res})/(\theta_{sat}-\theta_{res})$; relative saturation and λ is an empirical coefficient. To determine the transfer of salts in soil, the equation of salt transfer in soil is used in SWAP; vertically and in saturated and unsaturated conditions:

$$\frac{\partial(\theta c+Q\rho_b)}{\partial t}=-\frac{\partial(qc)}{\partial z}+\frac{\partial\left[\theta(D_{dif}+D_{dis})\frac{\partial c}{\partial z}\right]}{\partial z}-\mu(\theta c+Q\rho_b)-K_r Sc \qquad (4)$$

Where c: solute concentration in soil water ($gr\ cm^{-3}$), ρ_b: soil bulk density ($gr\ cm^{-3}$), q: flux rate ($cm\ day^{-1}$), Q: amount of salt adsorption on soil particle surface ($gr\ gr^{-1}$), μ: the first order rate coefficient of transformation (d^{-1}), S: the root water extraction rate (d^{-1}) and K_r: the root uptake preference factor. Freundlich nonlinear adsorption isotherm is used to determine the amount of solute adsorption on the surface of soil particles. The initial concentration of salts in soil and average concentration of groundwater are taken as the initial conditions while the irrigation and precipitation water concentration is taken as the upstream boundary condition. Downstream boundary condition is defined based on the flow direction and quantity. In order to study the decrease of adsorption as a result of water content decrease and salinity increase, the proposed functions of Feddes (1978) and Mass and Hoffman (1977) are used in SWAP, respectively. The multiplication theorem is employed to calculate the amount of real adsorption by the root in model:

$$S_a(z)=\alpha_{rd}\alpha_{rw}\alpha_{rs}\alpha_{rf}S_p(z) \qquad (5)$$

Where: $S_a(z)$ and $S_p(z)$ are the real flux and potential water adsorption rate by the root ($cm^3\ cm^{-3}\ S^{-1}$), (-) and α_{rd} (-),α_{rw} (-), α_{rs} (-) and α_{rf} (-) are reductive functions due to wet conditions (Oxygen deficit), drought stress, salinity stress and frozen soil conditions, respectively. There exist two algorithms for growth simulation. One is the simple algorithm of Doorenbos and Kassam (1979) and the other one can simulate the plant growth using an advanced model of WOFOST. The merit of the second algorithm is the simulation of real and potential biomass and also real and potential seed yield; while the simple one can only simulate relative yield. In the advanced algorithm, biomass growth rate ($Kg\ ha^{-1}\ d^{-1}$) is determined by the calculation of gross assimilation of the green canopy (A_{pgross}) ($Kg\ CO_2\ ha^{-1}\ d^{-1}$) in optimized condition. This function is exponential as below:

$$A_{pgross}=A_{max}\left(1-EXP(\varepsilon_{PAR}\times PAR_{L.a}/A_{max})\right) \qquad (6)$$

Where: A_{max}: the gross assimilation rate at light saturation ($Kg\ CO^2\ ha^{-1}\ d^{-1}$), ε_{PAR}: the initial slope or light use efficiency ($Kg\ CO^2\ J^{-1}$), $PAR_{L.a}$: The rate of light absorption at depth L in the canopy ($Jm^{-2}\ d^{-1}$). Light consumption efficiency is an index for a biophysical process and also is a function of crop type (C3 or C4) and temperature. Eq. 7 is utilized for modeling the biomass yield:

$$Y=C_e\sum_{t=1}^{N}\left(\frac{30}{44}TT_P^{-1}(t)A_{pgross}(t)-R_m(t)\right) \qquad (7)$$

(TT_p^{-1}) : a parameter to quantify gross real stabilization as a result of the stress for water logging, salinity, dryness or freezing conditions, R_m: maintenance respiration rate of plant ($Kg\ ha^{-1}\ d^{-1}$), N: plant growth length (d), C_e: average partitioning factor for root, stem, seed and leaf ($Kg\ Kg^{-1}$). Total produced biomass is partitioned among plant organs based on the partitioning factors. In order to evaluate the model reliability, a number of model evaluation indexes were used. These indexes include the coefficient of determination (R^2), root of mean square errors (RMSE), maximum error (ME), modeling efficiency (EF) and coefficient of residual (CRM).

SWAP linked to PESTmodel

In this research and in order for optimized parameter estimation, the WinPEST software was used which optimizes the parameters by nonlinear regression method (Doherty et al., 1995). The model divisions are parameter definition and recognition, observed data and eventually model run and parameter evaluation. Unknown parameters were defined in a PEST Template File (PTF). The information such as the number of parameters, observed data and the name of the required files were stored into a PEST Control File (PCF). The addresses of the values of simulation parameters were stored into a PEST Instruction File (PIF) which is linked to a PCF. The model was first run with preliminary estimation of the parameters and was continued iteratively comparing the real data stored in PCF and the simulated data stored in PIF.

Upper and lower boundaries

Meteorological data required for the upper boundary was stored as daily data of maximum and minimum temperature, humidity, radiation, wind speed and precipitation into a file. The lower boundary was the location of water table that the monthly time series of which were input into the model for each crop. The water table was varying between 1.9 m to 2.9 m during the tests. The shallowest water table happened in May and the deepest in August to September.

Optimization of soil hydraulic parameters

The number of soil profile layers was determined based on the soil texture. Soil profile was introduced to the model in two or three layers up to 100 cm depth and the coefficients of soil hydraulic functions (parameters of Eq.s 2 and 3) were input into the model. Amongst the parameters of the soil moisture retention curve and unsaturated hydraulic conductivity, θ_{sat} and K_{sat} were physically sensible and measured. For λ, θ_{res}, α and n, a preliminary estimate was made using transitional functions. Then, the optimization method was employed in order for more precise evaluations. PEST Model was linked to SWAP for optimization of the four mentioned parameters. The target function for the evaluation of the parameters was defined to be minimizing the differences between the values of the simulated and observed moistures (Eq. 8).

$$min\,\varphi(\theta,b) = \sum_{i=1}^{N} w_i \left[\theta_{obs}(z,t_i) - \theta_{sim}(z,b,t_i)\right]^2 \quad (8)$$

Where b (λ, θ_{res}, n, α): vector of unknowns, θ_{obs} (z,t_i) : observed moisture in depth z and time t_i, θ_{sim} (z, b, t_i): simulated values using the vector of unknowns' array, N: number of observations and w_i: weight factors of each observation. In the equation above, it was aimed to find a unique value for b in such a way that the target function is minimized. Unknown parameters were optimized for each of the field tests.

Optimization of solutes transport parameters

Zero concentration of precipitation salts was considered for the upper boundary condition and for the initial condition, concentration of soil salts was considered as a function of soil profile depth in the model. In SWAP, effective parameters on salt transfer in soil are mass flow or conduction, diffusion, hydrodynamic dispersion, adsorption and degradation. The relationship between the concentration and the adsorption of salts is evaluated based on Freundlich isotherm in SWAP. Optimized estimation of the parameters of salt transfer was made in reverse solving method together with linking to PEST model. These parameters were molecular diffusion factors (D_{dif}), dispersivity (λ) and the empirical factors of Freundlich adsorption isotherm (K_d, β). The target function for the evaluation of salt transfer parameters was defined to be minimizing the difference between the simulated and observed salinity of saturated extract (Eq. 9).

$$\min \varphi(EC_e, b) = \sum_{i=1}^{N} w_i \left[EC_{e-obs}(z, t_i) - EC_{e-sim}(z, b, t_i)\right]^2 \quad (9)$$

Where b (λ, θ_{dif}, K_d, β): vector of unknowns, $EC_{e\text{-}obs}$ (z,t_i): soil salinity of the observed saturated extract in depth z and time t_i and $EC_{e\text{-}sim}$ (z, b, t_i): simulated values using the vector of unknowns.

Plant parameters

The advanced model of plant was preferred for crop yield simulation (except for cotton and onion). The most basic input parameters are: time series of plant height, leaf area index, leaf specific area and maximum amount of CO_2 assimilation rate as a function of development stages, decreasing factor of CO_2 assimilation rate as a function of mean daily temperature and minimum daily temperature, partitioning of dry matter, maintenance respiration of organs, maximum rate of leaf area index increase, total daily temperature from germination until flowering and from flowering until maturity, light consumption efficiency, light absorption coefficient, soil suction amounts in terms of water adsorption (parameters of Feddes function), salinity threshold and adsorption decrease gradient. The main plant parameters were measured and input into the model. A part of these parameters was input into the model based on previous studies and literature reviews including weight partitioning, maintenance respiration, parameters of Feddes adsorption function, parameters of Mass and Hoffman adsorption function and light absorption coefficient. The parameters whose values were reported as a range in previous studies were reversely optimized together with linking to PEST. These include SLA, A_{max} and ε_{PAR} for which the target functions were defined as:

$$\min \varphi(DM, b) = \sum_{i=1}^{N} w_i \left[DM_{obs}(t_i) - DM_{sim}(b, t_i) \right]^2 \quad (10)$$

$$\min \varphi(SO, b) = \sum_{i=1}^{N} w_i \left[SO_{obs}(t_i) - SO_{sim}(b, t_i) \right]^2 \quad (11)$$

Where DM_{obs} and SO_{obs}: biomass and seed observed yield respectively, DM_{sim} and SO_{sim}: biomass and seed simulated yield in day t of the crop growth length, N: number of measurements in crop growing season and $b(\varepsilon_{PAR}, SLA, A_{max})$: vector of unknowns.

Results and Discussions

Calibration of SWAP model

To study its efficiency, SWAP was calibrated for each crop. Soil hydraulic parameters (λ, θ_{res}, n, α) for each crop in three depths were calibrated. Figure 1 shows the moisture observed and simulated data for wheat and sugar beet in depths 0-30 and 60-90 cm. Having optimized the hydraulic parameters, the simulated moisture data was in conformity with the observed data during the growth period. In modeling, it was often seen that in the second layer of soil, simulated moisture data is evaluated lower than the observed one. One reason can be texture change and error in the numerical solution of the governing equation. Calibration went on with the parameters of solute transfer in soil for each crop. Figure 2 shows the simulated and observed values of the salinity concentration in the saturated paste for wheat and sugar beet during growth period. Complete conformity was achieved between the observed and simulated values after parameter calibration. The last calibration belongs to plant parameters and crop yields in such a way that the differences between simulated and observed yield were minimized.

Crop yield prediction

Comparison of model yields and field measured yields shows that there exists high correlation between simulated and real data. Model validity statistics were more analogous in this research rather than previous researches. One reason can be complete calibration of the model. In this research, all parameters of the model containing soil hydraulic characteristics, solute transfer parameters and plant parameters were calibrated and optimized using PEST model. In order for model assessment and validity check, statistical indices of R^2, RMSE, ME, EF and CRM were determined. According to the results, the least coefficient of correlation belongs to sunflower (0.877) while the most one belongs to onion (0.982). Although the indexes of coefficient of determination, maximum error and modeling efficiency are acceptable for wheat and sunflower, but higher RMSE's have negative effects on the simulation process. Negative CRM implies that the model evaluates the yield of barley, cotton and sunflower often higher than real amounts while for wheat, sugar beet and onion, the yield is underestimated. Modeling efficiency at all cases is more than 0.8 which means modeling validity.

Determination of Production-Water-Salinity

Functions In order to determine the production-water-salinity functions, a wide range of yield data proportionate to salinity and irrigation depth is required. As a result of the insufficiency of field observations to estimate these functions, calibrated SWAP was used to increase the functions accuracy. Running the SWAP and for rapid alteration of salinity and irrigation depth, SWAP was linked to SENSAN model. Production functions of the selected crops were set for different amounts of salinity and irrigation depth. Mathematical models of 2nd order functions are presented in Eq.'s 12 to 16 for wheat, barley, cotton, onion, sunflower and sugar beet respectively.

$$y = -1221.8 + 204.71 I + 246.09 EC - 1.2009 I^2 - 28.7 EC^2 - 3.30 I \times EC \quad (12)$$

$$y = -1536.8 + 198.07 I + 110.4 EC - 1.2802 I^2 - 14.141 EC^2 - 1.001 I \times EC \quad (13)$$

$$y = 271.12 + 45.513I + 46.29EC - 0.18635I^2 - 9.1356EC^2 - 0.055I \times EC \qquad (14)$$

$$y = 39058 + 1147.4I - 12046\ EC - 4.0132\ I^2 + 618.4EC^2 - 42.121\ I \times EC \qquad (15)$$

$$y = 458.75 + 43.94\ I - 4.65\ EC - 0.2257\ I^2 - 4.5EC^2 - 0.481\ I \times EC \qquad (16)$$

$$y = -3706 + 885I + 834.36EC - 3.319I^2 - 117.8EC^2 - 4.037I \times EC \qquad (17)$$

In the equations above, I and EC represent the depth (cm) and salinity (ds/m) of irrigation water, respectively.

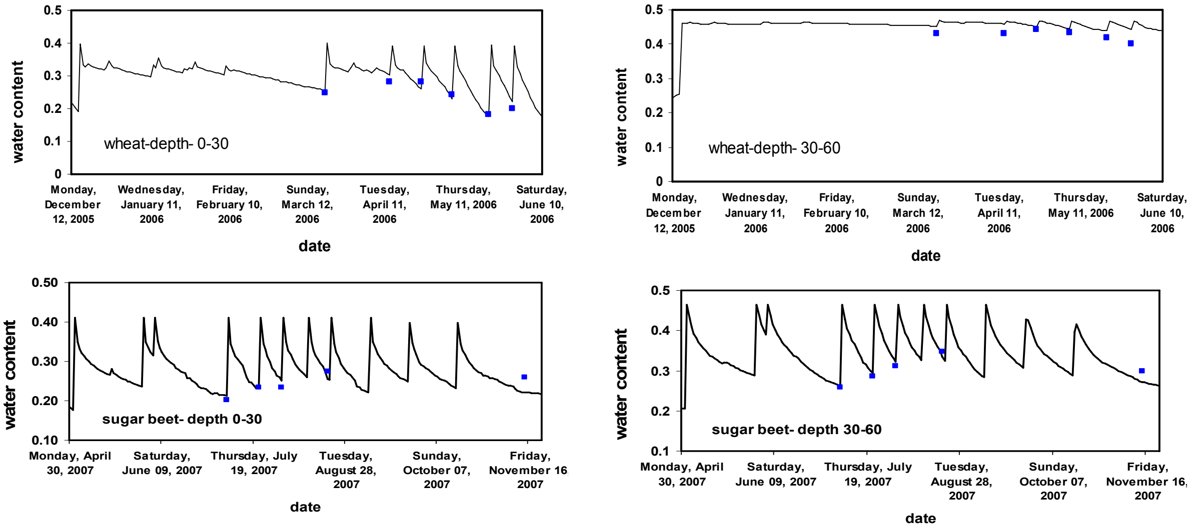

Figure 1. Simulated and observed moisture data (cm^3cm^{-3}) for wheat and sugar beet (Treatment Q_1LR_1-GU)

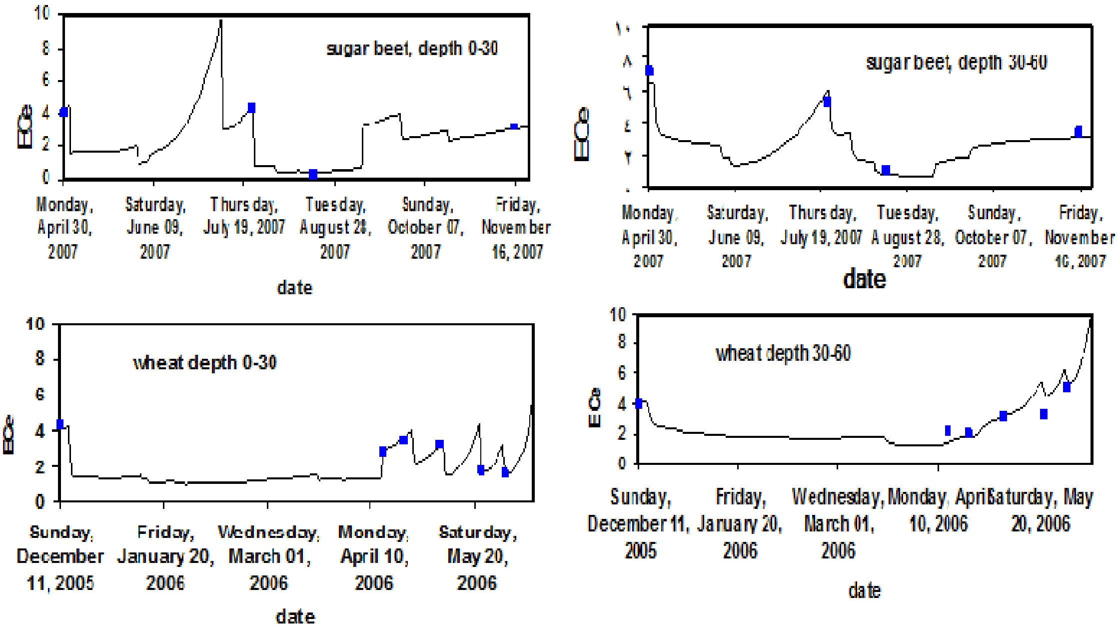

Figure 2. Simulated and observed soil salinity (mg cm-3) for wheat and sugar beet in different depths of soil (for both crops, simulation results are shown for Q2LR1-GU)

Conclusion

Results have shown that simulating models can be employed as a helpful tool for prediction of water and salt movement through soil profile and crop yield under various irrigation water qualities and quantities. According to the results of this research, accuracy of SWAP depends mainly on the accuracy of the input data and in case of complete calibration, it can be employed to simulate various scenarios of irrigation management with no temporal or spatial restrictions existing in field tests or being costly and time consuming. Production functions of the selected crops are the most applied and useful consequences of this study which are used for the determination of optimum irrigation depth for each crop, study of yield assessment indexes and the effects of salinity and water deficit, irrigation management and scheduling under limited quality and quantity of water and, finally, development of iso-yield curves (geometric place of different combinations of salinity and depth of irrigation water) to determine the crop yields under different water qualities and quantities.

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Effect of Different Degrees of Magnetized Water in Estimate of Some Soil Properties

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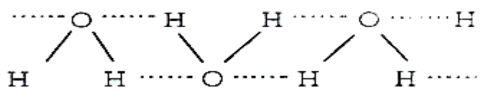
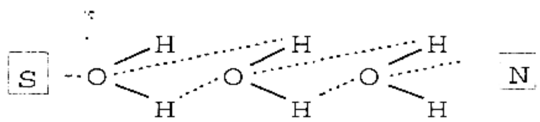
Abstract
Magnetized water is a metaphor, called the water which is passed through a magnetic field, or that is placed inside or close to it for a period of time. The magnetized water increases the totals Quartet Tetra hydra group, which makes it highly effective to dissolve (Colic et al 1998).Soil samples have been taken to be ready for laboratory analysis, after crushed and sieved it by sieve 2 mm, then Designed a device for magnetizing water sequence period to magnetide water for use it first to soil extracts and to estimate some chemical and physical properties of these samples (pH, EC, Ca^{++}, Mg^{++}, Na^{+}, K^{+}) page etal., 1982, as well as for use of this water, with a different degrees of Magnetizing, and in specified quantities to be added into Anvils contain clay soil and receive the filtrate from this Anvils that have periods of sequential filtering, to do the analyzes mentioned above.The results showed that the lowest value for PH is 6.5 at water magnetized for a period 20 minutes, these value at 15 minutes from the times of the filtrate, and the highest value of PH was 8.08 at water magnetized for a period of 10 minutes and is these value at 60 minutes from the times of the filtrate.Ec, Na was the lowest value is 30 mg. kg^{-1}, 1.6 ds . m^{-1} respectively at the same time of filtrate and the magnetization degree (magnetic water for 30 minutes and filtrated time at 60 minutes) While the highest value of EC, Na was 100 mg . kg^{-1}, 10.9 $ds.m^{-1}$ respectively at the same time of filtrate and the magnetization degree (magnetic water for 20 minutes filtrated time at 5 minutes).
Keywords Magnetized water, Soil extracts, Soil properties,

Introduction
Magnetized water is called for the water is passed through a magnetic field or a magnet that is placed inside or close to it for a period of time. The magnetized water increases the Tetra hedral group, which makes it highly effective for the melt, Water molecule is composed of oxygen atoms and tow hydrogen atoms, hydrogen atoms in the molecule attracts water with a single oxygen atoms in other molecules of water by Hydrogen-Bond. In general, in the water molecule there are partial oxygen atom and two atoms of hydrogen linked in isosceles triangle, and it is upper degrees 105.03 , so the molecule shape of the water as shown as:



When water passes through a magnetic field, it will oriented in one direction as shown below



This strong and very intensive magnetic field generated by a magnet tool made a change in water properties as the affect the hydrogen bonds in liquid water, which is heavily influenced by magnetic and electric field, leading to a change in the water properties, whether physical or chemical, causing an increase in the mobility of atoms of salts and therefore the hydrogen bonds will breakdown and adjust the properties of water and make it more capable of Dissolving

Materials and Methods
First:
1 - Soil sample has been obtaining in depth 20 cm for laboratory analysis in 2 mm diameter sieve.
2 - Soil extracts were prepared using a 1:2 water and different magnetic degrees.
1 -The first was extracted using distilled water.
2 - The second extracted using magnetized water for 5 minutes.

3 -The third extracted by using magnetized water for 10 minutes.
4 - The fourth extracted using of magnetized water for 20 minutes.
5 - Fifth extract was used magnetized water for 30 minutes.

The study adopted on a specific mechanism to magnetization the water which has been prepared by the researcher, which consists of:
1 – Water Pam to raise water.
2 -glass basin to collect water magnetic.
3 - Tow electrical pole in 25 per Chaos.
4 - plastic tubes.

Table 1. Specification of pole used in the experiments

Magnetic flux density (Gus.)	DC which passes the pole (ampere)	Diameter a wire (mm)	Resistance wire coiling	Length of the coil (m)	Number of turns	No
25	1.9	30	540	10	1100	1
25	1.9	30	540	10	1100	2

Second
1 - Soils have been placed on four pots (4 kg) and the height of the soil column was 30 cm.
2 - Water was added 200 ml above the field capacity for all samples were as follows:-

a - The first pot added a distilled water.
b - The second pot added magnetized water in 10 minutes.
c - The third pot added magnetized water for 20 minutes.
d - Fourth pot added magnetized water for 30 minutes.

3 - after the soil being in saturation point, will get the infiltrate water leaky from the bottom of pots, then we collected it in plastic containers and in synthesis during the following periods, (5, 15, 30, 45, 60 minutes) of each period separately to made the laboratory analyses for it including:

1 - PH using (PH Meter)
2 - EC
3 - Na using a Flamephotometer
4 - K using a Flamephotometer
5 – Ca^{++}
6 – Mg^{++}

Results and Discussion
First: Table (2) the results of laboratory analysis for the physical and chemical characteristics of soils, the soil texture was clayey, which made a difficulty in obtaining large quantities of infiltrated water in close times, and Table (3) results of the analysis of soil extracts by using a 1:2 water and different time for magnetic water.

Table (2): Some physical and chemical characteristics of soil study

No	analyses	unit	
1	Calcium carbonate	gm. kg $^{-1}$	240
2	Organic matter	gm. Kg^{-1}	19.0
3	Sans	gm. Kg^{-1}	233
4	Silt	gm. Kg^{-1}	350
5	Clay	gm. Kg^{-1}	417
6	Tecsture	Clay	-
7	Field capacity	%	18.75
8	available Potassium	mg.kg $^{-1}$	200
9	available phosphorus	mg.kg $^{-1}$	9.28

Table 3. Some physical and chemical characteristics of soil extracts with five magnetized degrees

No	Time (Sec)	PH	EC dS . m^{-1}	Ca^{+2} Meq . L^{-1}	Mg^{+2} Meq . L^{-1}	Na^{+1} mg.kg $^{-1}$	K^{+1} mg.kg $^{-1}$
1	0	7.71	0.37	1.4	0.2	7	7
2	5	7.6	0.78	2.4	0.4	19	11
3	10	7.75	0.65	5	2.4	16	9
4	20	8.07	0.74	3	0.4	18	10
5	30	8.08	0.69	4.6	0.2	17	11

Second: it should be noted here that the concentrations of the samples were obtained studied in two forms, according to the relationship between the magnetization and the elements concentration for five periods of the nomination, and the second case of the relationship between periods of filtration and the elements concentration for four degrees of magnetization of water.

1 – pH: The results of laboratory analysis of water showed that less value is (6.5) when the water magnetized for 20 minutes and is located at 15 minutes from the time of the filtrate, while the maximum value is (8.08) when the water magnetized for 10 minutes and is located at 60 minutes from the time of extracted, fig. (1). In implementing the graph and make the times of extract on the X-axis and compared it with the values obtained by the graph of the water Representative magnetized for a period of 20 minutes will be mixed in the highs and lows of any volatile, as for the highest value, the graph representing the water magnetized for a period of 10 minutes begins to rise gradually until up to a higher value (8.08) form (2).

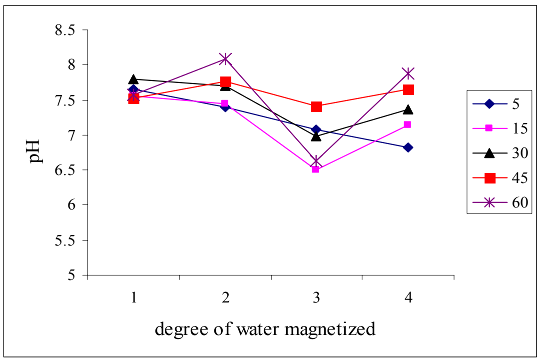


Fig. (1) Effect of magnetize water degrees on the PH values in the different periods of the infiltration

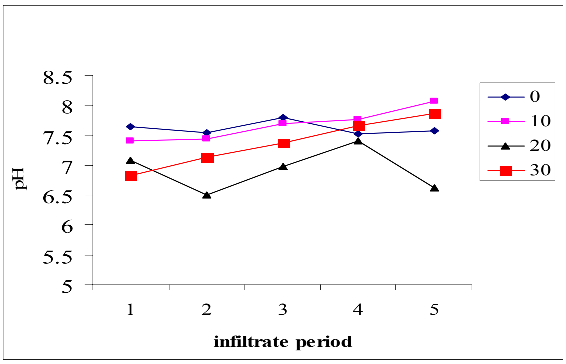


Fig. (2) Effect of periods of filtration on PH values at different magnetic degrees

2 - Electrical conductivity (Ec): In the first case noted that the lowest value was 1.6 dS . L $^{-1}$ at water magnetized for 30 minutes and is located at 60 minutes from the time of the water filtrate, while the maximum value is 10.9 dS . L $^{-1}$ at water magnetized for 20 minutes located at 5 minutes from the time of the water filtrate, and there are a convergence of values of electrical conductivity at a degree of 20 to magnetize periods for water filtrate (15, 30.45, 60) as in Fig. (3).
When makes the times of filtrate on the X-axis, the graph representing the water magnetized for a period of 30 minutes begins to plunge so deeply up to 30 minutes from the time of the filtrate and then returns to rise gradually and then return to his decline at the point of 45 minutes and will decrease untill 60 minutes of time water filtrate . As can be seen that the values of electrical conductivity less for all periods of magnetization and are very close when the amount 60 of the filtrate and this shows that there is a washing large salts per periods of magnetization, especially for the magnetization 20 and 30 occurred in periods of filtration developed 15 to 30 minutes, as shown in Fig.(4).

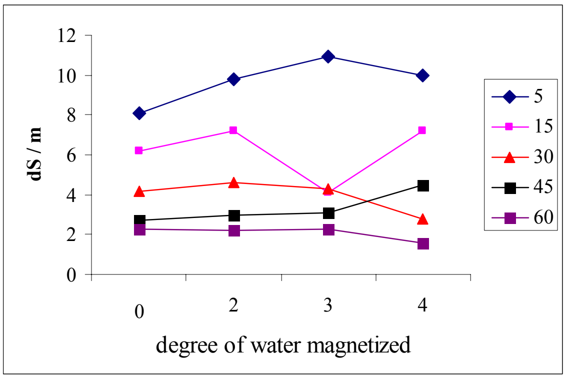


Fig. (3) Effect of magnetize water degrees on the Ec values in the different periods of the infiltration

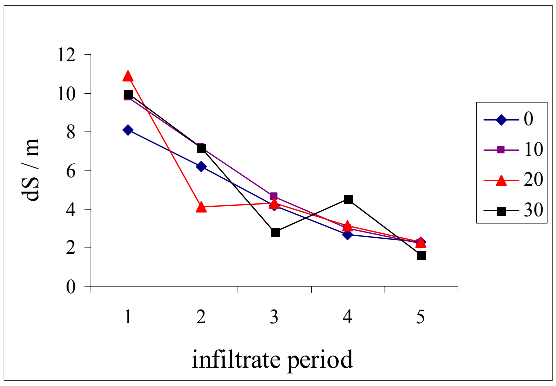


Fig. (4) Effect of periods of filtration on Ec values at different magnetic degrees

3 - Concentration of Sodium ions: The lower value was 30 mg. Kg $^{-1}$in the water magnetized for 30 minutes and is located at 60 minutes from the time of filtrate. The highest value was 100 mg. Kg $^{-1}$ at the water magnetized for 20 minutes and is located at 5 of the times of filtrate and when we make the times of magnetization on the X-axis (5). then when we change the case and makes the times of filtrate on the X-axis and compared it with values obtained by the graph represented by water magnetized for a period of 30 minutes will be a decline of average even up to 30 minutes from the time of filtrate and then settle down gradient to be up to 60 minutes from the time of filtrate, the form (6).

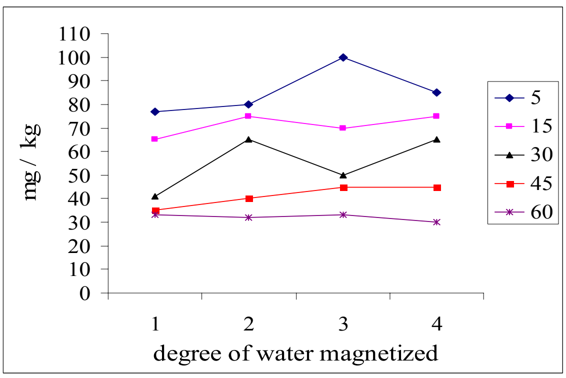


Fig.(5) Effect of magnetize water degrees on Na values at different periods of the infiltration

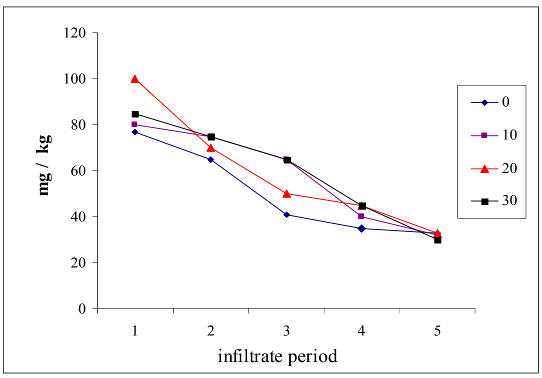


Fig. (6) Effect of filtration periods on Na values at different magnetic degrees

4 - Concentration of Potassium ions: the lowest values was14 mg. Kg -1 at the water magnetized for 20 minutes which is located at 60 minutes from the time of filtrate while the highest value was 36 mg. Kg -1 in distilled water which is located at 5 minutes from the times of filtrate and notes of the fig.(7) that the concentration of potassium in general be less as possible when the degree of magnetization 20 for all periods of filtration. When drawing the relationship and make the times of filtrate on the X-axis, the graphs representing the times of magnetization be a sharp decline with the increase in the infiltration period, with the observation that water magnetized for a period of 20 minutes recorded the highest value when the infiltration period for 45 minutes as shown in Fig. (8).

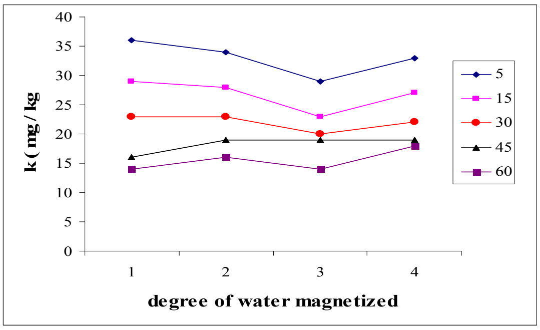


Fig.(7) Effect of magnetize water degrees on K values at different periods of the infiltration

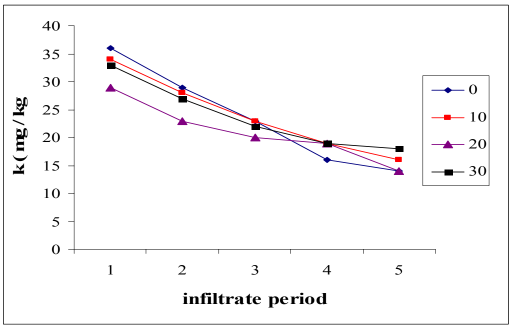


Fig. (8) Effect of filtration periods on K values at different magnetic degrees

5 - Concentration of Calcium ions: Figure (9) showed that the lower value is 25 meq / L at the distilled water and located at the point 60 minutes from the times of filtrate, while the highest value is 90 meq / L at 10 minutes from the times of magnetization which is located at 5 minutes from the times of filtrate, and notes here that the degree of magnetization, and the magnetic degree (10) clearly effect on the readings and all stages of the infiltration. in other case fig (10) noting that there is a convergence of the slopes for the curves down to the time of filtrate 60 minutes except for the curve on the magnetize period 10

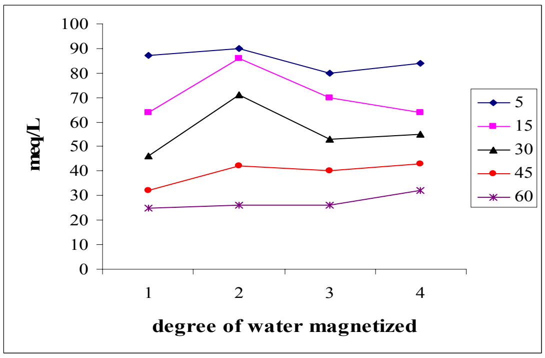


Fig.(9) Effect of magnetize water degrees on Ca values at different periods of the infiltration

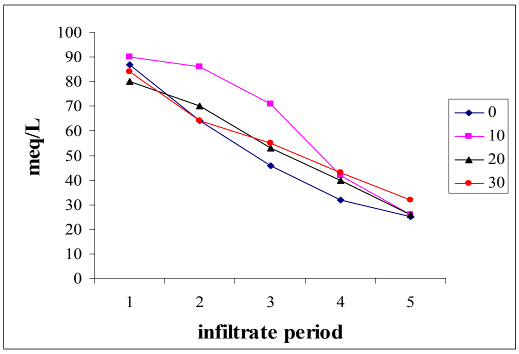


Fig. (10) Effect of filtration periods on K values at different magnetic degrees

6- Concentration of Magnesium ions: the lowest value was 20 meq / L at the water magnetized for 10 minutes and located at the point 60 minutes from the times of filtrate, while the highest value was 58 meq / L at the water magnetized for 20 minutes and is located at 5 minutes of times of filtrate. It was noted that there is convergence in the concentrations of magnesium at magnetization degree 30 for all periods of filtrate fig.(11). When the opposite relationship as the second case and make the times of filtrate on the X-axis will the curves generaly will go down with increase in the infiltration period with relatively high in curves values on magnetic water at 30 minutes Fig.(12).

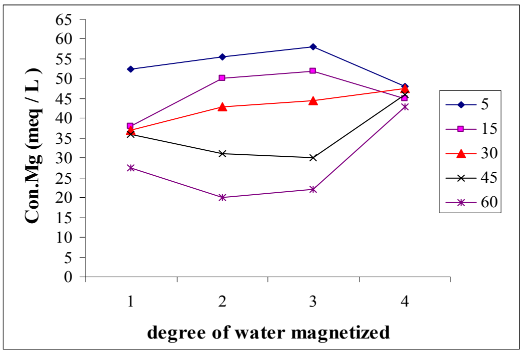


Fig.(11) Effect of magnetize water degrees on Mg values at different periods of the infiltration

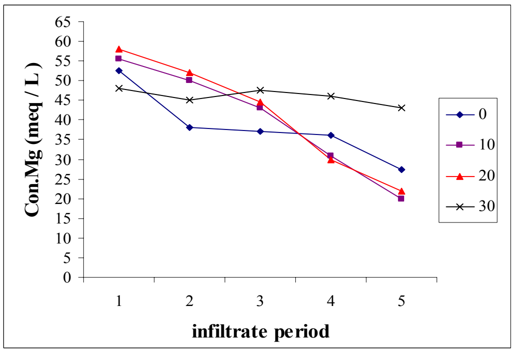


Fig. (12) Effect of filtration periods on Mg values at different magnetic degrees

Acknowledgements
The technology of magnetic change the features of the physical and chemical water and natural, which leads to improved filtering and energy to dissolve, so the process of separating salts from saline soils get much better and plants can absorb nutrients and fertilizers in the best during the period of germination, in addition to the water treated magnetically the wash salts from the soil to effectively increase the effectiveness of three times the water is magnetized in it, Should also note that after washing the magnetic soil, the content of nutrients in the obtained filtrate decreases with increasing the infiltration period, within the different degrees of magnetic water.

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Chelate-enhanced Phytoextraction and Phytostabilization of Lead Contaminated Soils by Carrot, *Daucus Carrota*

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Abstract
Chemically enhanced phytoremediation has been proven to be effective criteria for removing heavy metals from contaminated soils through use of high biomass plants. The objective of this study was to study the influence of different ethylenediamine tetraacetate (EDTA), nitrilotriacetic acid (NTA) and oxalic acid (HOx) concentrations on lead accumulation capacity of carrot (*Daucus carrota*). The results indicated that EDTA was more effective than NTA and HOx in solublizing soil Pb. The highest Pb content in shoots (342.2±13.9 mg kg^{-1}) and taproots (301±15.5 mg kg^{-1}) was occurred in 10 mM EDTA, while it occurs for capillary roots (1620±24.6 mg kg^{-1}) in 5 mM HOx, when soil Pb concentration was 800 mg kg^{-1}. The high obtained phytoextraction and phytostabilization potentials were 1208 (±25.6) and 11.75 (±0.32) g Pb ha^{-1} yr^{-1} in 10 mmol EDTA kg^{-1} soil and no chelate treatments, respectively. It may be concluded that chelates application increases Pb uptake by carrot. Consequently, this plant can be introduced as a hyperaccumulator to phytoextract and phytostabilize Pb from contaminated soils.
Keywords: Ethylenediamine tetraacetate (EDTA), Nitrilotriacetic acid (NTA), Oxalic acid (HOx), Phytoextraction potentials, Phytostabilization potentials.

Introduction

Lead (Pb) is one of the most widespread contaminants in soil that generally concentrates in topsoils. This heavy metal is a nonessential element in metabolic process and may be toxic to organisms even when absorbed in small amounts. Phytoextraction and phytostabilization are two types of phytoremediation technology that involves removing heavy metals from contaminated soils by hyperaccumulator plants. For phytoextraction to be a feasible remediation tool, plants should be able to both absorb and translocate large amounts of heavy metals into aboveground tissues and have high biomass and profuse root system (Alkorta et al. 2004a). Phytostabilization uses certain species to immobilize contaminants, through absorption and accumulation by roots, adsorption in roots or precipitation within the root zone (Prabha and Li 2007).

High affinity of soil constituents for heavy metals and very low solubility of their minerals decrease efficiency of phytoremediation process. To enhance these limitations, application of chelating agents has been proposed. In the many pot experiments described in the literatures the effect of EDTA has ranged from no significant to over 100-fold enhanced accumulation (Greman et al. 2001). The wide variation has been due to the metal species, the metal concentration in the soil, soil properties and the amount of EDTA applied (Evangelou et al. 2007). Huang et al. (1997) showed that EDTA enhances desorption of Pb from soil and subsequently increases the Pb concentration in soil solution. Although synthetic chelates (EDTA) have shown positive effects on the enhancement of metals phytoextraction from contaminated soils, their application can also be harmful. Because most of synthetic Aminopolycarboxylic acids (APCAs) are non-selective in extracting metals (Barona et al. 2001), have low biodegradability and could decrease plant growth even at low concentrations (Chen and Cutright 2001). Recently, natural APCAs such as NTA have been proposed as an alternative for EDTA. NTA, as a biodegradable chelating agent, forms strong complexes with metals (Quartacci et al. 2005). In spite of its expected positive properties, few studies have been performed with NTA as a ligand to assist phytoextraction of metals. Several experiments using natural low molecular weight organic acids (NLMWOA) have also been performed. Of these compounds, citric acid, oxalic acid and malic acid, because of their complexing properties, play a significant role in metal solubility (Nigam et al. 2001). So far, most studies on Pb in plants have been concerned with its effects on plant growth, accumulation and phytotoxicity (Liu et al. 2008). Chelates can not only desorbs heavy metals from the soil matrix into soil solution, but can enhance the bioavailability of Pb for translocation to the plant. So, the current study was conducted to determine whether application of different concentration of EDTA, NTA and oxalic acid can enhance the solubility of Pb and make it more bioavailable for root uptake. Also we wanted to assess the efficacy of kind and rate of chelates in facilitating Pb sorption by capillary roots and its translocation into taproot and shoot of carrot (*Daucus carrota*).

Materials and Methods

Soil collection and treatments

Soil sample was collected (upper 0-30 cm) from an agricultural field in the west of Tehran, Iran. The sample was sieved through a 5 mm sieve and air-dried for 4 days. The experimental soils were artificially contaminated with 0, 100, 200, 300, 500 and 800 mg Pb kg^{-1} as $Pb(NO_3)_2$. Each treatment of Pb contaminated soil (17 kg) was placed in a lysimeter (25cm diameter×30cm height) with three replications. The experimental soils were fertilize and equilibrated for 9 weeks, undergoing 9 cycles of saturation with non saline water (EC: 0.3 dS m^{-1}) and air drying processes. Some physical and chemical properties of the soil were measured by standard methods (Table 1). The total Pb concentration in the soil was measured using acid digestion (1:1 concentrated HNO_3 and deionized water (v/v), H_2O_2 30% and concentrated HCl) (Gupta, 2000).

Table 1. Some physical and chemical properties of the experimental soils

Clay (%)	Silt (%)	Sand (%)	pH (H_2O)	OC(%)	CEC(cmol kg-1)	EC (dS m^{-1})	Total Pb (mg kg^{-1})
12.2	21.1	66.7	7.65	0.42	14.3	0.81	10.5

Values are the mean of three replicates

Seeds sowing and cultivating condition

Carrot (*D. carrota*) was used due to its high biomass production (~ 20 t ha^{-1}) and profuse root system. In addition, it has exhibited strong transpiration rates. Ten carrot seeds were planted in each lysimeter. After germination, seedlings were thinned to five plants per lysimeter and grown for five months. The experiment was carried out in a greenhouse under controlled conditions. On the 140th day after sowing, EDTA (as Na_2EDTA salt), NTA (as H_3NTA salt) and Oxalic acid were applied as solution to the soil surface at rates of 0 (no chelate), 2.5, 5 and 10 mmol chelate per kg soil. Each treatment was replicated three times and was in a completely randomized block design.

The examination method of Pb in soil and different parts of plants

In order to study the effect of chelates on Pb concentration in the soil solution, 5.0 g of air-dried contaminated soil (<2 mm) were placed in a 125 mL Erlenmeyer flask. Then, 25 mL 0.01 M $CaCl_2$ extracting solution was added. After that, the suspension was shaken for 120 minutes at 120 rpm. At the end of time period, the suspension was centrifuged and filtered through a Whatman No. 42 filter paper. Pb contents in soil solution were analyzed using induce coupled plasma-optical emission spectrometry (ICP-OES).

Ten days after chelate application, plants were harvested by cutting the shoots exactly at the swelling formed in the taproot to shoot junction. Shoots, taproots and capillary roots were then washed thoroughly with 0.01 M HCl and deionized water to remove soil particles. The dry mass of plant tissues sample (shoot, taproot and capillary roots) was gravimetrically determined after heating at 70°C in a drying oven. The plant tissues were individually grounded in a ball mill, digested in a mixture of HNO_3, $HClO_4$ and H_2SO_4 (40:4:1, by volume) and filter through Whatman No. 42 filter paper (Gupta 2000). Pb content in the shoot, taproot and capillary roots was measured using ICP-OES. The absorbed Pb was expressed as mg kg^{-1} of dry weight.

The calculation method and equations of phytoremediation capability

The success of phytoextraction or phytostabilization process not only depends on the metal concentration in plant tissues, but also on sufficient biomass production. Carrot involved additional phytoremediation ability due to the phytostabilization mechanism which considering increases the capability of this plant in the decontamination Pb from soil. Therefore, the phytoremediation potential was calculated based on both equations as follows (Doumet et al. 2008):

$$PP_{[Shoot+Taproot]} = \left[\frac{C_{[Shoot+Taproot]} \times Y_{[Shoot+Taproot]}}{1000} \right] \qquad PP_{[Capillaryr\ oots]} = \left[\frac{C_{[Capillaryr\ oots]} \times Y_{[Capillaryr\ oots]}}{1000} \right]$$

where $PP_{[Shoot+Taproot]}$ is the phytoextraction potential ($kg\ ha^{-1}$), $C_{[Shoot+Taproot]}$ the shoot and taproot metal concentration ($mg\ kg^{-1}\ d.w.$) and $Y_{[Shoot+Taproot]}$ the annual shoot and taproot biomass productivity ($t\ d.w.\ ha^{-1}\ yr^{-1}$), $PP_{[Capillaryroots]}$ the phytostabilization potential ($kg\ ha^{-1}$), $C_{[Capillaryroots]}$ the capillary roots metal concentration ($mg\ kg^{-1}\ d.w.$) and $Y_{[Capillaryroots]}$ the capillary roots biomass productivity ($t\ d.w.\ ha^{-1}\ yr^{-1}$). The $PP_{[Shoot+Taproot]}$ and $PP_{[Capillaryroots]}$, therefore, reflect the amount of Pb extracted and stabilized by harvestable parts (shoots and taproots) and capillary roots of carrot from the soil during one growth season. Statistical analyses were performed using the SAS statistical package (version 8.2). All values reported here are the means of three replications.

Results and Discussion

Effects of chelates application on solubility of Pb

To study the post-harvest effects of chelating agents on soluble Pb in soil, the concentration of water-soluble Pb at the time of harvest was measured. The results indicated that both EDTA and NTA were effective in solubilising Pb in the soil. The highest concentration of soluble Pb fraction was obtained for EDTA and NTA applications, respectively, while for HOx and no chelate, the differences among treatments were not significant in most cases (Figure 1). Application of EDTA, NTA and HOx at 10 mmol kg^{-1} for 10 days and at level 800 mg Pb kg^{-1} of soil produced 671, 342 and 2.5 mg kg^{-1} of soluble Pb in soil, respectively. Thus, EDTA at its highest concentration solublised as much as 82% of total Pb content which is equal to a 463-fold increase in soluble Pb concentration compared to the no chelate treatment). The addition of 10 mmol NTA kg^{-1} of soil resulted in a significantly lower increase (a 236-fold increase) of soluble Pb concentration compared to 10 mmol EDTA kg^{-1}, in the 800 mg Pb kg^{-1} of soil (Figure 1c). The application of 2.5 and 5 mmol EDTA kg^{-1} at the highest soils Pb concentration produced 506 and 632 mg kg^{-1} of soluble Pb in soil, respectively, which were 348 and 436 times higher than the amounts obtained in the no chelate. The addition of 2.5 and 5 mmol NTA kg^{-1} led to 113 and 170 mg kg^{-1} increase in soluble Pb content (Figure 1 a,b).

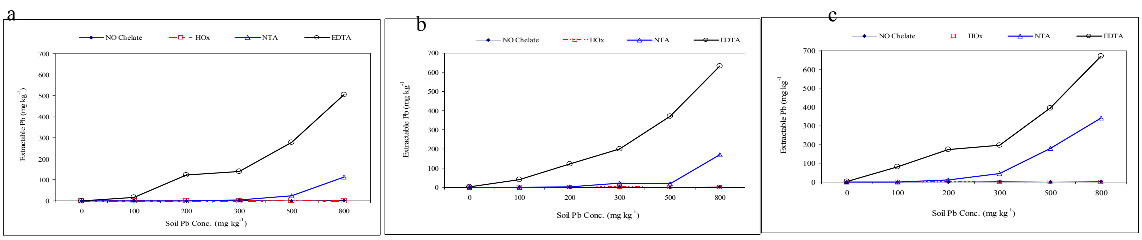

Figure 1. Change in the soluble Pb concentration in soil treated with EDTA, NTA and HOx with concentrations of 2.5 (a), 5 (b) and 10 (c) mM

The application of Pb to the soil has not significantly effect on soluble Pb concentration in the soil if the chelate was not applied, which confirms the generally low availability of Pb in the soil. Results of this study indicated that HOx and NTA were less effective than EDTA for mobilization of soil Pb. This fact can be due to the higher tendency of Pb to form Pb-EDTA complexes (log $K_{Pb\text{-}EDTA}$=18.3) rather than Pb-NTA complexes (log $K_{Pb\text{-}NTA}$=12.27) (Sillen and Martell 1964). Also, the fast rate of HOx and NTA biodegradation may be considered as a major factor determining the differences in soluble Pb concentration. This finding is supported by many researches. Luo et al. (2005) reported that the addition of EDTA at 5 mM concentration for 2 days, significantly increased concentration of soluble Pb, Zn and Cd in soil, which were 496-, 5- and 114-fold higher than these in control treatment. Although NTA is a weaker chelate than EDTA, it is a strong complexing agent compared with HOx. Due to its positive properties, several studies have been performed with NTA as a ligand to assist metals phytoextraction (Ruley et al. 2006).

Influence of chelates application on Pb uptake by plant

The Pb concentration profiles accumulated in the shoot and taproot of carrot are illustrated in Figs. 2a-c and 3a-c. The chelate application significantly enhanced Pb translocation to taproots and shoots. At 800 mg Pb kg^{-1} soil, the application of 2.5, 5 and 10 mmol EDTA kg^{-1} resulted in an elevated Pb concentration in shoot of 217.2±4.5 (Figure 2a), 238.2±6.5 (Figure 2b) and 342.2±13.9 (Figure 2c) mg Pb kg^{-1}, respectively. These, almost correspond to a 3.3-, 3.6- and 5.2-fold increase compared to the no chelate treatment (65.5±5.7 mg Pb kg^{-1}). A similar trend was observed for NTA and HOx in that shoot uptake increase 2.6-, 3.3- and 2.7-fold by NTA and 1.5-, 1.6- and 1.6-fold by HOx, respectively (Figure 2a-c). Shoot tissues extracted more Pb from the soil than taproot tissues in most Pb treatments. For instance, in the soil treated with 800 mg Pb kg^{-1}, when 2.5, 5 and 10 mmol kg^{-1} EDTA; NTA; and HOx were applied, the amount of absorbed Pb in taproot increased 2-, 3.9- and 5.5-fold; 2.2-, 1.7- and 2-fold; and 0.9-, 1.5- and 1.3-fold, respectively, compared to the no chelate treatment (Figure 3a-c). These results clearly indicated that the Pb uptake and accumulation depend principally on soluble soil Pb content. Therefore, it may be suggested that the predominant mechanism for Pb uptake by carrot was the concentration gradient between soil and plant tissues. The lower Pb extraction and uptake efficiency of NTA and HOx compared to EDTA may be attributed to their possible rapid biodegradability. Figure 4a-c shows Pb concentrations profiles in capillary roots versus soil Pb and applied chelates. The amount of Pb in the capillary roots decreased with the amount of chelate applied to the soil. At the highest concentration of applied EDTA, NTA and HOx, Pb content in capillary roots had a minimum values of 1038(±21.5), 764(±18.9) and 1067(±20.2) mg Pb kg^{-1}, respectively (Figure 4c). While maximum amounts of Pb in capillary roots are obtained when 5 mmol kg^{-1} HOx and no chelate were applied (Figure 4b). This might be due to translocation of Pb into taproots and shoots. The form Pb-EDTA complexes may enter the root system at disruptions of the endodermis and Casparian strip (Haynes 1980). Since there is no specific Pb transporter known for selective Pb uptake, the translocation of Pb into plant shoot is most likely in the form of Pb-EDTA complex through a passive uptake mechanism (Tandy et al. 2006a).

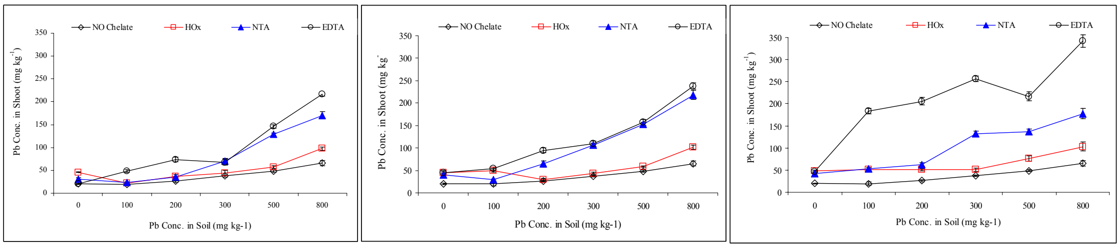

Figure 2. Effects of EDTA, NTA and HOx with concentration of 2.5 (a), 5 (b) and 10 (c) mM on Pb concentration in shoot of *Daucus carrota*. Values are means ± S.E. (*n*=3)

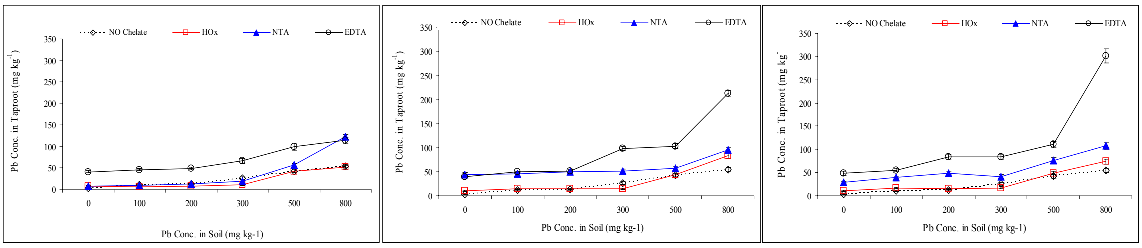

Figure 3. Effects of EDTA, NTA and HOx with concentration of 2.5 (a), 5 (b) and 10 (c) mM on Pb concentration in Taproot of *Daucus carrota*. Values are means ± S.E. (*n*=3)

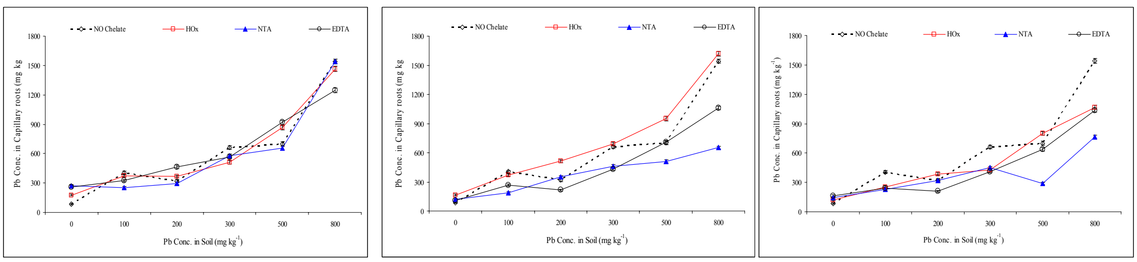

Figure 4. Effects of EDTA, NTA and HOx with concentration of 2.5 (a), 5 (b) and 10 (c) mM on Pb concentration in Capillary roots of *Daucus carrota*. Values are means ± S.E. (*n*=3)

Influence of chelates application on phytoremediation capability

The results indicated that the application of EDTA, NTA and HOx at 5 and 10 mM concentrations produced 832 (±8) and 1208 (±25.6); 598 (±24) and 548 (±26.5); and 363 (±18.2) and 346 (±17.5) *gr* ha^{-1} yr^{-1} Pb extraction from soil, respectively, when soil Pb level was 800 mg kg^{-1} (Fig. 5). A similar trend was also obtained in other Pb concentrations. The higher Pb phytoextraction efficiency by the chelate treatments was mainly due to the better plant growth and bioavailability of Pb in the soil. The highest Pb phytoextraction potential in one growing season was up to 1208 *gr Pb* ha^{-1} yr^{-1} after application of 10 mmol EDTA kg^{-1} of soil (compared to 212(±16.2) *gr Pb* ha^{-1} yr^{-1} extracted by control). This is about ten orders of magnitude greater than values reported by Neugschwandtner et al. (2008) for the phytoextraction potential of *Z. mays* (122.6 *gr Pb* ha^{-1} yr^{-1}, at 6 mmol EDTA kg^{-1}). Also, similar observations have been made by Doumett et al. (2008) with the application of 10 mM EDTA for Pb extraction by *Paulownia t.*, resulting in a phytoextraction potential 620 *gr Pb* ha^{-1} yr^{-1}. The maximum Pb phytostabilization potential (11.75±0.32 *g Pb* ha^{-1} yr^{-1}) occurred in 800 mg Pb kg^{-1} and no chelate treatment (Fig. 6). At the highest soil Pb concentration and in the presence of 5 and 10 mM of EDTA, NTA and HOx, the phytostabilization potential was 7.94 (±0.76) and 5.48 (±0.75); 6.29 (±0.21) and 8.33 (±0.95); 10.8 (±0.65) and 10.4 (±0.61) *gr Pb* ha^{-1} yr^{-1}, respectively (Fig. 6). These amounts are relatively high to successfully remediate the Pb contaminated soils. At presence of DETA and NTA, high amount of Pb translocates from capillary roots to the taproot and shoot. Thus, Pb concentration was decreased in the capillary roots.

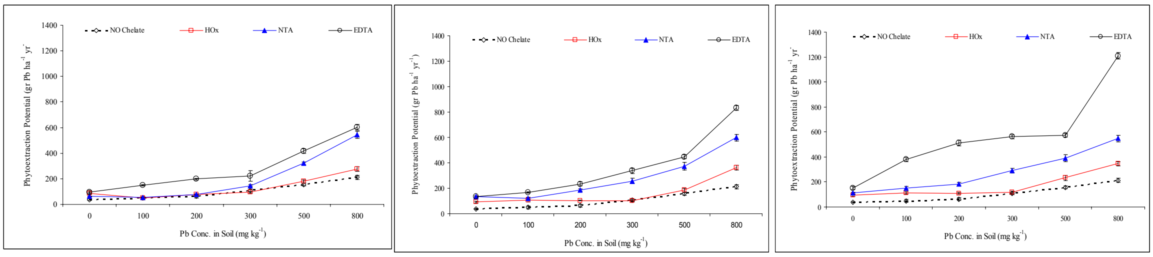

Figure 5. Effects of EDTA, NTA and HOx with concentration of 2.5 (a), 5 (b) and 10 (c) mM on the phytoextraction potential of *Daucus carrota*. Values are means ± S.E. (*n*=3)

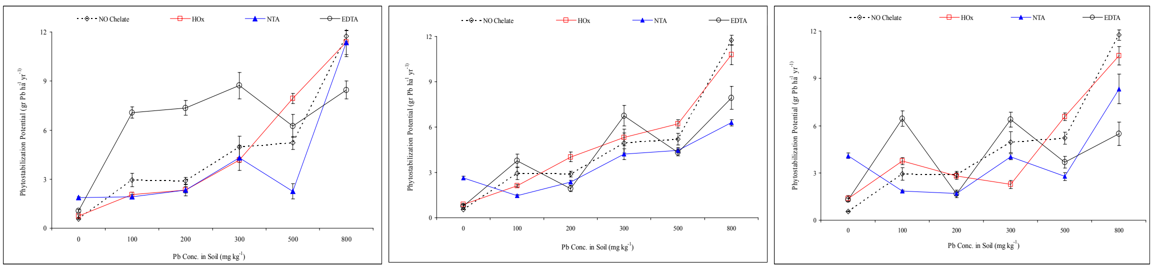

Figure 6. Effects of EDTA, NTA and HOx with concentration of 2.5 (a), 5 (b) and 10 (c) mM on the phytoestabilization potential of *Daucus carrota*. Values are means ± S.E. (*n*=3)

Conclusion

This manuscript can be concluded by the following points:

1. EDTA and NTA significantly increased Pb concentration in the soil solution.
2. The application of EDTA, NTA and HOx significantly increased the Pb concentration in taproots and shoots of carrot.
3. Pb and chelate application have positive effect on carrot growth, even at relatively high Pb concentrations. However, a yield reduction was observed on capillary roots biomass.
4. The chelates application has no effects on stabilization of Pb into capillary roots of carrot.
5. In order to minimize the unwanted and negative impacts of leaching of Pb-EDTA complexes to the groundwater, NTA could be regarded as a good candidate chelate for environmentally safe phytoextraction of Pb contaminated soils.

6. It may also be concluded that carrot can be introduced as a hyperaccumulator to enhanced phytoremediation Pb from contaminated soils.

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Cadmium Fractions as Affected by Long-Term Cultivation and Fertilization History in Some Calcareous Soils of Semi-Arid Region

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Abstract:

The potential hazards arising from cadmium accumulations in cultivated soils through long-term phosphate fertilizer applications along with the loss of P to surface waters have been public concern. Cadmium (Cd) is potentially toxic to the environment, and its bioavailability is related to the chemical fractions it occurs in the soils. The objective of this study was to assess continuous cultivation and fertilizer applications effects on soil Cd fractions in calcareous cultivated and adjacent virgin lands. Twenty one paired surface soil samples from cultivated soils and adjoining virgin lands were studied. The soils were analyzed for physical and chemical properties. Different Cd fractions were determined by sequential extraction procedure. In general, the mean contents of Cd fractions for cultivated soils were as exchangable Cd= 1.3, carbonate bound Cd= 0.81, oxide bound Cd= 0.38, organically bound Cd= 0.33, residual Cd= 0.32 mg kg^{-1} accounting for 41%, 26%, 12%, 11% and 10% of total Cd fractions, and those for adjacent virgin lands were as 1.2, 0.82, 0.37, 0.33, 0.31 mg kg^{-1} accounting for 40%, 27%, 12%, 11% and 10% of total Cd fractions, respectively. Intensive cultivation significantly ($P \leq 0.01$) increased the contents of carbonate bound Cd (an increase of 20%) and organically bound Cd (an increase of 21%) fractions in the Typic Endoaquepts. Long-term use of cultivated soils increased the values of most soil Cd fractions in the studied soil types as compared with virgin adjoining soils, likely due to high application of phosphorus fertilizers. It can be concluded that phosphate applications can increase the risk of Cd movement into the food chain by fertilizer-induced changes in Cd phytoavailability.

Key words: cadmium, fertilization history, fractions, calcareous soil

Introduction

Cadmium (Cd) is potentially toxic to the environment, and their behavior is related to the chemical associations that occur in soils. Thus, the mobility and bioavailability of Cd depend strongly on the physical and chemical properties of the soils. Cadmium can be associated with soil fractions: the exchangeable fraction, considered quickly available; the fraction bound to carbonates, susceptible to changes of pH in soil; the fraction associated to iron and manganese oxides, thermodynamically unstable under anoxic conditions; the organic matter fraction, that can be degraded, leading to a release of soluble trace metals; the residual fraction that is not available by oxy-reduction reactions or solubilization, and presents fewer environmental risks (Tessier et al., 1979).

Continuous application of phosphorus (P) fertilizers for increasing yield may cause a risk of Cd contamination in agricultural lands (Butt et al., 2008). Phosphorus fertilizers derived from sedimentary phosphate rock may contain high concentrations of Cd (Loganathan et al., 2003). The phosphate-fertilized soils are reported to have higher Cd content than in natural uncultivated soils (Butt et al., 2008). Cadmium is an important soil and water pollutant which is generally identified as the most common toxic heavy metal to reach the food chain through crop plant transfer (Chang et al., 2009). Cadmium accumulation in soil and Cd uptake from annually applied P fertilizers at the rate currently being used could be an environmental issue after long-term applications of P fertilizers.

Knowledge of the total cadmium concentration in the soil alone does not help in understanding the mobilization in soils. Thus to appreciate the potential effects of cadmium and its complexity in mobilization, one must understand the various forms of the element present in the soil. This has led to a shift of attention in recent years from the determination of total cadmium in soils to other techniques, which include the quantitative distribution of metal among various chemical phases, such as adsorptive or exchangeable, carbonate, reducible, i.e. Fe-Mn oxide, organic and residual phase (e.g. Ahumada et al., 1999; Howari and Banat, 2001). Sequential extraction or chemical fractionation could be the source of the aforementioned information,

enabling identification and quantitative determination of various forms of the same chemical element (Gworek and Mocek, 2003)
Crops with a high phosphorus demand may affect content and distribution of chemical forms of Cd and its bioavailability. While it has been documented that long-term cultivation affects physical and chemical properties of soil, little is known about its effect on the solid-phase species of soil Cd in calcareous soils of north-west Iran. The objectives of this study were to: 1) determine the distribution of cadmium chemical forms in cultivated soils and adjoining virgin lands, 2) evaluate the relationships among these fractions and with selected soil properties.

Materials and Methods

Soil sampling and analysis

A total of 42 surface soil samples (0–0.30 m) (21 cultivated and 21 virgin) belonging to 14 soil series were collected from the major sugar beet-growing soils and adjacent virgin lands. Typic Calcixerepts (TC), Typic Haploxerepts (TH), Typic Endoaquepts (TE), Vertic Endoaquepts (VE), and Vertic Calcixerepts (VC) are the major soil types occurring in the areas studied. The soil samples were air-dried and ground to pass through a 2-mm sieve before use. The pH was determined using 1:5 soil to 0.01 *M* $CaCl_2$ suspension by a glass electrode and particle size distribution was determined by the sedimentation procedure using the pipette method after dispersing the soil with sodium hexametaphosphate (Gee and Bauder 1986). The total $CaCO_3$ in soil expressed as the calcium carbonate equivalent was determined by a rapid titration method (Rayment and Higginson, 1992). Organic matter was determined by wet digestion (Nelson and Sommers, 1996). Cation exchange capacities of the soils were determined by the 1 *M* NaOAc, pH 7 method. The available Cd was determined by DTPA extraction method as described by Lindsay and Norvell (1978).

Fractionation procedure

The sequential extraction method of Kashem and Singh (2001) modified from Tessier et al. (1979) was used to partition Cd into five operationally defined fractions: (1) exchangeable (Ex-Cd), (2) carbonate-bound (Car-Cd), (3) oxide-bound (OX-Cd), (4) organically-bound (OM-Cd), and (5) residual (Res-Cd). Two grams of each soil sample, 2-mm sieve fraction, was placed in a 50 mL polycarbonate centrifuge tube and following extractions were performed sequentially: Ex-Cd, Sample extracted with 20 mL of 1 *M* NH_4OAc, pH 7 for 2 h at 20 ºC on a rolling table, Car-Cd, residue from the Ex-Cd fraction extracted with 20 mL of 1 *M* NH_4OAc, pH 5 for 2 h at 20 ºC on a rolling table, OX-Cd, residue from Car-Cd fraction extracted with 20 mL of 0.04 *M* hydroxylamine hydrochloride ($NH_2OH.HCl$) in 25 % acetic acid (v/v) at pH 3, reaction time 6 h in a water bath at 80 ºC with occasional shaking, OM-Cd, residue from OX-Cd fraction extracted with 15 mL of 30 % H_2O_2 (adj. pH 2), reaction time 5.5 h in a water bath at 80 ºC with occasional shaking. After cooling, 5 mL of 3.2 *M* NH_4OAc in 20 % (v/v) HNO_3 was added and sample was shaken on a rolling table for 0.5 h at 20 ºC and finally diluted to 20 mL with water, Res-Cd, the residue from OM-Cd fraction, extracted with 7 *M* HNO_3, reaction time was 6 h in a water bath at 80 ºC with occasional shaking.

Paired t-test was performed to compare mean differences in soil Cd fractions between cultivated soils and the adjacent virgin lands. Correlation/regression analysis of data was carried out using the Statview program (Abacus Concepts, 1996).

Results and Discussions

Characteristics of soils

Mean (±S.D.) values of selected soil characteristics and cadmium chemical fractions for five major soil types under sugar beet cultivation and adjoining virgin lands are presented in Table 1. The soils are calcareous and alkaline. Continuous cultivation was associated with changes in soil properties, in particular in soil organic carbon (SOC) content and CEC. A pronounced significant decline ($P \leq 0.01$) in CEC contents (a drop of 6.9%, Table 1) was detected in the TC. This could be attributed to decomposition and decrease of SOC by continuous cultivation. Significant positive relationship between CEC and SOC contents (r = 0.54, $P \leq 0.05$) confirms this statement. Data from long-term cropping system experiments have repeatedly shown that continuous cultivation depletes

SOC and destroys soil quality compared with native vegetation, regardless of cropping system (Schlesinger and Andrews, 2003). Changes in soil physical and chemical properties such as organic matter, pH, CEC or soil structure as a result of intensive agricultural land use has been reported (Lal, 1985; Jaiyeoba, 2003).

Cd contents and forms

The mean contents and changes in five fractions of Cd and DTPA-extractable contents of Cd (plant available Cd) with cultivation are presented in Table 1. The amounts of Cd extracted with DTPA ranged between 0.57-0.79 with an average of 0.68 mg kg^{-1} for cultivated soils and between 0.59-0.75 with an average of 0.66 mg kg^{-1} for adjacent virgin lands. These concentrations are lower than the value of 3 mg/kg reported as a toxic level for soils (Pais and Benton Jones, 1997). The available Cd contents increased in TC, TH, and VC soil types as a result of cultivation, but this increase was only significant ($P \leq 0.05$) in TC.

The concentration of Ex-Cd ranged from 0.40 to 2.5 mg kg^{-1}, with a mean value of 1.3 mg kg^{-1} for the cultivated soils, and from 0.43 to 2.4 mg kg^{-1}, with a mean value of 1.2 mg kg^{-1} for the adjoining virgin land. The largest portion of Cd was found in the Ex-Cd fraction for both the cultivated and the virgin soils and all soil types followed by Car-Cd fraction. Berti and Jacobs (1996) also found the greater percentage of soil Cd in the exchangeable fraction. Long-term cultivation resulted in changes in Ex-Cd contents. The concentration of Ex-Cd increased in TC, TH and VE soil types although this increase was not significant.

Car-Cd contents ranged from 0.063 to 2.0 mg kg^{-1}, with a mean value of 0.81 mg kg^{-1} for the cultivated soils, and from 0.10 to 2.0 mg kg^{-1}, with a mean value of 0.82 mg kg^{-1} for the adjoining virgin land. These values are comparable to those found by Jalali and Khanlari (2008) for Car-Cd (0.7-2 mg/kg) in natural calcareous soils. Several studies have reported that Cd is the most mobile heavy metal in soils and that a large part is associated with the readily leached exchangeable and carbonate bound fractions (Jalali and Khanlari, 2008). The values of soil Car-Cd fraction increased in the TC, TE and TH subgroups. Intensive cultivation significantly ($P \leq 0.01$) increased the contents of Car-Cd (an increase of 20%) in the Typic Endoaquepts.

The OX-Cd, OM-Cd and Res-Cd fractions are of minor importance after the first two Cd fractions (Ex-Cd and Car-Cd). The concentration of OX-Cd ranged from 0.089 to 0.64 mg kg^{-1}, with a mean value of 0.38 mg/kg for the cultivated soils, and from 0.098 to 0.59 mg kg^{-1}, with a mean value of 0.37 mg kg^{-1} for the adjoining virgin land. The values obtained for these soils are comparable to the OX-Cd values reported by Jalali and Khanlari (2008) for oxides bound Cd fraction in natural calcareous soils. OX-Cd contents increased in TC and TE subgroups.

The concentration of OM-Cd ranged from 0.089 to 0.79 mg kg^{-1}, with a mean value of 0.33 mg kg^{-1} for the cultivated soils, and from 0.40 to 0.60 mg kg^{-1}, with a mean value of 0.33 mg/kg for the adjoining virgin land. The small amount of Cd in the organic matter fraction is consistent with evidence that Cd does not appear to form a strong complex with OC (Sposito et al. 1982). OM-Cd contents increased in all soil types except for the Typic Haploxerepts. Intensive cultivation significantly ($P \leq 0.01$) increased the contents OM-Cd fraction in the TE (an increase of 21%) and VE (an increase of 14%) subgroups.

The cadmium present in the residual fraction is mainly associated with silicates and other stable crystalline components and is considered to have been present during formation of the soil minerals. That is, its presence in soils is generally not ascribed to anthropogenic activity and is considered to be biologically unavailable. The concentration of Res-Cd ranged from 0.054 to 0.64 mg kg^{-1}, with a mean value of 0.32 mg kg^{-1} for the cultivated soils, and from 0.11 to 0.61 mg kg^{-1}, with a mean value of 0.31 mg kg^{-1} for the adjoining virgin land. The Res-Cd contents increased in all soil types except for the Typic Calcixerepts and Vertic Calcixerepts. mean relative abundance of the different Cd fractions for both the virgin and the cultivated soils followed the order: Ex-Cd>Car-Cd>OX-Cd>OM-Cd>Res-Cd. The values of all soil Cd fractions increased in the cultivated Typic Calcixerepts and Typic Endoaquepts subgroups. Butt et al. (2008) also reported a definite increase in the overall cadmium levels in the farmed soils compared to the unfarmed soil as a result of intensive cropping and phosphate fertilization.

Table 1: Mean ± standard deviation (S.D.) values of selected soil characteristics (physical and chemical) and cadmium forms for cultivated soils and adjacent virgin lands

variable	Typic Calcixerepts			Typic Endoaquepts		
	Cult.	Virgin	% Change	Cult.	Virgin	% Change
OC[a] (%)	1.1 ± 0.63	1.4 ± 0.63	21	0.76 ± 0.010	0.79 ± 0.22	3.8
CCE[b] (%)	13 ± 7.7	15 ± 6.6	13	12 ± 7.2	9.7 ± 6.5	-24
Clay (%)	33 ± 16	31 ± 15	-6.5	28 ± 6.2	26 ± 1.2	-7.7
CEC[c] (cmol$_c$ kg^{-1})	27 ± 4.0	29 ± 5.3	6.9***	28 ± 4.5	26 ± 6.1	-7.7
pH (0.01 M CaCl$_2$)	7.7 ± 0.16	7.7 ± 0.17	0	7.7 ± 0.49	7.7 ± 0.57	0
Ex-Cd[d] (mg kg-1)	1.4 ± 0.84	1.3 ± 0.79	-7.7	0.69 ± 0.0090	0.71 ± 0.025	2.8
Car-Cd[e] (mg kg-1)	1.1 ± 1.1	0.91 ± 0.66	-21	0.49 ± 0.33	0.41 ± 0.34	-20*
OX-Cd[f] (mg kg-1)	0.42 ± 0.14	0.36 ± 0.13	-17	0.36 ± 0.056	0.36 ± 0.050	-2.8
OM-Cd[g] (mg kg-1)	0.38 ± 0.17	0.36 ± 0.18	-5.6	0.23 ± 0.021	0.19 ± 0.023	-21*
Res-Cd[h] (mg kg-1)	0.29 ± 0.16	0.33 ± 0.12	12	0.49 ± 0.21	0.46 ± 0.22	-6.5
DTPA-Cd[i] (mg kg-^{1})	0.69 ± 0.044	0.67 ± 0.045	-3.0*	0.58 ± 0.025	0.60 ± 0.011	3.3

variable						
	Cult.	Virgin	% Change	Cult.	Virgin	% Change
OC[a] (%)	0.87 ± 0.42	1.1 ± 0.20	20	1.6 ± 0.14	1.2 ± 0.23	-33
CCE[b] (%)	3.7 ± 4.6	6.4 ± 5.3	42	28 ± 0.71	26 ± 1.4	-7.7
Clay (%)	39 ± 7.6	39 ± 10	0	57 ± 2.3	56 ± 1.4	-1.8
CEC[c] (cmol$_c$ kg^{-1})	28 ± 8.1	30 ± 8.5	6.7	37 ± 1.3	33 ± 1.9	-12
pH (0.01 M CaCl$_2$)	7.3 ± 0.24	7.4 ± 30	1.4	7.6 ± 0.035	7.2 ± 0.32	-5.6
Ex-Cd[d] (mg kg-1)	1.2 ± 0.70	1.2 ± 0.76	-8.3	0.45 ± 0.069	0.46 ± 0.051	2.2
Car-Cd[e] (mg kg-1)	0.76 ± 0.72	0.72 ± 0.67	-5.6	0.56 ± 0.062	0.57 ± 0.037	1.8
OX-Cd[f] (mg kg-1)	0.32 ± 0.16	0.38 ± 0.15	16	0.36 ± 0.084	0.44 ± 0.062	18
OM-Cd[g] (mg kg-1)	0.33 ± 0.24	0.41 ± 0.16	20	0.15 ± 0.067	0.14 ± 0.079	-7.1
Res-Cd[h] (mg kg-1)	0.35 ± 0.15	0.26 ± 0.14	-35	0.23 ± 0.10	0.25 ± 0.066	8.0
DTPA-Cd[i] (mg kg-^{1})	0.69 ± 0.072	0.67 ± 0.057	-3.0	0.66 ± 0.021	0.63 ± 0.0070	-4.8

variable			
	Cult.	Virgin	% Change
OC[a] (%)	1.1 ± 0.10	1.8 ± 0.30	39
CCE[b] (%)	6.3 ± 3.2	8.5 ± 4.6	35
Clay (%)	46 ± 5.3	49 ± 8.8	6.1
CEC[c] (cmol$_c$ kg^{-1})	30 ± 1.2	32 ± 0.60	6.3
pH (0.01 M CaCl$_2$)	7.4 ± 0.042	7.4 ± 0.064	0
Ex-Cd[d] (mg kg-1)	1.9 ± 0.93	1.4 ± 1.3	-36
Car-Cd[e] (mg kg-1)	1.1 ± 1.0	1.2 ± 1.1	8.3
OX-Cd[f] (mg kg-1)	0.29 ± 0.11	0.30 ± 0.17	3.3
OM-Cd[g] (mg kg-1)	0.33 ± 0.34	0.29 ± 0.34	-14*
Res-Cd[h] (mg kg-1)	0.29 ± 0.0090	0.20 ± 0.081	-45
DTPA-Cd[i] (mg kg-^{1})	0.67 ± 0.0040	0.68 ± 0.0070	1.5

a, Organic carbon; b, calcium carbonate equivalent; c, Cation Exchange Capacity; d, exchangeable Cd; e, carbonate-bound Cd; f, oxide-bound Cd; g, organically-bound Cd; h, residual Cd; i, DTPA extractable Cd.
* $P \leq 0.05$; ** $P \leq 0.01$; according to paired t-test results

The mean contents of cadmium fractions for cultivated soils were as Ex-Cd= 1.3, Car-Cd= 0.81, OX-Cd= 0.38, OM-Cd= 0.33, Res-Cd= 0.32 mg kg^{-1} accounting for 41%, 26%, 12%, 11% and 10% of total fractions, respectively. The mean amounts of cadmium fractions for adjacent virgin lands were as Ex-Cd= 1.2, Car-Cd= 0.82, OX-Cd= 0.37, OM-Cd= 0.33, Res-Cd= 0.31 mg kg^{-1} accounting for 40%, 27%, 12%, 11% and 10% of total fractions, respectively (Fig. 1). The
Highly significant correlation was recorded between exchangeable and carbonate bound cadmium fractions (r = 0.77, $P \leq 0.001$) for cultivated and (r = 0.82, $P \leq 0.001$) virgin soils, indicating a dynamic relationship between these forms (Bolan et al., 2003). Stepwise regression analysis showed that Car-Cd and OM-Cd jointly explained 69% and 78% of the variation in Ex-Cd, with the partial contribution of Car-Cd being 58% and 67% in the cultivated and virgin soils, respectively (Table 2). Most of the variation in Ex-Cd (58%) in cultivated and (67%) in virgin soils was accounted for by Car-Cd.

Table 2. Step wise multiple regression equations relating exchangeable Cd to other Cd fractions in cultivated and virgin soils

Ex-Cd in cultivated soils	R^2
Y= 0.532 + 0.905 Car-Cd	0.58***
Y= 0.175 + 0.775 Car-Cd + 1.387 OM-Cd	0.69***
Ex-Cd in virgin soils	
Y= 0.388 + 0.961 Car-Cd	0.67***
Y= -0.009 + 0.882 Car-Cd + 1.388 OM-Cd	0.78***

*** Significant at 0.001 probability level

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Study the Possibility of CuO and ZnO Nanoparticles for the Removal of Heavy metals from waste waters

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Abstract

This study investigated the adsorption of Cd2+, Cu2+, Ni2+, and Pb2+ from aqueous solutions with novel nanoparticle sorbents (ZnO, and CuO) using a range of experimental approaches, including, pH, competing ions, sorbent masses, contact time, scanning electron microscopy (SEM), transmission electron microscopy (TEM), and x-ray diffraction (XRD). Tests were performed under batch conditions to determine the adsorption rate and uptake at equilibrium from single and multiple component solutions. The maximum uptake values (sum of four metals) in multiple component solutions were 360.6, 114.5 mg g–1, for ZnO, and CuO, respectively. Sorption equilibrium isotherms could be described using the Freundlich model in some cases, whereas others isotherms did not follow this model. Furthermore, a pseudo-second order kinetic model was found to correctly describe the experimental data for all nanoparticles. Scanning electron microscopy, energy dispersive X-ray before and after metal sorption, and soil solution saturation indices showed that the main mechanism of sorption for Cd2+ and Pb2+ was ion exchange, whereas both Cu2+ and Ni2+ sorption were due to ion exchange and precipitation.

Keywords: Nanoparticles; heavy metals; remove; waste water

Heavy metals removal from waste water using TiO_2, and Al_2O_3 nanoparticles

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Abstract
This study investigated the adsorption of Cd2+, Cu2+, Ni2+, and Pb2+ from aqueous solutions using nanoparticle sorbents (TiO2, and Al2O3) with a range of experimental approaches, including, pH, competing ions, sorbent masses, contact time, scanning electron microscopy-energy dispersive X-ray (SEM-EDX), transmission electron microscopy (TEM), and X-ray diffractometry (XRD). Experiments were performed in batch conditions to measure the adsorption rate and uptake at equilibrium with single and multiple component solutions. The maximum uptake values (sum of four metals) with multiple component solutions were 114.6, and 49.4 mg g–1, for Al2O3, and TiO2, respectively. Based on the mean metal removal by the three nanoparticles, In some cases, the sorption equilibrium isotherms were described using the Freundlich model, whereas this model was not followed by other isotherms. A first order kinetic model also correctly described the experimental data with all nanoparticles. Scanning electron microscopy, energy dispersive X-ray both before and after metal sorption, and soil solution saturation indices indicated that the main sorption mechanism for heavy metals was attributable to adsorption. These nanoparticles may potentially be used as efficient sorbents for heavy metal removal from aqueous solutions.

Keywords: heavy metals; nanoparticles; removal; multicomponent solutions

Studies on the Novel Strategies in Soil and Irrigation Water for Remediation of Arsenic

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Abstract
Arsenic toxicity has become a global concern owing to the ever-increasing contamination of water, soil and crops in many regions of the world. It is released and contaminated in agricultural soil by natural weathering, industrial production and mining. Arsenic concentration in plant has tendency to relate to arsenic concentration in soil. Also, arsenic contaminated in water is major factor of human health risk. Thus, to limit the detrimental impact of arsenic compounds, novel and efficient strategies are required. The present paper reviews appropriate, low cost and novel methods for the elimination of arsenic as a significant environmental problem from contaminated soil and irrigation water. In this Study, we will discuss recent advances and their potential applications that many of which are based on the use of new nanotechnology products for arsenic remediation from contaminated soil-water systems.
Keywords: Arsenic, Remediation, Soil Contamination, Irrigation Water, Nanotechnology.

Introduction

Arsenic contamination is increasing day by day in many regions of the world and has become a global problem. Arsenic (As) is a metalloid ubiquitously present in soils, normally at trace quantities (Matschullat, 2000). Worldwide, natural soil concentrations are around 5mg/kg, and this varies depending on the origin of the soil (Mandal & Suzuki, 2002). The arsenic content of agricultural soils (Andersson, 1992; Notter, 1993) and of natural waters (Hallberg, 1991) has increased during recent decades. The increased level of arsenic has mainly resulted from mining, industrial, agricultural applications (pesticides, fertilizers) and geochemical processes. Thus problem is dramatically severe due to food chain contamination and the only solution is to localize the metalloid in non-edible parts of the crops and seedlings (Sahu et al., 2007). Arsenic (As) is widely distributed in the environment, originating either from as in the soil parent material or from discharge of as onto land as a result of human activities. Consequently, people and livestock are being exposed to as via contamination of drinking water and consumption of food grown in As-contaminated soil or irrigated with As-contaminated water (Banejad and Olyaie). Widespread use of arsenicals as pesticides has significantly contributed to the elevation of arsenic concentrations in soils (Adriano, 2001).
Several studies have linked long-term exposure even to small concentrations of arsenic with cancer and cardiovascular, pulmonary, immunological, neurological and endocrine effects. As a consequence, arsenic is considered highly toxic and there is a tremendous demand for developing efficient methods for arsenic removal from soils and waters (Shih 2005).

Chemistry and toxicity of arsenic

Arsenic is the twentieth most abundant element in the earth’s crust, fourteenth in the seawater and the twelveth most abundant element in human body (Mandal and Suziki 2002). Arsenic is a metalloid or inorganic semiconductor. It occurs with valence states of –3, 0, + 3 (arsenite, As[III]), and +5 (arsenate, As[V]). Because the valence states –3 and 0 occur rarely, this discussion of arsenic chemistry focuses on As(III) and As(V). Arsenic forms inorganic and organic compounds. Inorganic compounds of arsenic include hydrides (e.g., arsine), halides, oxides, acids, and sulphides (Smedley and Kinniburgh 2002). The dominant organic forms found in water are methyl and dimethylarsenic compounds (Fig. 1, Hung et al. 2004). Methylated arsenic species, such as monomethylarsonous acid (MMA(III)), monomethylarsonic acid (MMA(V)), dimethylarsonous acid (DMA(III)), dimethylarsonic acid (DMA(V)) can be formed through biomethylation by microorganisms under favourable conditions. Generally, As (V) is more prevalent in surface water while As(III) is more likely to occur in anaerobic ground waters.

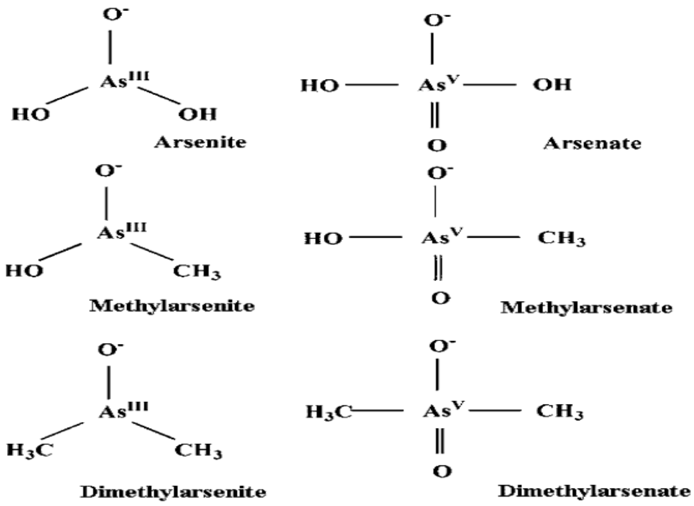

Figure 1. Arsenic species found in water (Hung et al. 2004)

The toxicity and mobility of arsenic are determined by its oxidation state, thus the behaviour of arsenic will change depending on the biotic and abiotic conditions of water. Generally, inorganic forms are more toxic and mobile than organoarsenic species, while arsenite is considered to be more toxic than arsenate. It has been reported that As(III) is 4 to 10 times more soluble in water than As(V) (US EPA, 2002). Moreover, it has been found that As(III) is 10 times more toxic than As(V) and 70 times more toxic then MMA(V) and DMA(V). However, the trivalent methylated arsenic species, i.e., MMA(III) and DMA(III) have been found to be more toxic than inorganic arsenic because they are more efficient at causing DNA breakdown. The toxicity of different arsenic species varies in the order: arsenite > arsenate > mono-methylarsonate (MMA) > dimethylarsinate (DMA) (Jain and Ali 2000).

Conventional arsenic removal technologies

Arsenic retention and mobility in surface water and groundwater are of great concern because of their toxic effects in the environment. Several treatment technologies have been adopted to remove arsenic from drinking water under both laboratory and field conditions. Conventional technologies for removing arsenic from drinking water include:

Oxidation and reduction

Oxidation is a previously required step to transform As(III) species in more easily removable As(V) species. Simple direct aeration is slow, but a number of chemicals, including gaseous chlorine, hypochlorite, ozone, permanganate, hydrogen peroxide, calcium peroxide, manganese oxides and Fenton's reagent (H_2O_2/Fe^{+2}) can be employed to accelerate. Chlorine is a rapid and effective oxidant, but it may react with organic matter, producing toxic and carcinogenic trihalomethanes as by-products. Potassium permanganate effectively oxidizes arsenite, and it may be a widely available inexpensive reagent suitable for developing countries. Hydrogen peroxide can be an effective oxidant if the raw water contains dissolved iron, which often occurs in conjunction with arsenic contamination, allowing the occurrence of Fenton reactions. Ultraviolet radiation alone or with suitable light absorbers such as TiO_2 can be also convenient options for As(III) oxidation.

Precipitation and coagulation methods

Precipitation and coagulation methods for arsenic removal from water depend upon the co-precipitation of both water in soluble arsenates and inorganic oxides of other metals. The water insoluble inorganic oxides are produced by the hydrolysis in the arsenic contaminated water of added coagulants such as alum (aluminum sulfate), ferric chloride. The coagulant must be uniformly mixed into the arsenic-contaminated water in order to obtain the maximum arsenic removal efficiency. If alum is the coagulant, the pH of the contaminated water must be very close to neutral pH whereas ferric salts are useful coagulants over a wider pH range (Ahmed and Rahman, 2000). The usual range of coagulant addition to the contaminated water is between 5 and $50 mgL^{-1}$. Table 1 summarizes the advantages and disadvantages of precipitation/coagulation methods for arsenic removal from water.

Table 1. Precipitation/coagulation for arsenic removal from water

Method	Advantages	Disadvantage
Co-precipitation	No monitoring of a breakthrough is required, with relatively low cost simple chemicals	Serious short and long-term problems with toxic sludge, multiple
Alum coagulation	Durable powder chemicals normally available	chemicals requirement, operation requires training and discipline
Iron coagulation	More efficient than alum on weight basis	Efficient pre-oxidation is compulsory Medium removal of As(III)
Lime softening	Most common chemicals	Re-adjustment of pH is required
Sorption techniques	No daily sludge problem	Requires monitoring of break through or filter use. Requires periodical regeneration or medium shift
Activated alumina	Relatively well known and commercially available	Re-adjustment of pH is required

Adsorption

Aluminum oxides (activated alumina), iron oxide/hydroxides, titanium dioxide, cerium oxide, or reduced metals can be used as adsorbents (Ravenscroft et al., 2009). The technology is very simple, does not require chemical addition and is useful at community or household levels. Granular iron hydroxide, proved to be a good material, able to retain As(V) and As(III) (Driehaus, 2002). Granular iron oxide is another similar successful material, containing less than 70% of Fe2O3. Commercial titanium dioxide, cerium oxide and manganese dioxide proved to be also effective.

Membrane processes

Membrane methods have been applied primarily to purify brackish water or seawater for use as drinking water. Membrane processes can remove arsenic through filtration, electric repulsion, and adsorption of arsenic-bearing compounds. Arsenic compounds could also be directly removed from aqueous solution by separation process such as reverse osmosis (RO), nanofiltration (NF), ultrafiltration (UF) and electrodialysis (ED), with efficiency reaching 100% for RO and NF. One of the major advantages of membrane processes over adsorption is that removal efficiencies are relatively less affected by the chemical composition and pH of the feed (Sato et al. 2002). This is especially true for RO and NF systems. Table 2 shows performances of arsenic removal by various membrane processes and it is based on the work reported by Ning (2002) and Sato et al. (2002).

Table 2. Arsenic removal by membrane processes

Membrane process	Removal efficiencies (%)		
	Total As	As (V)	As (III)
RO	-	96-100	40-85
NF	95-99	60-100	10-75
ED	>95	-	-
UF	-	50-65	10-53

Ion exchange

Synthetic ionic exchange resins, generally of polymeric matrix (polystyrene cross-linked with divinylbenzene), linked to charged functional groups, can be applied for As removal; quaternary amine groups, are the preferred groups. Arsenate removal is efficient, producing effluents with less than 1 mg L^{-1} of arsenic, while arsenite, being uncharged, is not removed, and a previous oxidation step is necessary. Commonly, resins are pretreated with hydrochloric acid, to establish chloride ions at the surface, which are easily displaced by arsenic. Arsenate removal is relatively independent of pH and influent concentration. $HAsO_4{}^{2-}$ has adsorption ability higher than that of H_2AsO^{4-}. Competing anions, especially sulfate, TDS, selenium, fluoride, and nitrate, interfere strongly and can affect run length Suspended solids, SS, and precipitated iron can cause clogging. The most important producers of ion exchanger materials have introduced new tailored anionic exchangers to attain values below 10 mg L^{-1}.

In Table 3 are compared the conventional technologies with their advantages and disadvantages.

Technologies	Advantage	Disadvantages
Oxidation and reduction	Simple. Small installation costs. Easily applied to large water volumes. Arsenite can be directly oxidized by a number of chemicals and/or UV light.	Some oxidants produce toxic and carcinogenic by-products. Needs further removal treatment.
Precipitation and coagulation	Solid obtained can be removed through sedimentation and filtration. Simple. Easily applied to large water volumes. Effective when As(V) is the only pollutant.	Solids rather unstable and inadequate for direct disposal as they will produce As-containing liquid residues. Low removal efficiency. pH needs adjustment. Disposal of the arsenic-contaminated coagulation sludge may be a concern.
Adsorption	Simple. Not other chemicals required. Highly selective towards As(V). Effective with water with high TDS. Useful at community or household levels.	Moderate efficiency. Regeneration needed. Interferences: Se, F^-, Cl^- and $SO4^{2-}$. Application of point-of-use treatment devices needs regeneration and replacement.
Membrane processes	Low space requirement. Capable of removal of other contaminants, if any.	High running costs, high investment costs, high tech operation and maintenance. Toxic wastewater. Re-adjustment of water quality is required
Ion exchange	Effective removal. Not pH and influent concentration dependent.	As (III) is not removed. Sulfate, TDS, Se, F^- and $NO3^-$ interfere. SS and precipitated iron cause clogging. May require pretreatment

New nanotechnology products for arsenic remediation

The application of nanotechnology to the purification and treatment of water supplies to make the potable (Banejad et al, 2011). Various nano-scale materials and nanptechnologies are in development. These technologies may also enable the more efficient and effective removal of arsenic from water and soil. The removal of arsenic by nanoparticles has shown promising results with titanium dioxide nanoparticles. Another promising technology for treating polluted water in situ is the use of zero-valent nanoscale iron particles. Use of zero-valent nano-iron has been shown to significantly reduce pollution for organic solvents, fertilizers, pesticides, and contaminant metals, without the formation of undesirable byproducts. Zero-valent nano-iron is particularly attractive because it can be injected directly into arsenic contaminated water and soil without the need to remove the water prior to treatment.

Metal (hydr)oxide nanomaterials were synthesized using multiple synthesis techniques and the resulting media (titanate nanofibers, nanostructured spheres, and nanoparticle impregnated surfaces) were assessed for their potential to remove an important environmental pollutant in water (arsenate). The hypothesis was that nanotechnology offers the ability to control, characterize, and tailor the fabrication of materials for specific applications. Arsenic was selected as a representative environmental pollutant because of recent regulatory changes to a lower maximum contaminant level and its ability to form strong inner-sphere complexes with metal (hydr)oxides.

Discussion

Arsenic in drinking water is a problem just about anywhere in the world, particularly in developing parts of Asia (Bangladesh, Pakistan and India). Arsenic contamination is largely a natural phenomenon, and no preventive measures can usually be taken, so only remediation technologies can help to minimize the effect. Arsenic can be removed from water in various ways like water purification techniques. Some of technologies are traditional treatment processes coagulation/filtration, lime softening, iron/manganese oxidation, and membrane filtration, which have been used to improve removal of arsenic from drinking water in water treatment plants. Technologies such as ion exchange, manganese greensand filtration and adsorption on activated alumina have been employed in small and domestic systems these are cheap and easily adoptable by local community after small information training. Innovative technologies, such as permeable reactive barriers, biological treatment, phytoremediation (using plants), and electrokinetic treatment, are also used to treat arsenic-contaminated water. However, many of these techniques are at the experimental stage and some have not been demonstrated at full-scale. It is recommended that a combination of low cost chemical treatment like ion exchange, filtration and adsorption along with bioremediation may be useful option for arsenic removal from drinking water.

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Comparison of kinetics desorption of heavy metals (Cd, Zn and Pb) in soils formed from different parent materials in the presence of Leonardite

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Abstract

The aim of this study was the effected of leonardite on kinetics desorption of heavy metals in soils formed from different parent materials including Syenite, Siyenit-Calcite, Schist and Gabbros rocks. Leonardite is a rich source of humic and fulvic acid and act as important cation sorbents, contributing strongly to proton and trace metal cation buffering in soils and aquatic ecosystems. The samples were analyzed for soil physical and chemical properties and mineralogy of clay fraction (XRD-XRF). Soil samples were polluted with heavy metals for two months then treated with 0, 2 and 5% of leonardite. The desorption kinetics of heavy metals were carried out for Cd, Zn and Pb at the concentration of 1000, 4000 and 10000 mg kg^{-1}, respectively, in 2, 24, 48, 168, 360, 720, 1080, 1440 and 2160 hrs during 90 days period. Based on the results of X-ray diffraction analysis, smectits were dominant clay minerals in studied soils. Parabolic diffusion, first order and power function kinetic models found to fit best with the results of desorption kinetic data. According to the results, there were differences in the diffusion coefficient rate (K_d) and values of initial desorption for the studied soil, and Syenite soil had the highest values. Statistical analysis showed that the desorption kinetic parameters such as the amount of released heavy metals, diffusion coefficient (K_d) and initial desorption significantly ($P<0.001$) increased as the leonardite levels increased in the different soil types as follows: Siyenite > Siyenite-Calcite > Schist > Gabbros.

Keywords: Desorption, Kinetics, Clay Minerals, Leonardite, and Heavy Metals

Introduction

Heavy metals reach the soil through natural geologic processes or anthropogenic activities. For example, Pb, Cd and Zn reach the soil by smelting, fertilizers, sewage sludge, and some industries. These are the major sources of metal enrichment in soils and may cause to significant environmental problems due to their toxicity and persistence (Alloway, 1995). Therefore, assessments rate of desorption heavy metals in soils with different clay minerals in control health of environmental is important. Also low rate of desorption of heavy metals in soils contaminated is considered a major obstacle in soils cleaning process. On the other hand, release of heavy metals with different reactions such as complexion, ion exchange, and adsorption/desorption and precipitation reactions dependent on the chemical forms of heavy metals and properties of components soils.

Many kinetic studies have been made by researchers with different adsorbents. For this purpose, we used influence of humic substance for estimate amount and rate of desorption heavy metals with kinetic method. In recent years, some of the character of natural organic materials (lignite, leonardite, etc...) potential as a binding material for the purpose of cleaning heavy metals is used in various fields. Leonardite is a rich source of humic and fulvic acid (up to 90%) and act as important cation sorbents, contributing strongly to proton and trace metal cation buffering in soils and aquatic ecosystems (Tipping, 2002).

Although humic substances can be extracted from peat, composts or soils. The special merits of lignite and weathered coal (leonardite and oxihumolite) have high content of exchangeable functional groups that makes them an effective medium for the removal of metals from wastewater (Machovic et al., 2000; Mizera, et al., 2007). These groups are the active centers of the natural polymer for ion exchange. The leonardite can also be considered as ion-exchange materials (Murakami et al., 2001).

The objectives of present study were 1) The estimated type and amount of clay minerals in soils with derived different parent materials, 2) Application kinetic models, and the selection of the best model for the estimated amount and rate of desoprsiyon heavy metals, 3) The effects of leonardite on amount and rate of desorption heavy metals in different soils, 4) The survey relationship of desorption kinetic parameters (a, b, K_d, and q) and clay minerals.

Materials and Methods

Surface soil samples (0-30 cm) collected from soils each derived from different parent materials including Syenite (1), Syenite-Calcite (2), Schist (3) and Gabro (4) from Akpınar-Kırşehir province located in the central of Anatolia Turkey. X-ray diffraction analysis were performed on the <2 mm clay fraction. X-ray diffractograms were obtained from a Shimadzu XRD-6000 defractometer employing a Cu K_α radiation source from oriented clay. The semi-quantitative mineralogical composition of the clay fraction treated by Mg-saturation, Mg-plus ethylene glycol-saturation, K-saturation, and K saturation alongside heat were determined by X-ray diffraction analysis. For XRF analysis, soils were ground and then sieved to a particle size less than 65 µm finally, were prepared by fluxing of powder samples with $Li_2B_4O_7$.

Soil samples were polluted with heavy metals for two months then treated with 0, 2 and 5% of leonardite. The desorption kinetics of heavy metals were carried out for Cd, Zn and Pb at the concentration of 1000, 4000 and 10000 mg kg^{-1}, respectively, in 2, 24, 48, 168, 360, 720, 1080, 1440 and 2160 hrs during 90 days period. Kinetic equations commonly used in soil desorption studies including first-order, parabolic diffusion, power function and simple Elovich. The equations thus obtained were tested for goodness of fit by least- square regression analysis. All data were analyzed by analysis of variance (ANOVA) for a tow factor factorial experiment. Duncan's new multiple range test (MRT) was used compare to means. The minimum level of significance was set at Ps= 0.05.

3. Results and discussion

3.1. Characteristics of soils

The main physical and chemical characteristics of the studied soils and leonardite are shown in table 1. Among the soils Syenit soil has slightly acidic pH while others soils are very slightly alkaline. Syenite and Gabbros soils indicate moderate organic carbon content (1.7 and 1.2 %). Gabbros soil has the highest clay content (40%) while other two soils have pattern similar form clay (20-22%) content. Schist soil contains more $CaCO_3$ (10.7%) than others soils. Syenite soil had lower CEC than that of others.

3.2 Kinetics desorption of heavy metals

Physical and chemical properties of the soils are depending on type of parent materials. However, desorption rate and the variability of heavy metals depended on the type of clay minerals, the amount of leonardite, time, and Fe_2O_3, Al_2O_3, MnO, CaO, and MgO concentrations. On the other hand, chemical properties of heavy metals are important in complex formation, resulting to variability in trend of adsorption and desorption of heavy metals.

Based on the results of X-ray diffraction analysis, smectits were dominant clay minerals in studied soils and schist soil contained considerable amount of vermiculite mineral (41.3%). Moreover, smectite minerals accounted for more than 86, 55.3, 48.4 and 40.6% of the clay fraction for gabbros, syenite, schist, and syenite-Calcite soils, respectively. The results showed that Fe_2O_3 (7.54%) and Al_2O_3 (15.1%) were dominant minerals gabbros and syenite soils, respectively (Table 1). Generally, chlorite and illite minerals are digenetic or legacy of formed soils on the low-grade metamorphic rocks. The result is in line with Kadioglu et al (2006) who showed that refrigerator mountain contained high potassium in syenite rocks.

Table 1, Physical and chemical properties of the selected soils and leonardite

Parameters	Soils				Leonardite
	Syenite	Syenite-Calcite	Schist	Gabbros	
pH (0.01M $CaCl_2$ 1:5)	6.37	7.11	7.35	7.25	3.51
EC (dS m^{-1})	0.24	0.29	0.21	0.34	-
$CaCO_3$ (%)	2.0	1.0	10.7	4.9	-
O.C (%)	1.7	2.12	0.6	1.2	36.9
Clay (%)	20	28	22	40	-
Texture	S.L	CL	S.C.L	C	-
CEC	18	30	24	35	67
HA+FA (%)	-	-	-	-	28
Smectite (%)	55.2	40.6	48.4	86	-
Kaolinite (%)	20.2	17.4	7.1	12.5	-
Chlorite (%)	0.0	34.8	0.0	0.0	
Vermiculite (%)	0.0	0.0	41.3	0.0	-
Illite (%)	24.6	7.3	3.2	1.5	-
Al_2O_3 (%)	15.11	12.55	9.81	12.34	-
Fe_2O_3 (%)	3.22	5.80	5.50	7.54	-
MnO (%)	0.12	0.14	0.12	0.13	-
CaO (%)	2.11	6.33	19.33	8.21	-
MgO (%)	1.01	4.96	3.57	3.46	-

EC: Electrical Conductivity, CEC: Cation Exchange Capacity, HA: Humic acids, OC: Organic Carbon and A_{va}: Available

Minerals with differences in surface properties as an example clay minerals and oxides, sorb different functional moieties of source humic acid, leading to fractionation of the HA (Specht et al., 2000). On the other hand, the manner adsorption of dissolved organic matter (DOM), HA and FA with Mn-Fe-Al oxides and clay minerals is important. Wang and Xing (2005) showed that HA sorbed on both kaolinite and montmorillonite has much higher sorption capacity and higher sorption linearity than those to in the original HA.
The result of variance analysis kinetic desorption of heavy metals are shown that in table 2. Time, leonardite and time × leonardite interactions were effective factors on the amount of desorption of heavy metals ($P<0.001$).

Table 2, The kinetic desorption of variance analysis for heavy metals

Soil		Reference			
		Time	Leonardit	Leonardite-Time	Error
Syenite	Cd	3754.2***	107959***	83.57ns	69.10
	Zn	433111***	1373707***	4672.0***	1381.0
	Pb	256239***	2099137***	45476.4***	1366.8
Syenite-Calcite	Cd	4593.9***	10511***	430.4***	4.25
	Zn	5503***	1646601***	1421.8***	11.97
	Pb	992.8***	12925***	456.8***	4.84
Schist	Cd	5422.8***	5317.3***	341.7***	3.35
	Zn	2489.3***	7996.5***	625.9***	0.93
	Pb	516.95***	173.9***	58.8***	0.07
Gabbros	Cd	3316.8***	6391.9***	450.1***	2.04
	Zn	395.87***	1768.4***	108.9***	1.10
	Pb	99.94***	961.8***	6.20***	1.17

The amount of the initial desorption of heavy metals were differ in soils toward control samples. Therefore, the amount of desorption increased and on the low contact time (2 and 24 h) was highly effective, and decreased with time contact. This result showed that the increase in contact time decreased the metals subsequent ability to desorb from the clay minerals. Desorption of heavy metals according to the comparison of the average value are as follows;

Syenite > Syenite-Calcite > schist > gabbros

A wide range of kinetic models have been applied to describe the metal-ion sorption and desorption from soil and minerals surfaces (Sparks, 1989). Parabolic diffusion, first order and power function kinetic models found to fit best with the results of desorption kinetic data. Figure 1 has showed only kinetic models for Zn desorption. According to the results, there were differences in the diffusion coefficient rate (K_d) and values of initial desorption for the studied soil, and syenite soil had the highest values (Cd_{L0}=485, Zn_{L0}=2433 ve Pb_{L0}=1506 mg kg^{-1}). Because, application of leonardite levels cause to decrease pH value in syenite soil (Cd: 6.1 to 5.30, Zn: 5.9 to 5:07, and Pb: 5.16 to 4.60), however there was not change pH in other soils. Soil solution pH is an important parameter affecting sorption–desorption reactions. Adsorption of FA/HA on to the surface of clay minerals decreased under high pH conditions, favoring the dissolution of HA and desorption of FA (Abate, and Masini., 2005). Also coating the surface clay minerals with HA, cause reduced the amount of negative charge and has been increased desorption of heavy metal.

Statistical analysis showed that the desorption kinetic parameters such as the amount of released heavy metals, diffusion coefficient (K_d) and initial desorption significantly ($P<0.001$) increased as the leonardite levels. Also, initial desorption of heavy metals in syenite soil was highest than the other soils (Cd: 715 to 1025, Zn: 3704 to 4740, and Pb: 2190 to 4006 mg kg^{-1}).

The most important output of this study was to application of some materials containing humic and fulvic acids (leonardite) to absorption or desorbtion of heavy metals from contaminate soils, must be done owing to its characteristics.

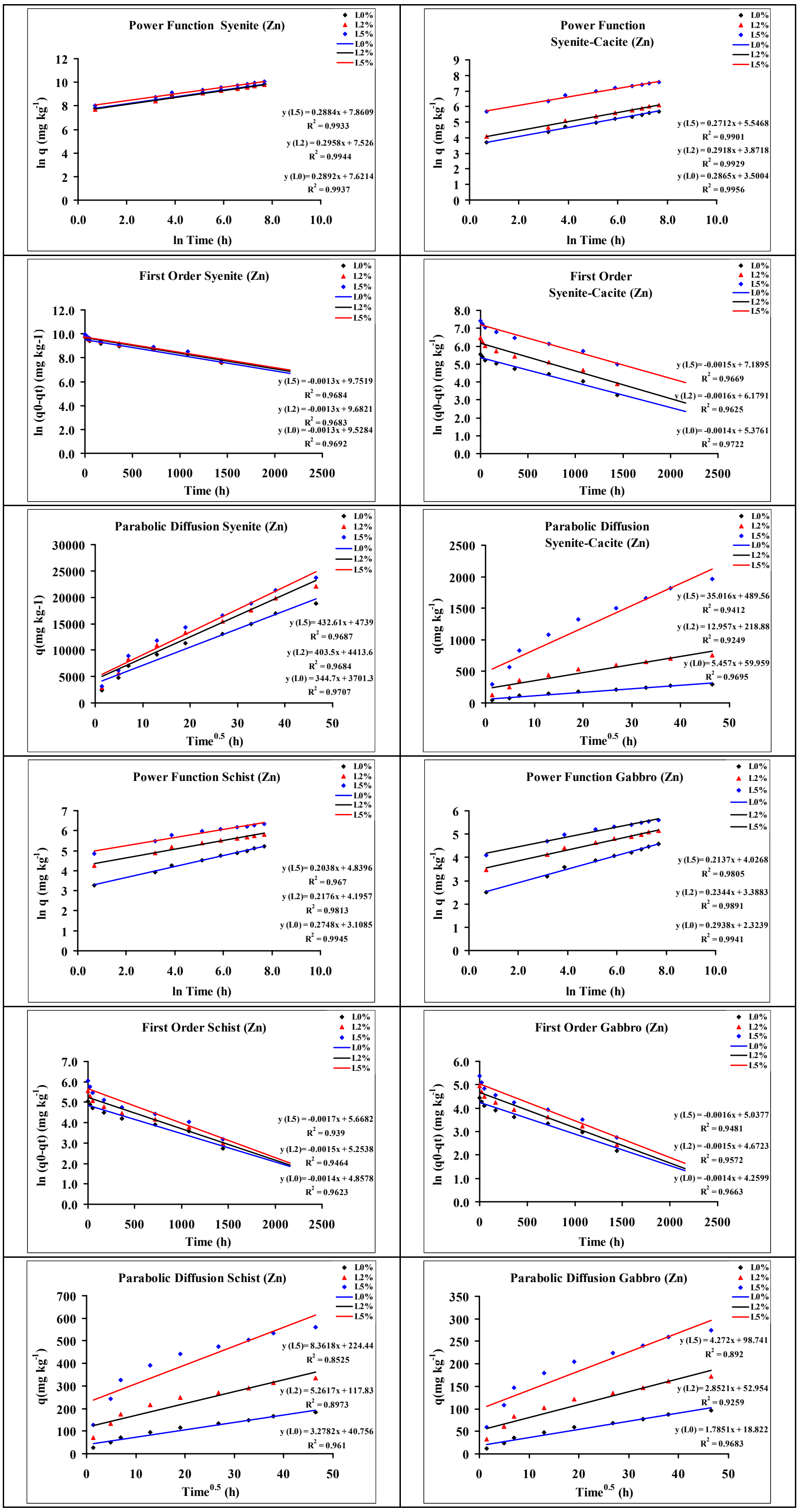

Fig. 1 Kinetic desorption of Zn in studied soils

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Chenopodium botrys L. halophyte weed can phytoremediate Pb-contaminated soils

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Abstract

Phytoremediation is a developing technology for remediate contaminated soils with heavy metal such as lead. Finding out a suitable plant species for phytoremediation is soil scientists responsibility. This species should be native for contaminated areas to be prevented biodiversity. Chenopodium botrys L. is a native plant for Iran and turkey. Soils around the highways in big cities are contaminated with lead resulting from air pollution. It enters into the soil with rain or irrigation water. The target of this research is to find out if Chenopodium botrys L. is useful for phytoremediation pd-contaminated soils. In this experiment applied pollution to the soil with 0,150,300,600,900,1200 ppm lead ($PbNO_3$) with three replicates.In the 45 days pollution soil were abandoning for chemical equilibrium in the field capacity after planting and harvesting the roots and shoots were collected. The concentration of lead was read with ICP. The results show that there was a non-linear positive relationship between the lead concentrations in the soil and the accumulation of Pb in plant roots and shoots. It was observed that by increasing the lead concentration in soil, its accumulation in plant tissues increased. The major lead accumulation occurred in the roots rather than shoots. The measured lead accumulated in roots was 97 mg/kg while it was 57 mg/kg in harvested shoots on 100 mg/kg soil-Pb. Since radish can be seeded up to 2 or three times a year in the same soil, and because its high biomass, it can be used to remediate topsoil's contaminated with lead from air.

Keywords: Lead, phytoremediation, chenopodium botrys L, contaminated soil

Introduction

Today soil and water contamination with heavy metals is one of the problems, which considered as threaten to produce agricultural crops, life of human and other live organisms (Gerritse et al.,1982). The increasing influx of heavy metals into water bodies from industrial, agricultural, and domestic activities is of global concern because of their well documented negative effects on human and ecosystem (Mataka et al., 2006). Pollution of the environment with heavy metals is a growing concern, and generally occurs in well-developed countries. Soil contamination is an adverse phenomenon. Some of pollutants heavy metals transports through soil by water and enters into ground water or uptakes through plants root systems and enters the human's food chains, which can threat human's health on some ways(Wilson,2000).Lead is the most common of the heavy elements. Several stable isotopes exist in nature, Pb being the most abundant. Lead is toxic to most living organisms at high exposure and there is no demonstrated biological need. Since most of the physiologically active tissues of plants are involved in growth, maintenance, and photosynthesis, it can be expected that lead might interfere with one or more of these processes. Indeed, such interactions have been observed in laboratory experiments at lead concentrations greater than those normally found in the nature, except near smelters or mines (Pais and Jones,2000).Wide variations in soil lead levels have been reported, natural levels of non contaminated lead in soils are usually between 10-150 mg/Kg soil. But according to Safety and Environment roles in different regions of the world, the range of 50-150 mg/Kg was known as an allowance range for soil(alloway et al.,1990). Lead accumulates in the body organs (i.e., brain), which may lead to poisoning or even death. The gastrointestinal tract, kidneys, and central nervous system are also affected by the presence of lead. Contaminated soils and waters pose a major environmental and human health problem, which may be partially solved by the emerging phytoremediation technology. This cost-effective plant-based approach to remediation takes advantage of the remarkable ability of plants to concentrate elements and compounds from the environment and to metabolize various molecules in their tissues. Toxic heavy metals and organic pollutants are the major targets for phytoremediation. In recent years, knowledge of the physiological and molecular mechanisms of phytoremediation began to emerge together with biological and engineering strategies designed to optimize and improve phytoremediation.Phytoremediation is a green technology for the sustainable remediation of surface soils contaminated with heavy metals. It is an innovative, novel & potentially inexpensive technology using metal polluted soils, sledges & sediments(chaney et al.,2000). One of the basic factors in phytoremediation is selection of appropriate plant species, which has high potential in accumulation of heavy metal in their organs, especially aerial organs that considered as end of phytoextraction. In definition a hyper-accumulator plant has ability in heavy metals accumulation in its aerial organs 100 times more than a non hyper-accumulator plant(lasat,2000). In phytoremediation researches, great interest has been developed for the identification of autochthonous plant species, which can accumulate elevated amounts of heavy metals in their tissues, with the aim of employing them for phytoremediation of contaminated soils. These plant species have special mechanisms to cope with higher levels of metals in growth medium.Pb concentration that was reported in various plants by researchers is very various. This variation may be due to environmental and genetic factors. Although concentration of Pb is vary in different parts of plants, but occasionally in some plants usual concentration of Pb was lower than 3 $mgKg^{-1}$ (Henry, 2000). The highest amount if Pb accumulation was shown in *Brassica junce*(shen et al.,2002),that not only ,has high potential on Pb accumulation in its roots, but also can transport them to aerial organs.

Chenopodium botrys L. is known as one of the accumulator plants for heavy metal such as lead. This plants specie has ability of Pb accumulation not only in their roots, but also can transfer it to their shoots, that are the phytoextraction end. The target of this research is to find out if Chenopodium botrys L. is useful for phytoremediation pd-contaminated soils.

Material and methods

Experiments were done in a set of 28 pots (1element × 4 repeats × 7treatments) according to Complete Randomly Design. Soil was passing on the 400 μm (No. 40) sieve. Initial specific gravity of soil was 1.344 g cm^{-3}. Any pots were filled by 7 Kg soil. In this experiment applied pollution to the soil with 0, 150, 300, 600, 900, 1200 ppm lead (PbNO3) with three replicates. In the 45 days pollution soil were abandoning for chemical equilibrium in the field capacity after planting and harvesting the roots and shoots were collected. In this study amount of transpiration was measured by gravity methods, and plants were harvested after the end of shoot and roots growth. Fresh weight of plants in any pots was measured, and then harvested plants were washed by distillation water. Washed plants dried in oven in130 °C, and then grained. Pb concentration in Plants grained substances was obtained in wet oxidation methods by ICP instruments.

All values reported in this work are means of four independent determinations. The mean values are given in tables. All the data has been statistically analyzed by one way analysis of variance (ANOVA) in randomized complete block design to check the variability of data and validity of results. Comparison between control and treatment was done by LSD test (Gomez and Gomez, 1984). All tests were performed with SPSS software.

Results and Discussions

Results from concentration of total Pb, soluble Pb, and Pb in roots and shoots of *Chenopodium botrys L*, by ICP instruments was shown in table1. Data exist in this table shows means of Pb accumulated in shoots and roots of *Chenopodium botrys L.* respectively are in ranges of 2.1-57.8 and 8.1-96.7 mg/Kg.

found that the Pb concentration in shoot and in roots of *Chenopodium botrys* was respectively 5.6-54 mg Kg^{-1} and 22.8-97.7 mg Kg^{-1}. Pb concentrations of roots and stems were respectively1.8-18 and 7.6-32.2 rather than usual Pb concentration of same plant (Asadi and Homaii, 2007).

Table1: Concentration of total Pb, soluble Pb, and Pb in roots and aerial organs of *Chenopodium botrys L.*

Total Pb in soil (µgr/Kg)	Soluble Pb in soil (µgr/Kg)	% Soluble to total Pb in soil	Pb in root (mg/Kg)	Pb in shoot (mg/Kg)	Extracted Pb (g/ha/year)
35.5	0.48	1.35%	8.132	2.115	17.9
139.75	0.56	0.40%	22.807	5.676	27.65
250.25	0.56	0.22%	43.980	11.346	59.62
561.19	0.52	0.09%	53.209	21.552	126.01
798.26	0.64	0.08%	71.938	42.933	258.4
1142.5	0.68	0.06%	82.795	50.413	179.8
1150.25	0.6	0.06%	96.753	57.830	178.64

It was observed that by increasing the lead concentration in soil, its accumulation in plant tissues (roots and shoots)increased. The major lead accumulation occurred in the roots rather than shoots. The measured lead accumulated in roots was 97 mg/kg while it was 57 mg/kg in harvested shoots on 100 mg/kg soil-Pb.

Results shows that the translocation rate of root to shoot in a hyper accumulator plant is 0.2, also, this rate for *Chenopodium botrys L.* is 0.46 that near to 0.5. It is well demonstrated that means of Pb accumulation in *Chenopodium botrys L.* roots nearly is similar to Pb accumulation in its shoots. this indicates,except of other plants, *Chenopodium botrys L.* has high potential in translocation of Pb from roots to its shoots(Fig2).

Data obtained from table (1) shows that *Chenopodium botrys L.* averagely storages about5.6% total of Pb in soil on dry substance of its shoots. According to definition of extraction rate for plants (concentration of Pb in plant biomass to concentration of Pb in soil), *Chenopodium botrys L.* as a hyper-accumulator plant has a high potential in uptake and storage of Pb in its aerial organs and shoots.This ability is very effective in phytoextraction.

From this results well documented that, beside of low accumulation Pb in roots system of *Chenopodium botrys L.* , we can use of this hyper-accumulator plant to phytostablozation thought its distributed and extant roots systems. Storage of contaminated Pb in roots system of *Chenopodium botrys L.* lead to organification of heavy metal of Pb, and keeps it from the leaching cycle and entering in to ground waters and or some of mineral reactions in soil for a long times.

In fig (1) shows that Pb accumulation per ha.year on aerial organs of *Chenopodium botrys L.* in ranges of 13.8-258 gr for different soil contaminated treatmets .Results indicates the increase in Pb concentration in soil, causes the concentration of Pb in shoots of *Chenopodium botrys L.* be increased. This trend justifying that this plant can be a potential candidate for pb phytoremediation from contaminated soil.

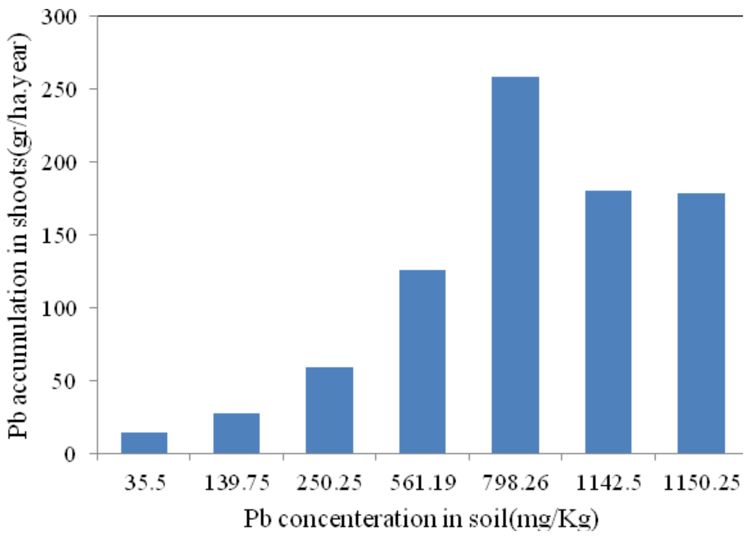

Fig1: Accumulation of Pb in shoots of *Chenopodium botrys L.* respects to Pb concentration of soil.

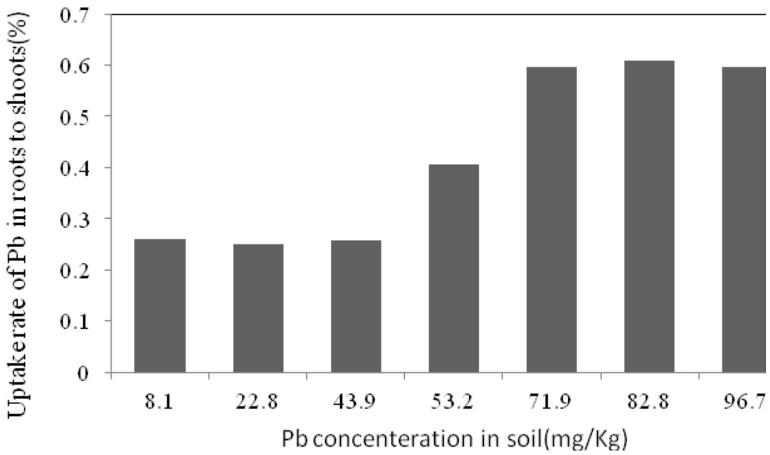

Fig2: Uptake rate of Pb in shoots to shoots of Chenopodium botrys L. respects to Pb concentration of soil.

Acknowledgements:

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Portulaca oleracea L. is a better choice for phytoextract lead-contaminated soils than cadmium

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Abstract
Soil contamination is an undesirable phenomenon in the worldwide. Phytoremediation is the name given to technologies that use plants to clean up contaminated sites. The ability of a plant to adsorb different heavy metals is various. The purpose of this research is to investigate the effect of two heavy metals, Lead and cadmium, on Portulaca oleracea L.Purslane is a native weed on the Middle East. For this research we considered extra concentrations of lead and cadmium in soils to compare the effects of them. The total Cd-concentration in soil was 0-10-20-40-60-100 ppm ($CdCl_2$) and the total Pb-concentration in soils was 0-150-300-600-900-1200ppm ($PbNO_3$). When plants were fully developed, it was harvested and divided into shoot and root parts. The lead and cadmium concentrations in shoots and roots were measured by ICP. In this research the highest removal amount of Pb was 64 gram per hectare at one harvest when the total Pb concentration in soil was 900 mg/kg (Ten times more than threshold lead concentration in soil). The highest removal amount of Cd was 38 gr/hec at one harvest when the Cd concentration in soil was 10 (Ten time more than threshold Cd concentration in soil) and in high concentraction more than 40 ppm of soil-Cd, purslane was not a good choice for phytoextraction because of low biomass and low root growth. So the ability of removing lead from soil is more than cadmium in purslane when the concentrations of these metals are about ten times more than threshold.

Keywords: Phytoextraction,lead,cadmium, Portulaca oleracea L.

Introduction
Soil pollution by heavy metals is a global problem causing vast areas of agricultural land to become non-arable and hazardous for both wildlife and human populations(Garcıa et al.,2003).
Phytoremediation, or the use of plants to remove, destroy or sequester hazardous substances from the environment, has received considerable attention recently because, unlike traditional remediation of these environmental contaminants which usually involves expensive excavation and removal of soil for treatment, it is perceived to be environmentally friendly and relatively low cost(Kinniburg et al.,1976).Iran is an arid to semi aired country and has extant of halophytes plants. This plants specie has ability of heavy metals accumulation not only in their roots, but also can transfer it to their shoots, that are the phytoextraction end. Lead and cadmium toxicity have become an important issue due to their constant increase in the environment. The present study was carried out to investigate the effect of two heavy metals, Lead and cadmium, on *Portulaca oleracea* L. This is C4 plant and a hyper-accumolatur(Mazhari et al.,2011), that has low water needs and wiedly growts in salin soil, so is considered as a halophyte plants(Kumamoto et al .,1990) Purslane plants are a Cd hyper-accumolator, and has high potential to uptake and acccumolation of Cd in their stems and aerial organs(Mazhari et al.,2010).also have ability to translocation of some of heavey metals such as Pb from roots to their stems(Mazhari et al.,2011). In ccontrast with plants sensive to saline soil, Salin tolearability of halophytes increases by increasing in salt concenteration and osmosic effects. In the other hand, halophytes has this ability to uptake water from sline soil(Blaylock et al.,1994). According to this assumption , for Purslane is halophyte which has high tolerance in arid and saline soil, it could has high tolerability and accumulation forheavy metales,Pb and Cd. in deffination a hyper-accumulator plant has ability in

heavy metals accumulation in it's aerial organs 100 times more than a non hyper-accumulator plants(Lasat, M., 2000). lead and cadmium for their long time toxic effects, always must be keep in allowance limit. Soil contamination by thise heavy metals is a serious environmental problem that limits plant productivity and threatens human health. Cadmium (Cd) is one of the most highly toxic environmental pollutants in the atmosphere, soil and water, and in excessive amounts can cause serious problems to all organisms (Benavides et al.,2005). Toxic levels of Cd may be caused by contaminated soil characteristics with abundant Cd or by agricultural manufacturing, mining and other waste disposal practices, or by use of metal-containing pesticides and fertilizers in agricultural soils (Radotic et al.,2005). according to Environmental Conservation Institue Pb is common heavy metals twhich contaminating environments. it treateds human health and cause seriuse disease . Some problems stem from low dislocation and high sedementration of lead(Garbisu et al .,2001, Reeres et al.,1999) .Plants of Brasicasse and Fabasse has famoused as hyper-accumolator of heavy metals(Chaney, R.L. Brown, S.L., 2000). the highest amount if Pb accumulation was shown in *Brassica junce*(Baker, A.J.M. Brooks, R.R. 1989, Shen ZG et al.,2002),that not only ,has high potential on Pb accumolation in its roots, but also can translocate them to aerieal organs ,whiche considered as the most importants end of phytoextraction.In the recent years as effective option for hyper accumulator plants ,using of some plants such as zeamyes,sunflower which have considerable biomass,beside some activities in order to inreasing in heavy metal uptake by these plants (Blaylock et al.,1997, Deram et al .,2000) Succesfullness in phytoremediation is based on producing high portion of biomas and high accumulation of heavy metals in plants organs. Altuough, some of hyper- accumulator plants for their low growth and producing little biomas,are n't approperiate in phytoremediation. The aim of the present research was to investigate the effects of two heavy metals, Lead and cadmium, on dry weight and productivity of Purslane L. also the potential of this plants on accumulations of heavy metals were investigsted.

Material and methods

Experiments were done in a set of 56 pots (2element × 4 repeats × 7treatments) according to Complete Randomly Design. Soil was passing on the 400 µm (No. 40) sieve. Initial specific gravity of soil was 1.344 g cm^{-3}. Any pots were filled by 7 Kg soil. For this research we considered extra concentrations of lead and cadmium in soils to compare the effects of them. The total Cd-concentration in soil was 0-10-20-40-60-100 ppm ($CdCl_2$) and the total Pb-concentration in soils was 0-150-300-600-900-1200ppm ($PbNO_3$). In this study amount of transpiration was measured by gravity methods. When plants were fully developed, it was harvested and divided into shoot and root parts. Fresh weight of plants in any pots was measured, and then harvested plants were washed by distillation water. Washed plants dried in oven in130 °C, and then grained. The lead and cadmium concentrations in plants grained substances were obtained in wet oxidation methods by ICP instruments. All values reported in this work are means of four independent determinations. The mean values are given in tables. All the data has been statistically analyzed by one way analysis of variance (ANOVA) in randomized complete block design to check the variability of data and validity of results. Comparison between control and treatment was done by LSD test. All tests were performed with SPSS software.

Results and Discussions

Fig1 shows trend of Cd portion in shoots of *Purslane* respects to Cd concentration of soil.

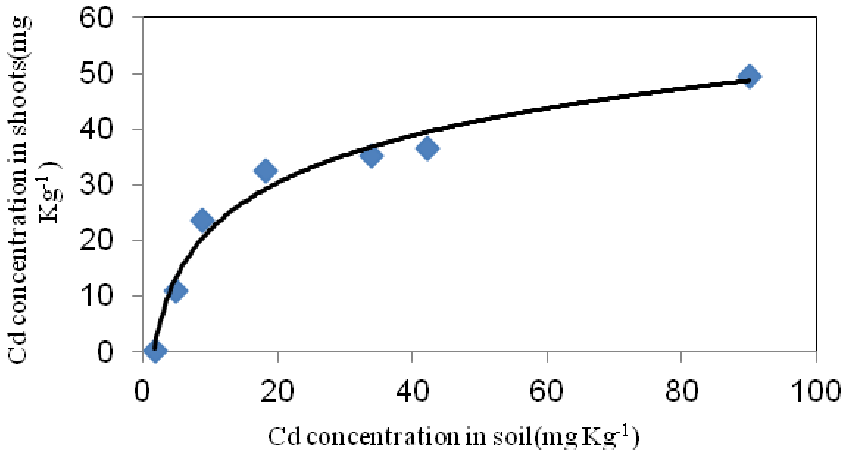

Fig1: Cd concentration in the shoots of *Purslane* versus Cd concenteration in soil

Results demonestrated in figure (1) shows by increasing in the Cd concenteration in soil the Cd concenteration in shoots and aerial organs grows up.Becauses of storage about 95% of total Cd in their shoots, *Purslane* is considered as a hyper accumulator plant, that has ability on high portion of Cd in its shoots and aerial organs. Even thought high accumulation of Cd in shoots and aerial organs, cannot itself ideal reason to apply this plant for phytoextraction, then its productivity must be considered respect to different Cd treatments in soils.Fig(2) shows the productivity of *Purslane* in different Cd treatments in soil.

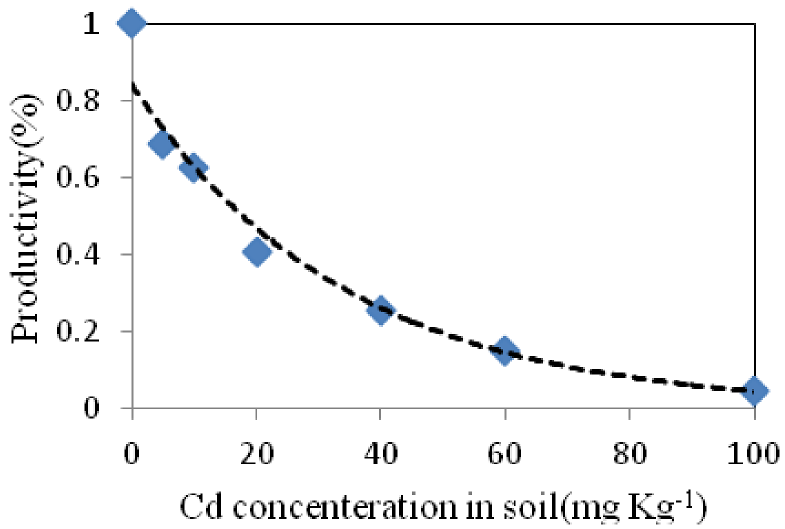

Fig2: Productivity of *Purslane* in different Cd treatments in soil.

In other hand excite of undistrubed an extent rooting system and low developmen in roots due to increasing Cd concenteration in soil well documented that *Purslane* is not a appropriate option for Cd phytorizofiltration in soil. Fig3 demonstrates that dry weight of *Purslane* by increasing in different levels of Cd concentrations in soil is decreasing significantly.

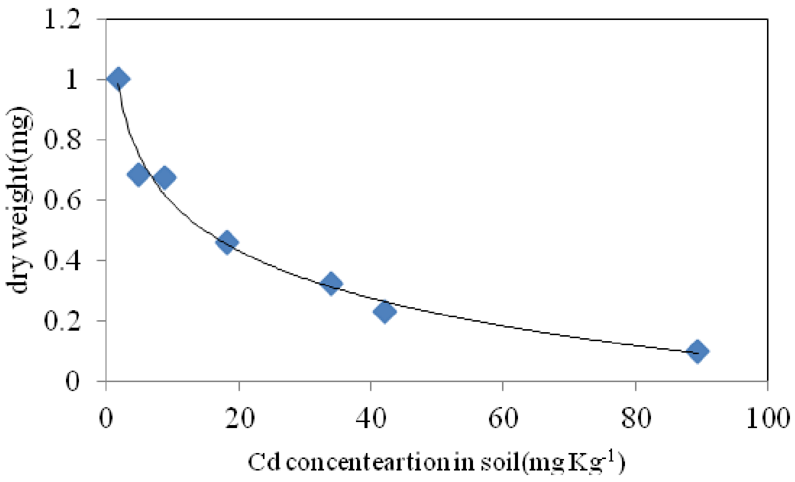

Fig3: Dry weight of *Purslane* in different Cd treatments in soil.

Fig 3shows trend of Pb portion in shoots of Purslane respects to Pb concentration of soil. Results shows, except of 1200mg Kg^{-1}, increasing in Pb concentration of soil causes the increasing amount of Pb in shoots of Portulaca oleracea. This result indicated although Pb was identified as contaminated element for plants, but in this case to some levels of Pb contaminations has increased growths of plants(Fig4). Absorbed Pb in plants to certain concentration (900µg) was acted as a factor to improving plants growth and accounted as a nutrient substance by plants, which applied in cellular interactions.

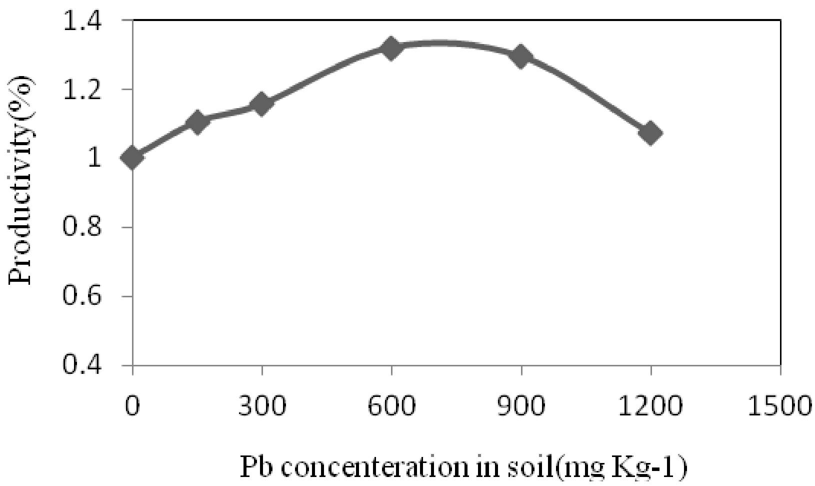

Fig4: Productivity of *Purslane* in different Pb treatments in soil.

Generally, selection suitable plant species, that has high potential to reduce of soil contamination, is very important to phytoextraction. *Purslane* is known as one of the accumulator plants for heavy metal such as lead. This plants specie has ability of Pb accumulation not only in their roots, but also can transfer it to their shoots,because of harvesting and extraction of heavy metals in aerial organs is applicable and do simplicity, these are very important to phytoremendiation. Instead amount of Pb accumulated in roots of plant is higher than its shoots.

Fig5 shows trend of Pb portion in shoots of *Purslane* respects to Pb concentration of soil. This indicated that *Purslane* has accumulated higher amounts of Pb in its aerial organs versus increasing in the leveles of Pb in soil. Amounts of Pb uptakes per ha year was increasing so the leveles of Pb concenteration in soil has increased.

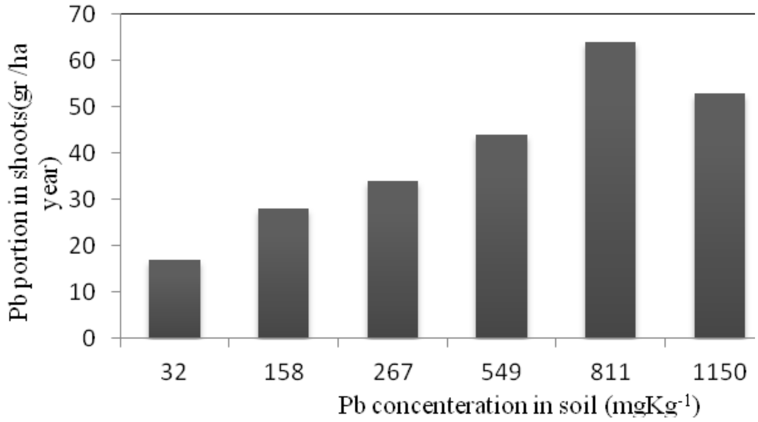

Fig5 : trend of Pb portion in shoots of *Purslane* respects to Pb concentration of soil

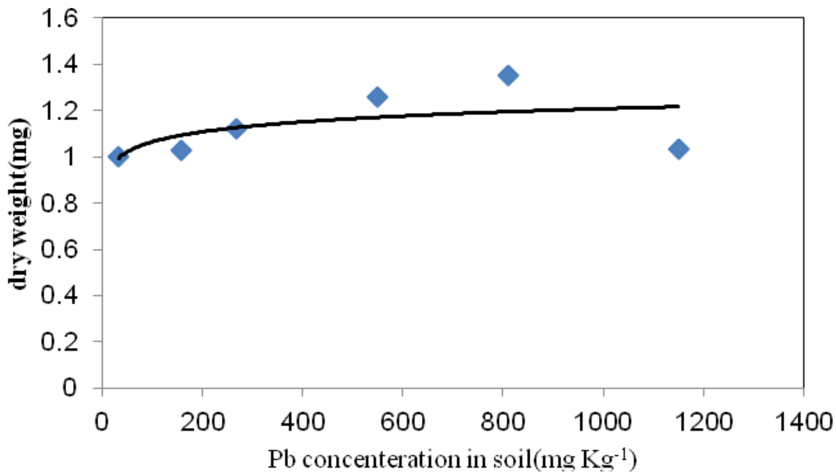

Fig6: Dry weight of *Purslane* in different Pb treatments in soil.

It is also well documented, the ability of removing lead from soil is more than cadmium in purslane .Because of low biomass and low root growth purslane was not a good choice for phytoextraction.

Acknowledgements

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Compare the Cd-phytoextraction in two halophyte weeds:chenopodium album L. and chenobodium botrys L.

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Abstract
Contaminated soils cause serious problem for environment and people health, so it needs an efficient solution. Cadmium is one of the important heavy metals and it makes environment and soils polluted and it may caused by overusing of phosphor fertilizer. Phytoremediation is a new recommendation and remediation technology for controlling and reducing these heavy metals from contaminated Sites. In this method special kind of plants transport heavy metals from soils to their roots and shoots so the rate of these heavy metals reduce in soils. Halophyte weeds *chenopodium botrys* and *chenopodium album* have a high potential for Cd- phytoextraction.the objective of this study is to find the difference between these two weed species in Cd-contaminated soils. Soils were contaminated with 0-5-10-20-40-60-100ppm cd in form of cdcl2. The plants were harvested at maturity growth stage and cadmium concentrations were analyzed by FCP in roots and shoots. The result of this experiment shows that increasing cadmium causes serious decrease in performance of both plants and this decreasing is stronger in chenopodium botrys. The rate of accumulating cadmium in shoots of chenopodium botrys in all treatment of soils is more than chenopodium album. With increasing cadmium pollution the biomass of chenopodium botrys decrease more than chenopodium album. Although chenopodium botrys has more accumulation of cadmium in its shoots but because it has less biomass than chenopodium album this make the output of cadmium in chenopodium botrys decreases in gr/ha by increasing the cadmium pollution in the soil.chenopodum album in high density of pollution has more capability of removing cadmium from the soil.

Keywords: Phytoremediation; Chenopodium album L; Chenopodium botrys L; Contaminated soils

Introduction

The accumulation of heavy metals in soils is one of the serious concerns for environment and people's health. Cadmium is one of the important heavy metals with strong effect on crop quality and human's health, as a first described by Fredrich stromerger in 1817 cadmium intoxication can cause kidney, bone and pulmonary damages. cadmium can inter the environment through industrial wastes, atmospheric deposition, composted materials, landfill runoff ,phosphate fertilizers and municipal sewage sludge (alloway, 1995;Adriano, 1986;meran 1991).in Iran using phosphate fertilizer is one of the major source of cadmium contamination(moteshare zadeh et al,2008). There are many methods in order to purify and cleaning contaminated soils. Soil remediation includes a wide range of costs according to various factors such as soil properties ad soil condition (Lasat, 2000). Because of high costs of methods we need an inexpensive clean up technologies. Phytoremediation is a method in which the plants are used to reduce the concentration of pollutant in the media contained them. These plants degrade or remove metals, pesticides, solvents and other contaminants from pollutant soils. For example the cost of this method is lower than the traditional processes (McGrath et al. 1994). The plant can be easily monitored and re use valuable metals and its less harmful method (Henry, 2000). phytoremediation contaminants are generally removed from the site by harvesting the plants. phytoextraction is one of the phytoremediation subdivisions. Phytoextraction involves the removal of toxins, especially heavy metals and metalloids, by the roots of the plants with subsequent transport to aerial plant organs (Salt et al. 1998; Lombi et al. 2001a). Hyperaccumulator plants can accumulate some elements 100 times more than other plants (lasat, 2000). Thaspi caerulescen , *Arabidopsis halleri* (Robinson et al., 1998; Bert et al., 2000), Hydrilla and tobacco (akiko hokura;ryoko onuma;nobuyki kitajima Tokyo2004) are four famous hyperaccumulator plants that can accumulate cadmium. Iran is one of the countries with largest collection of halophyte weeds in the world, because of this, the climate and soils conditions in Iran we purpose the using of these weeds for phytoextraction. In this experiment we compare the effect of cadmium of two kind of halophyte weeds; *chenopodium album* L. and *chenopodium botrys* L.

Materials and methods

In this experiment the soils was contaminated with 0-5-10-20-40-70-100 ppm cadmium in the form of (cdcl2) with four replications. We have 2 sets of pots and 2 kind of weed *chenopodium album* L. and *chenopodium botrys* L. to compare their phytoextractions. The pots had drainage system for drainage water. the soil was sandy clay loam(sand50% silt 24% clay 26%) in texture.the PH,EC,Organic carbone,cation exchange capacity(ECE),calcium carbonate and initial cd of the soil was 7.58 , 6.71 (mmohs/cm), 0.7, 14, 7.5 ,1.22 mg/kg respectively. And the bulk density of soil was 1.33 gr/cm3. cdcl2 were dissolved in distilled water and then sprayed on soils. Then the soil has been irrigated to reach the field capacity. the crops irrigated by dropped irrigation system. The irrigation water EC was 1.2 mmohs/cm with ph 7.4 and initial cd 0.0013 mg/lit. the seed were cultivated in experimental pots after about 8 week the plant harvested at maturity growth stage and then roots have been separated from experimental soils.the plants taken for analyzing then plant samples were washed with distilled water and dried in oven at 70 C° for 48 hours then it was digested in concentrated HNO3 HClO4-H2SO4(40-4-1) acids. Cadmium concentration were analyzed by ICP in roots and shoots

Results and discussion:

The results of this experiment show that increasing cadmium causes decrease in performance of both plants. Both plants dry matter decreases with increasing cadmium but chenopodium botrys dry matter decrease more than chenopodium album. Figure 1 shows the relationship between dry matter and concentration of cadmium in soil

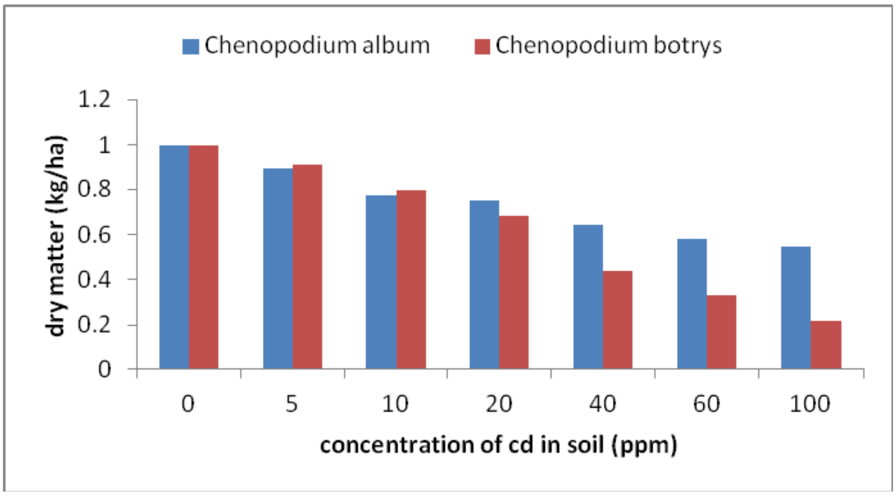

Figure1. The relationship between soil Cd concentration and dry matter

Figure 2 shows the relashionship between the amount of Cd in soil and cadmium on shoots of both plant and it shows that chenopodium botrys has more Cd in its shoots than chenopodium album

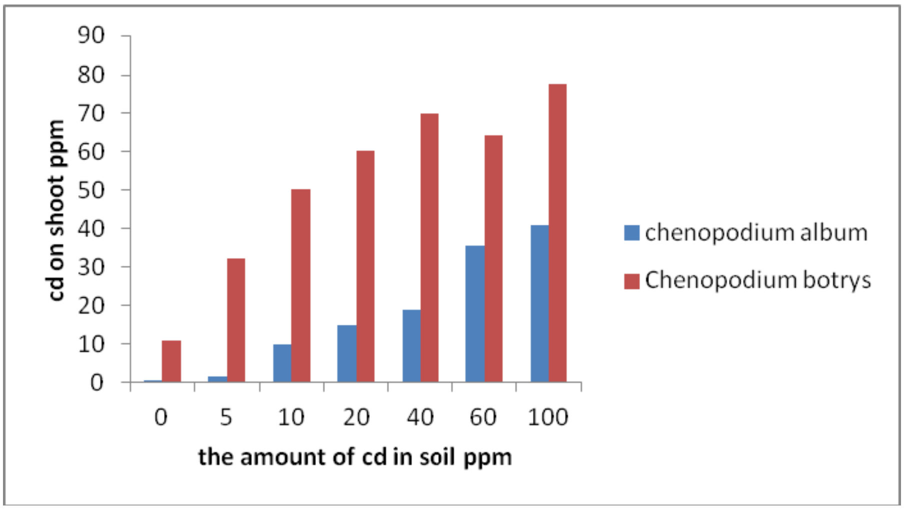

Figure2. The ralationship between the amount of Cd in soil and Cd on shoot

Figure3 shows the relation between Cd removal by shoots and Cd concentrations in soil .it shows that Cd removal by shoots of chenopodium album increases whit increasing Cd up to 100 ppm in soil and we know that plants were used for phytoextraction in two ways in one way plants are hyperaccumulator and in other way despite the contamination in soil plants have enough biomass and high accumulation

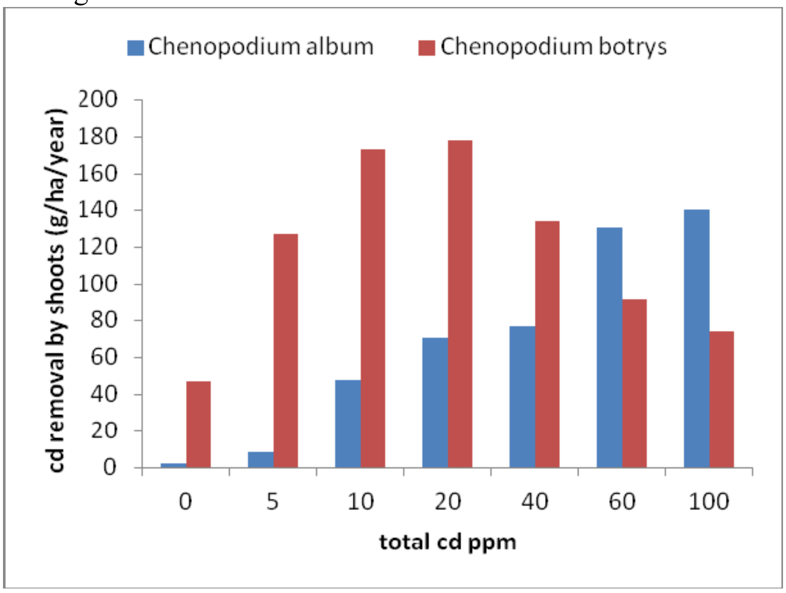

Figure3. The Relationships between total Cd in soil and Cd removal by shoots

In this experiment chenopodium botrys is better plant for phytoextraction of soils with less than 40 ppm Cd concentration. But it suggested using chenopodium album for soils with high contamination because it has enough biomass even in high concentration of Cd in soil

Acknowledgements

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Imperceptible Death of Urmia Lake: Ecological, Biological and Urban Consequences

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Abstract

Urmia Lake (Orumieh) in northwest of Iran (37°N to 38.5°N and 45°E to 46°E), near Turkey border, is the largest salty lake inside Iran and the second great saline lake in the world. Average depth of the lake is 16 m and salt amount vary from 185 to 220g/ltr. It is known as a rare and important ecological, economic zone in the region and one of the Biosphere Reserve of UNESCO. Moreover, it is the second great reserve of Artemia that have high economic values. Unfortunately, water level and area of the lake shows a great decrease in the recent years that leads the lake to the risk of drying and salinity of the soil. This is mainly because of dam constructions and devoting water sources to agricultural farms. The other reason is climate changes. This would be a big disaster to agricultural lands and people who live there as well as disturbing the home of almost 212 species of birds. More than 20000 Flamingo and 200 to 500 White Pelican live here. Unique ecosystem of the lake will be destroyed completely in near future and a salt desert with more than 5000 km2 area will appears overlaid with a 50-60 cm thick salt deposits. Afterwards, it is feared that wind will carry out these salts and chemicals and pesticides deposited in the lake basin to adjacent areas as far as 300 km (up to Zanjan plain) and these will damage agricultural lands, pollute the ecosystem and cause variety of diseases in nearby urban and rural areas. The same process is happening in nearly dried Aral Lake currently as winds erode the Lake basin and deposit ten million tones of dust on near by habitats. Along with them water pollution may lead to outbreak of epidemic and non epidemic diseases. Relying on these evidences it may be predictable that if Urmia Lake dries, the entire region will be affected negatively for several generations.

Keywords: Urmia Lake ecosystem, soil and water pollution, salt winds.

Combating Desertification of Great Loot Desert, Central Iran

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Abstract
Iran, located in the big desert belt of Atlantic Ocean to China, has % 11.2 agriculture lands, % 8.75 forest, % 53.7 rangelands ang %20.8 deserts. The deserts are generally classified in the subtropical, warm, arid and semiarid groups which conforms almost about 340,000 km^2 (less than one fifth of its total area) of which 120,000 km^2 is subjected to moving sands. Loot Desert with the extent of 100,000 km^2 is the most arid area in the Iranian central plateau. Therefore, most of Iran is dominated with arid and semiarid climate and is in high level of desertification hazard. Desertification phenomenon occurs due to brittle and sensitive specifications of its dominate climate and human unconscious contacts with environment. Also, wind as a dominant process in the area causes deflated features. One of the most peculiar geomorphic and geological features of the two Alborz and Zagros mountain ranges is that, there is always a parallel low and auxiliary range, formed mostly of gravel and mud hills. So each change will be noticed as the first influential chain in other desertification processes, mainly climatic situations, destroying jungles, underwater source decrease, inappropriate methods of mining, population growth and inappropriate agriculture methods. Consequently, man has the power to control desertification through proper utilization of land and water through some activities including increase of water use efficiency in agriculture production, extension of sustainable resources use of energy, harnessing of floods through increasing the capacity of reservoir dams, control of excessive water consumption and finally use of new technics of plant breeding to produce the more tollerant generation of desert plants.
Keywords: Desertification, Alborz and Zagros mountains range, Loot Desert

Studying of cadmium transfer from phosphate fertilizers to maize seedlings

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Abstract
This research was conducted to investigate the amounts of cadmium (Cd) in some of phosphate fertilizers, available in Iranian Markets, and their bioavailability to maize seedlings. To this aim, a pot experiment was carried out using Neubauer bioassay method in a completely randomized design with P-source and substrate factors in four replications. P-source factor was included a DAP of Iran, a TSP of Indonesia, a TSP of China, a SSP of Iran, a DAP of Iran, and regent grade KH_2PO_4 as the Control; and the substrate treatments were sand and mixed soil/sand. After 35 days, the seedlings were harvested and then, the roots were accurately extracted from the substrates. In sand substrate, analysis of variance showed significant effects of P-source factor on Cd concentration in shoots and roots, Cd uptake by roots, and total Cd uptake. Also, Cd uptake by roots, total Cd uptake, and Cd concentration in roots in all P-fertilizer treatments were higher than that of the Control. In mixed soil/sand substrate, only concentration and uptake of Cd of/by roots were significantly affected by P-source factor and their amounts were higher than that of the Control in all P-fertilizer treatments except in the case of DAP of Iran treatment; also, the highest amounts of Cd concentration and Cd uptake of/by roots were observed in the SSP of Iran treatment.
Keywords: Cadmium, heavy metals, Neubauer bioassay, phosphate fertilizer.

Nickel Removal by the Aquatic Plant, Coontail (Ceratophyllum Demersum L.)

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Abstract

With increasing of population, water resource polluting process intensified and humans demand for new and inexpensive methods for remediation and improving of water quality. Nickel in low concentration is necessary to ecosystem survival but in high concentrations harmful to life and considered as a dangerous pollutant. This element leads to water resource pollution through contaminated sources such as wastewater (municipal and industrial). Phytoremediation with aquatic plants is a new, effective and inexpensive method for improving water quality and wastewater. In this study, Nickel removal by the aquatic plant, coontail (Ceratophyllum demersum L.), native hydrophyte of most of Iran's rivers, was reviewed after optimum growing pH nomination, within 14 days cultivation in contaminated Hoagland nutrient solution, at the four different concentration of nickel (0, 1, 2, 4, and 6 mg L^{-1}). With daily analysis of nickel concentration in cultivation solution and also initial and final concentration of this element in plant, nickel phytoextraction potential evaluated, and nickel biological effects on coontail grows with biomass production was studied. Maximum removal efficiency was 50% calculated from 6 mg L^{-1} metal concentration. Maximum bioconcentration factor and maximum uptake index calculated from 6 mg L^{-1} metal concentration were 338.65 and 5.05 mg, respectively. Maximum (3.6 g/day) and minimum (1.27 g/day) biomass production index caused from 0 mg $L^{-}1$ and 6 mg L^{-1} of pollutant concentrations. Nickel phytoremediation in hydroponic system with coontail is conceivable and we proffer potential evaluation of coontail in nickel phytoremediation of industrial wastewater.

Keywords: Nickel, Wastewater Phytoremediation, Ceratophyllum demersum L., Phytoextraction, Aquatic Plant

Lead and Cadmium Soil contamination in an urban playground of Iran

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Abstract
There are atmospheric deposition of heavy metals such as lead and cadmium in the soil of playground which are near in the heavily traveled streets. Children are exposed to metals present in soil because of their hand-to –mouth behavior. Children are also more susceptible than adults to any potential negative health effects of metal ingestion. Therefore, in this study lead and cadmium soil contamination was investigated in Khanevade park of Karaj city. Sampling were done from the surface soil (0 – 10cm) of the playground. For determining the total cadmium and lead concentrations, soil samples were digested with a concentrated acid mixture of HNO3–HClO4. Cadmium and lead concentrations of the extract were determined using an Atomic absorption spectrometer. The results showed that the concentrations of Pb and Cd were 39 & 2.75 mg.kg $^{-1}$, respectively. The concentrations of Pb and Cd in the studied urban playground are relatively high, which can be a risk for health of children play in this playground. As there is not any factory or industrial activities near the park, it can be concluded that traffic is the main source for the pollution of the soil of the play ground.
Keyword: soil, Lead, Cadmium, contamination, playground

Introduction

Heavy metals in urban soils may come from human activities, such as industrial and energy production, construction, vehicle exhaust, waste disposal, as well as coal burning. These activities send heavy metals into the air and the metals subsequently are deposited into urban soil as the metal containing dust falls. (Chen et al., 2005).
Lead and cadmium are common pollutants in urban soils. The main sources of Pb and Cd are the leaded fuel used in the past (Fernández Espinosa & Ternero Rodríguez, 2004) and the tyre wear respectively (Chronopoulos et al., 1997; Moller et al., 2005). Lead and cadmium have no essential biological function and are highly toxic to plants, animals and humans. Lead spoil the nervous systems (Lin et al., 1998). The major hazard to human health from cadmium is its chronic accumulation in the kidneys (Chronopoulos et al., 1997).
Most children are exposed to metals present in soil because of their hand-to-mouth behavior, which puts them in more frequent contact with soil than most adults. Children practice both deliberate ingestion of soil and involuntary ingestion through mouthing dirty hands and objects (ATSDR, 2000). Children are also more susceptible than adults to any potential negative health effects of metal ingestion. While their smaller body mass increases the relative exposure to a given quantity of contaminants (per kg body mass) (Schütz et al., 1997). Moreover, because their nervous system is not fully developed, they are more sensitive to neurotoxin metals such as Pb (Klaassen, 1996).
Therefore the current study was desined to investigate the soil contamination to lead and cadmium in a playground (*Khanevade Boustan* of *Karaj* city), located near the streets with heavy traffic.

Material and Methods

Study Area
Selected play ground located in Karaj city. The area has a temperate climate with annual average temperature and rainfall about 16°C and 300 mm, respectively.

Figure1. Study area

Soil sampling

Soil were collected from the playground at the depth of 0-10cm,using a sampling stainless steel augers. As there were two soil textures(clay loam and sandy loam), samples were randomly collected at each location (16 points of clay loam and 5 points of sandy loam). The samples were air dried and ground to enable passage through a 2 mm sieve. Soil analyses were carried out by the following methods: particle size distribution by the hydrometer method (Gee and Bauder 1986); organic matter (OM) content by the Walkley-Black procedure (Nelson and Sommers 1996); and pH values using a glass electrode in mixture of soil and deionized water (1:5, w/v). For determining of the total Pb and Cd concentrations, soil samples were digested with a concentrated acid mixture of HNO_3–$HClO_4$ and heated at 95°C for 2 h. After cooling, the extract was diluted, filtered, and made up to 50 ml with 1% HNO3.The cadmium and lead concentrations of the extract was determined by an Atomic Absorption spectrometer. Also, total Nitrogen percent, available soil phosphorus and available soil potassium were determined (Nelson and Sommer, 1982, Olsen, 1954 and Knudsen et al *.,* 1982).

Result and

Soil properties as well lead and cadmium concentrations are presented in the Table1. The results showed that the concentrations of the cadmium are 2.5 and 2.37 $mg.kg^{-1}$ in the clay loam and sandy loam soils. According to the Table 3 showing the natural range of the lead and cadmium in the soil (Purohit and Agrrawal, 2006), it can be said that the amount of cadmium is slightly higher than natural range. Therefore, it seems that, this amount of cadmium are resulted in traffic especially tyre wears, since the playgrouned was located near the streets with heavy traffic . As the potential risks from soil metal intake depend on the amount of metangested and the bioaccessibility of that metal, the potential risks from soil cadmium intake in this playground should be taken under consideration.

Our results also showed that the concentrations of the lead were 39.5 and 38.5 $mg.kg^{-1}$ in the clay loam and sandy loam soils, respectively (Tabel 2).the comparision between the content of the lead in the studied children playground and table 3cleared that lead concentration was't higher than its natural range. This findings may be due to the fact that the establishment of the playground was before the using of the leaded fuel (Fernández Espinosa and Ternero Rodríguez, 2004).

Table 1. Soil properties of the studied location with clay loam texture.

Properties	
pH	7.57
EC (**ds/m**)	1.71
N (**%**)	0.15
K (**%**)	0.021
P (**ppm**)	4
Pb (**ppm**)	39.5
Cd (**ppm**)	2.37

Table 2.Soil properties of the studied location with sandy loam texture

Properties	
pH	7.47
EC (**ds/m**)	0.195
N (**%**)	0.05
K (**%**)	0.012
P (**ppm**)	4.3
Pb (**ppm**)	38.5
Cd (**ppm**)	2.5

Table 3. Admissible limit heavy metals in the soil (Purohit and Agrrawal, 2006)

Average($mg.kg^{-1}$)	**Natural range in the soil ($mg.kg^{-1}$)**	**Heavy metal**
0.35	0.01-2	Cadmium
19	10-70	Lead

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Lead Accumulation and Its Effect on Zinc Distribution in Different Organs of Maize (*Zea Mays* L.)

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Abstract

In order to investigate the accumulation of lead in shoots and roots of maize and its effect on zinc distribution, a factorial experiment in randomized completely design was performed. Treatments contained four levels of Pb: [0 (control), 100, 200 400 mg/kg] and four cultivars of maize: (SC301, SC302, DC370 and SC500). In this experiment soil was contaminated to different levels of lead. After end of incubation period and vegetation of plants in greenhousecondition concentrations of leadand zincin shoot and rootorganswere measured. The results indicated that the effect of cultivar of, levels of pollution and interaction, there were significant ($p<1\%$) for lead and zincin root and shoot of plant. According to mean compares between cultivars, the SC500 asthehighest concentration of Pb in shoot and the lowest content of Zn in shoot. In compare with control, cultivars of SC301, SC302 and DC370, with increasing in concentration of Pb in shoot, content of Zn in shoot increased but in plant root, decreased. In SC500, by increasing concentration of Pb in shoot, content of Zn in shoot decreased.Unlike the shoot, with increasing concentration of Pb in root, the amount of Zn in root, was decreased. Compare with the control, the Pb concentrations in root and shoot were greatly increased under the high Pb exposed. The highest concentrations of Pb and Zn in shoot of maize were 92.64and 68.2 mg kg^{-1}, respectively and the highest concentrations of Pb and Zn in roots of maize were 205.18and 110.23 mg kg^{-1}, respectively.

Keywords: Soil pollution, Lead, Zinc, Maize

Introduction:

Many parts of the world, especially Industrial and near urban areas, are highly contaminated by heavy metals resulted from human activity (Ravera, 2001). Among all these heavy metals,Lead (Pb) is one of the mostubiquitously distributed toxic elements in the soilas a result of long-term using of phosphatic fertilizers, sewage sludge application, dust from smelters, industrial waste and bad watering practices in agricultural lands. Lead toxicity causesnegative effects on morphology, growth and photosynthetic processes of plants. High level of Pb also causes inhibition of water imbalance, enzyme activities, and alterations in membrane permeability and disturbs mineral nutrition (Sharma and Dubey, 2005).

Zinc has been identified as a component of over 300 enzymes in plants. It is required for the maintenance of membrane integrity, protein synthesis, and DNA and RNA stability (Coleman, 1992) Akinci (2010) reported that Element uptake by roots, shoots and leafs was negatively affected by in raised lead concentrations.

Maize (*Zea mays* L.) is an important cereal crop of Iran and world. It is grown mainly for grain and oil purposein Iran. Its cultivation in Iran About 320,000 hectares of field corn were grown with a production of 2560000 tons an average grain yield of 8000 Kg per hectare in 2011(Nuraky et al, 2011).

In this research, the extent of lead uptake by corn seedlingsand its impact on Zinc Distribution in different organs of Maize (*Zea Mays* L.) were studied. The research was conducted in greenhouse condition at the University of Tehran during March up to April 2011.

Materials and Methods

Soil testing: This study was performed as factorial experiment in randomized completely design with three replications. Treatments contained four levels of Pb: [0 (control), 100, 200 400 mg/kg] and four cultivars of maize: (SC301, SC302, DC370 and SC500). The soil used in greenhouse experiment was collected from depth of 0-30 cm from the research station of Agriculture and Natural Resources of University of Tehran located in Karaj, Iran.Chemical and physical properties of soil sample were measured (table 1) regarding the standard methods.

Pot experiments and treatments: Maize seeds for the present investigation were provided by Iranian seed and plant improvement institute. In this experiment, air-dried soil samples were artificially polluted by Pb (lead (II) chloride). The screened soil was placed in plastic pods, and each pod was filled with 1000 g (DW) of soil.The soils were allowed to stabilize for 45 days in a green house, and then seeds were cultured.The pots were placed in the greenhouse with controlled light and temperature (25°C–30°C).The plants were harvested 45 days after the addition of Pb.

Determination of lead and zinc concentration:In order to determine the amount of lead and zinc in plants,harvested corn shoots and roots were oven dried at 70°C for 48 h. The plants samples were ashed in a muffle furnace at 480-500°C for 5 h. The inorganic ash was dissolved in HCL (1N) solution and this solution was analyzed for Pb and Zn by atomic absorption spectrometry (Schimadzo- AA670) according to Waling et al. (1989).

Statistical analysis:The data recorded were statistically analyzed using SAS-9.1 software. Duncan's multiple range test at 5% probability level was applied for mean separation of significant parameters.

Table 1. Physical and chemical characteristics of soil

Ec dS/m		pH		SP	Soil Texture	Sand %	Silt %	Clay %
46		8		40	Clay Loam	33	30	37
Extracted with DTPA (mg/kg)					K available (mg/kg)	P available (mg/kg)	Total N (%)	Pb (mg/kg)
Mn	Cu		Zn	Fe				
14.80	2.50		1.48	5.54	147.13	15.17	0.095	1.5

Results and Discussion

According to the (Table. 1), effects of cultivar, pollution and cultivar×pollution on lead contents of the shoot and roots were significant (P<1%). Effects of cultivar ×pollution in the roots were not significant (p>0.05). Also effect of cultivar on the zinc contents of roots, was significant (P< 5%).

Table 2. Analysis of variance for Lead and Zinc in shoot and root organs

Source of variation	d.f	Lead (mg/kg)		Zinc (mg/kg)	
		shoot	root	shoot	root
Cultivar	3	1786.24 **	2574.62 **	535.78 **	464.97 *
pollution	3	3555.57 **	61733.54 **	208.70 **	771.76 **
Cultivar × pollution	9	619.18 **	849.77 **	193.94 **	128.79 ^{ns}
Error	32	1.49	5.8	25.76	154.87
C.V	-	5.51	2.91	11.71	16.85

Notes: * and ** (significant at 5% and 1% probability level), ns (no significant)

Mean comparisons (Table. 3) indicated that the maximum lead concentration in shoot and root parts were related to the SC500 and SC302 cultivars respectively, at the 400 ppm level. On the other hand, the highest zinc concentration in shoot and root parts were related to the SC302 and SC301 cultivars, respectively.

Table 3. Results of mean comparisons between treatments

Treatment	Lead (mg/kg)		Zinc (mg/kg)	
Cultivar	shoot	root	shoot	root
500	39^{a}	77.77^{c}	36.36^{c}	66.717^{b}
370	14.99^{c}	90.74^{b}	41.64^{b}	74.36^{ab}
301	11.65^{d}	64.23^{d}	42.89^{b}	81.8^{a}
302	23.08^{b}	97.32^{a}	52.4^{a}	72.47^{ab}
Lead pollution (mg/kg)				
Control	7.93^{d}	13.97^{d}	40.89^{b}	85.63^{a}
100	11.93^{c}	44.58^{c}	39.68^{b}	67.67^{b}
200	22.59^{b}	93.16^{b}	43.67^{b}	71.05^{b}
400	46.27^{a}	178.34^{a}	49.05^{a}	70.92^{b}

Notes: Different letters within the same column indicate significant difference at $p<0.05$ according to Duncan's MultipleRange Test.

Pb concentration in Shoots and roots

Fig. 1 shows the lead concentration (mg/kg) at the different parts of the plants. Our results showed that the accumulation of lead increases with increasing lead levels. Results of statistical analyses showed that the Pb concentration in root is higher than shoots.These result confirmed the researches of Małkowski (2002); Akinci (2010) andQuanlin (2007).

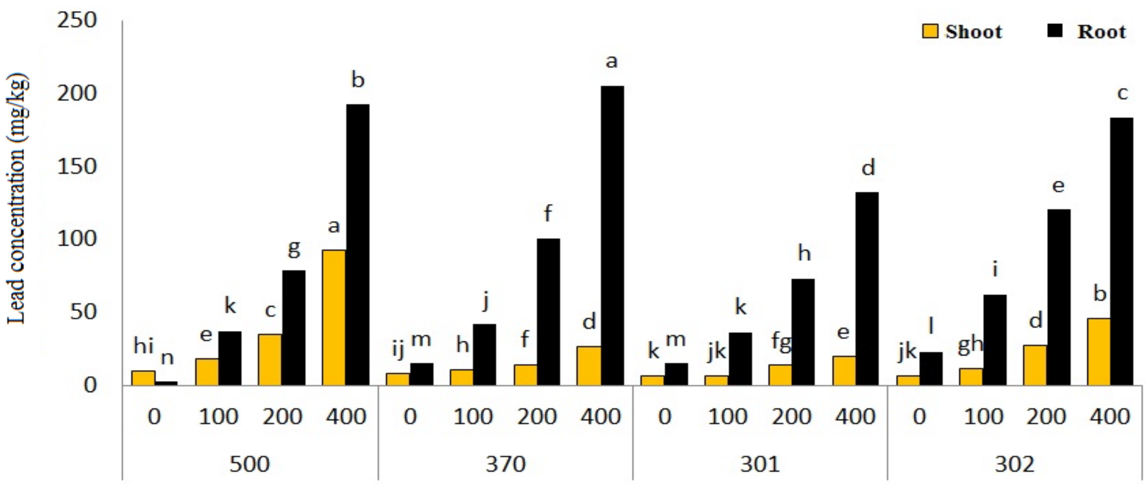

Fig 1. Lead concentration in root and shoot (0, 100, 200 and 400 are lead treatments- 500, 370, 301 and 302 are corn cultivars)

Notes: Different letters within the same column indicate significant difference at $p<0.05$

Interaction of lead and zinc

Fig. 2 shows that the zinc concentration of shoot was increased with increasing lead concentration in the shoots of DC370, SC301 and SC302 cultivars, whereas the zinc concentration of shoot was decreased in SC500 cultivar.

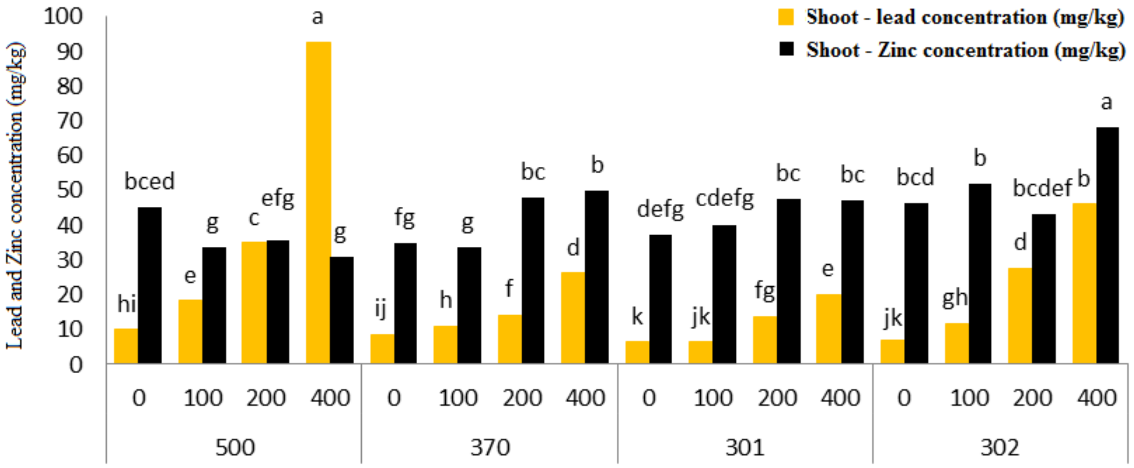

Fig 2. Lead and Zinc contents in shoot (0, 100, 200 and 400 are lead treatments- 500, 370, 301 and 302 are corn cultivars)

Notes: Different letters within the same column indicate significant difference at $p<0.05$

According to the (fig. 3) the zinc concentration was decreased with increasing lead concentration in the roots of all cultivars.

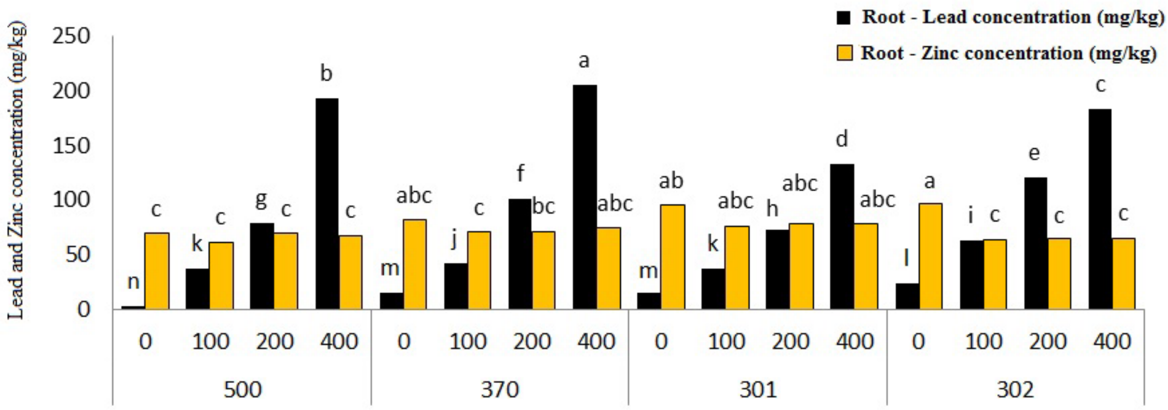

Fig 3.Lead and Zinc concentration in root (0, 100, 200 and 400 are lead treatments- 500,370, 301 and 302 are corn cultivars)

Notes: Different letters within the same column indicate significant difference at $p<0.05$

Results of this study indicated that the accumulation of lead is not uniform among the shoot and root parts of corn seedling So that Pb concentration in root is higher than shoots. The highest and the lowest concentrations of Pb in shoot of maize were 92.64 (SC500) and 6.33 (SC301) respectively, these amounts for Zn concentrations were 68.2 mg kg^{-1}(SC302) and 30.93 mg /kg (DC500). The highest and the lowest concentrations of Pb in root of maize were 205.18(DC370) and 2.43 (SC500) respectively, these amounts for Zn concentrations were 96.97mg kg^{-1}(SC302) and61.43 mg /kg (SC500).

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Germination and seedling growth of purslane (*Portulaca oleracea* L.) under different levels of Copper, Lead and Cadmium

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Abstract

To evaluate the effects of cadmium (Cd), copper lead (Pb) and (Cu) on germination and early growth components of purslane (*Portulaca oleracea* L), an experiment was carried out on based completely randomized design with three replications. Different concentrations of Cd, Cu, Pb contains (0(control), 15, 30, 45 and 60 mg/L) were applied. Germination percentage, seedling length, seedling weight and seedling Vigor index were measured. The results showed a significant effect of treatments on studied traits. With increasing in levels of metals, germination percentage, seedling length, seedling weight and seedling vigor index reduced. The highest germination percentage was related to Pb at 15 mg/L concentrations; however, in other levels of Pb, germination process was reduced. The highest and the lowest seedling length observed in control and Cd (60 mg/L) treatments respectively. It was observed that the higher levels of Cd and Cu had maximum inhibitory effects on seedling vigor index.

Keywords: purslane, heavy metals, germination, seedling growth

Introduction:

Soil pollution by different kinds of heavy metals has become a critical environmental concern due to its potential adverse ecological effects. Although, heavy metals occur naturally at low concentrations in soil systems, they are considered as soil contaminants because of their acute and chronic toxicity. Some of them such as lead and cadmium are special concern due to its relatively high mobility in soils (Das, et al., 1997). Cu^{2+} pollution has become a major environmental problem due to the long term use of copper containing fungicides, industrial and urban activities (e.g. air pollution, city waste and sewage sludge) and the application of pig and poultry slurries high in copper (Marschner, 1995). Ouzounidou et al (1995) reported that When Cu^{2+} is used in excess amount, can be considered as a toxic element, leading to total inhibition of growth, with disturbance of the mitosis, inhibition of root elongation and damage to root epidermal cells and root cell membranes. The toxicity of Cd and Pb to seed germination and seedling growth were reported by Kalimuthu and Siva (1990), Hasnain et al (1995) and Shafiq et al (2008). The aim of this study was to investigate the effect of different heavy metals such as Cd, Pb and Cu on purslane seed germination and seedling growth.

Materials and Methods

In order to study the effects of different levels of heavy metals include: Cd (Cadmium Nitrate), Cu (Copper Sulfate) and Pb (Lead Nitrate) on seed germination and seedling growth of purslane, an experiment was carried out on based on completely randomized design with 3 replications and 13 treatments. Experimental treatments included; Cd, Cu and Pb in four levels (15, 30, 45, 60 ppm) and Control (distilled water). Purslane seeds (50 seeds per Petri dish) after disinifection with Sodium hypochlorite were washed and placed on petri dishes. Planted petri dishes were filled with 8 ml of treatment solution levels. Control groups were filled with only distilled water. Petri dishes were placed inside the incubator with a temperature of 24 ± 2. After 7 days, number of germinated seed, root and shoot length, root and shoot dry weigh were measured. Seedling vigour index 1 and 2 were determined by the following formula (Reddy and Khan, 2001):

$$\text{Seedling Vigour Index 1} = \text{germination percentage} \times \text{seedling length}$$

$$\text{Seedling Vigour Index 2} = \text{germination percentage} \times \text{seedling dry weight}$$

The analysis of variance of the data was done by SAS, 9.1 software. The means compare were separated by Duncan's multiple range test (DMRT) at the 5% probability level.

Results and Discussion

According to analysis of variance (Table 1) final germination percentage and seedling length significantly affected by treatments ($p<0.05$). Also the effect of treatments on seedling dry weight, seedling vigour index 1 and seedling vigour index 2 was significant at 0.01% probability level.

Table1: Analysis of variance for purslane seed germination and early growth characteristics

S.O.V	df	Final Germination Percentage	Seedling Length	Seedling dry weigth	Seedling vigour index1	Seedling vigour index 2
Treatment	12	104.1*	10.48*	1.3**	25196**	0.0048**
Error	26	36.9	0.11	0.35	580	0.001
C.V	-	12.4	14.6	21.15	18.85	21.42

* and **: Significant at the 5% and 1% probability levels, respectively.

The highest germination percentage value was observed in 15 ppm of Pb. Whereas the lowest of this parameter resulted in 45 ppm of Pb (fig. 1). Uruc and Yilmaz (2008) reported that germination percentage values in *Phaseolus vulgaris* and *Rumex scutatus* was strongly inhibited by higher concentrations of Pb. However they were observed that *Rumex scutatus* seed germination was resistant to Cd toxicity at most levels used.

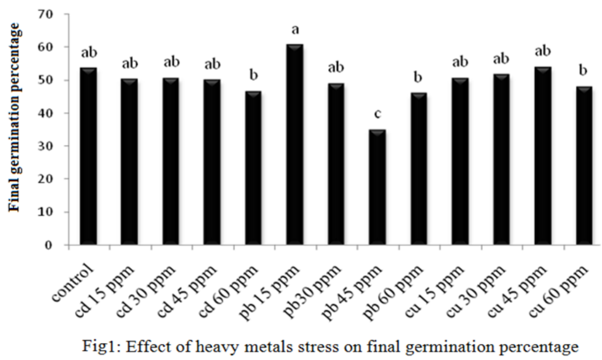

Fig1: Effect of heavy metals stress on final germination percentage

Purslane seedling length constantly decreased with increment of each metal level. According to (fig 2), the highest and the lowest seedling length was related to control treatment and cadmium 60 ppm, respectively. Similary, Oncel et al (2000) and Aydinalp and Marinova (2009) were reported that the reduction of root length in wheat and alfalfa plants at Cd stress condition. Studies showed that Cadmium affect mitotic index in root cells at very high concentrations (Zhang and Yang, 1994).

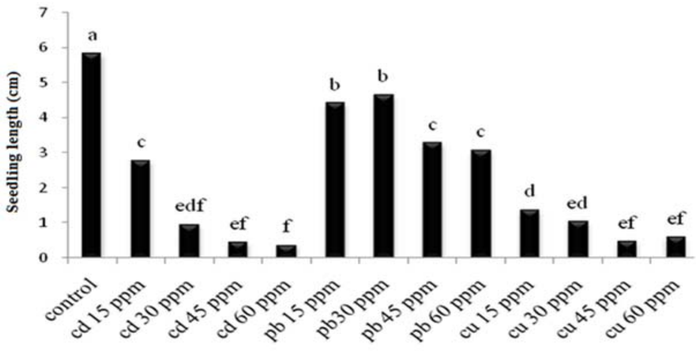

Fig2. Effect of heavy metals stress on seedling length

The lowest concentrations of Pb had stimulatory effects on seedling dry weight and higher seedling dry weight was related to Pb at 15 ppm, whereas the lowest value of seedling dry weight was observed in Cd at 30 and 45 ppm (fig 3). The Similar results obtained from *L.leucocephala* by Shafiq et al (2008).

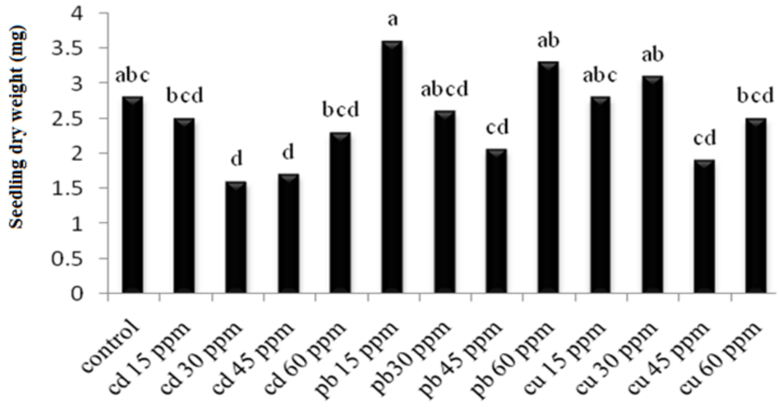

Fig3. Effect of heavy metals stress on seedling dry weight

Seedling vigour index 1 constantly decreased with increase in metal levels (fig 4). The High concentrations of Cd and Cu (40 and 60 ppm) showed lowest value of seedling vigour index 1. Highest value of seedling vigour index 2 was related to Pb at 15 ppm (fig 5). Saberi et al (2010) reported that Cd and Cu caused a significant reduction in seedling vigour index of Atriplex lentiformis.

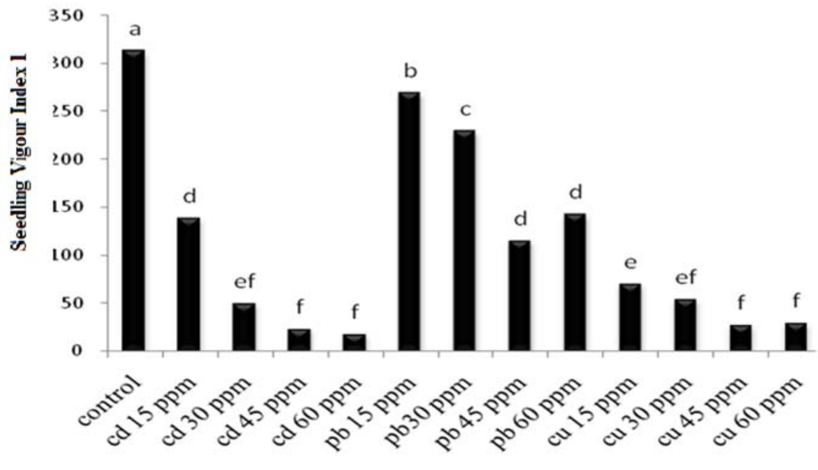

Fig4: Effect of heavy metals stress on Seedling vigor index 1

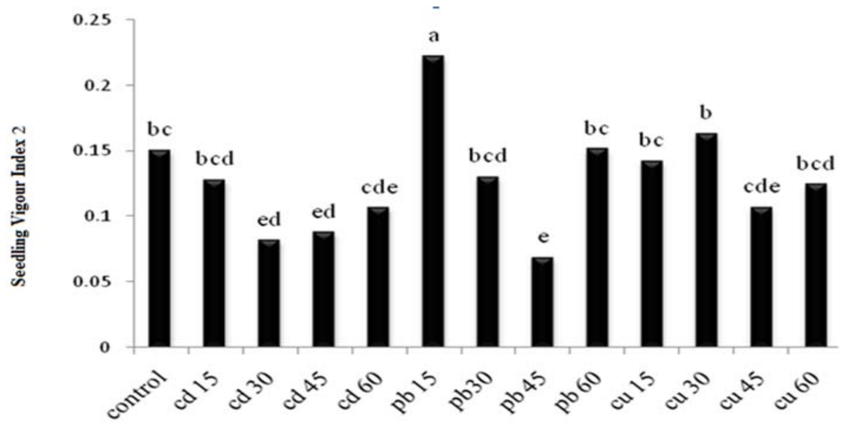

Fig 5: Effect of heavy metals stress on Seedling vigor index 2

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Effect of zinc fertilization on cadmium toxicity in Zea maize Grown in zinc-deficient soil

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Abstract

The effect of increasing application of zinc (Zn) and cadmium (Cd) on shoot concentrations of Zn, Cd, Fe and Mn was studied in maize cultivars. Plants were grown in severely Zn-deficient calcareous soil treated with increasing Zn (0 and 10 mg kg^{-1} soil) and Cd (0, 10 and 25 mg kg^{-1} soil) and harvested after 45 days of growth under greenhouse conditions. Increasing Cd application to Zn-deficient plants tended to decrease Zn concentrations in plants, whereas in plants with adequate Zn, concentrations of Zn were either not affected or increased by Cd. The results show that maize was more sensitive to both Zn deficiency and Cd toxicity. Cd application significantly increased Cd and Fe concentration of plants. Highest increase in Fe concentration observed in 25:10 treatment in B cultivar (159 mg kg^{-1}). Amount of Mn concentration was decreased with cadmium application. This decrease was 27% in 25:10 treatment then blank treatment. Increases in Zn application tend to enhance in Mn concentration. But cadmium increase in higher concentrations was deactivated the positive effect of Zinc.

Key words: Cadmium, Fe, Mn, Zea mays, Zinc concentration

Introduction

Cadmium is a toxic heavy metal released from smelting and absorbed and bioaccumulated by organisms along the food chain. Cadmium binds to organic molecules by forming bonds with sulfur and nitrogen, thereby inactivating proteins, and is one of the three contaminants which, according to the EPA, pose the most threat to the environment. The factors affecting Cd uptake and expression of Cd toxicity in plants, the plant nutritional status with respect to zinc (Zn) is the one which has been most studied. Generally, Zn applications decrease Cd uptake and accumulation in plants (Honma and Hirata, 1978). Hart et al. (2002) attributed the competitive interaction between Cd and Zn for uptake to the existence of a common transport system on the plasma membranes. Supporting these results, Gomes et al. (2002) showed that uptake of Cd is mediated through a Zn-transporter protein across the plasma membrane of yeast cells. Zinc was shown also to interfere with phloem-mediated Cd transport in durum wheat, possibly by competing with Cd for binding sites of a common transporter protein on the plasma membranes of sieve tube cells (Cakmak et al., 2000). Most experiments dealing with effects of increasing Zn treatments on Cd accumulation in plants have been carried out in soil or solution cultures containing normal or excess levels of Zn. Instead, we have used a severely Zn-deficient calcareous soil to study the effects of increasing Zn and Cd applications on the shoot growth and Cd toxicity. For better characterization of the relationship between Cd and Zn, maize cultivars differing in tolerance to Zn deficiency were used in the experiment, and the concentrations of Fe and Mn of plants were also measured.

Materials and methods

Seeds of maize were sown in pots containing 4 kg of Zn-deficient calcareous soil prepared from Kordan area in Karaj. This soil had a sandy loam texture, a pH of 8.3 and contained 0.9% organic matter and 6% CaCO3 as measured by the standard methods given in Jackson (1958). Total Zn and Cd concentrations of the soil were 0.4 mg Zn kg^{-1} and 1.13 mg Cd kg^{-1}, measured after wet-ashing as described by Jackson (1958). Before potting, soils were treated with 2 levels of Zn (0 and 10 mg kg^{-1} soil) and 3 levels of Cd (0, 10 and 25 mg kg^{-1} soil) in the forms of $ZnSO_4$, $7H_2O$, and $Cdcl_2$, H_2O. Each pot received 600mgN kg^{-1} soil as $Ca(NO_3)_2$, 200 mg P kg^{-1} soil as KH_2PO_4 and 2.5 mg Fe kg^{-1} soil as FeEDTA. All nutrients were mixed homogenously with soil before sowing. After 40 days of growth under greenhouse conditions, shoots were harvested and dried at 75°C for determination of shoot dry weight and concentrations of Cd, Zn, Fe and Mn. Cadmium, Zn, Fe and Mn were measured by Shirnadzu AA-660 Atomic Absorption Spectrophotometer (Jackson, 1958).

All sampling and measurements were carried out by using three replications. The results of experiment were subjected to variance analysis using MSTAT-C packet and SAS9 program. Least significant difference was used to compare the main treatment and interaction effects at p<0.0l.

Results

Cadmium application significantly increased Cd concentration of plants (Fig. 1). At each level of Zn treatment, cultivar B had a higher Cd concentration than cultivar A. Varied level of Zn in soil did not consistently affect Cd concentration of plants. Zn treatment tended to increase Cd concentration. Increasing Cd application to Zn-deficient plants tended to decrease Zn concentrations in plants, whereas in plants with adequate Zn, concentrations of Zn were either not affected or increased by Cd. Zinc application enhanced Zn concentration of plants, and the enhancement in Zn concentration was more evident. For plants exposed to Zn deficiency, increasing Cd application reduced the Zn concentration of the plants. When plants were treated adequately with Zn, increasing Cd enhanced Zn concentration in cultivars (Fig. 2). Iron and Mn concentrations of plants were higher in Zn-deficient plants. When Cd was not added, Zn deficiency enhanced the Mn concentrations of plants but not affected Fe concentration. Cadmium enhanced Fe under Zn deficiency and was sasme affect at adequate Zn application (Figs. 3). Zn treatment tended to decrease Mn concentration and increasing Cd application to Zn-deficient plants tended to decrease Zn concentrations in plants (Figs. 4). Cd application significantly increased Cd and Fe concentration of plants. Highest increase in Fe concentration observed in 25:10 treatment in B cultivar (159 mg kg^{-1}). Amount of Mn concentration was decreased with cadmium application. This decrease was 27% in 25:10 treatment then blank treatment (Figs. 4).

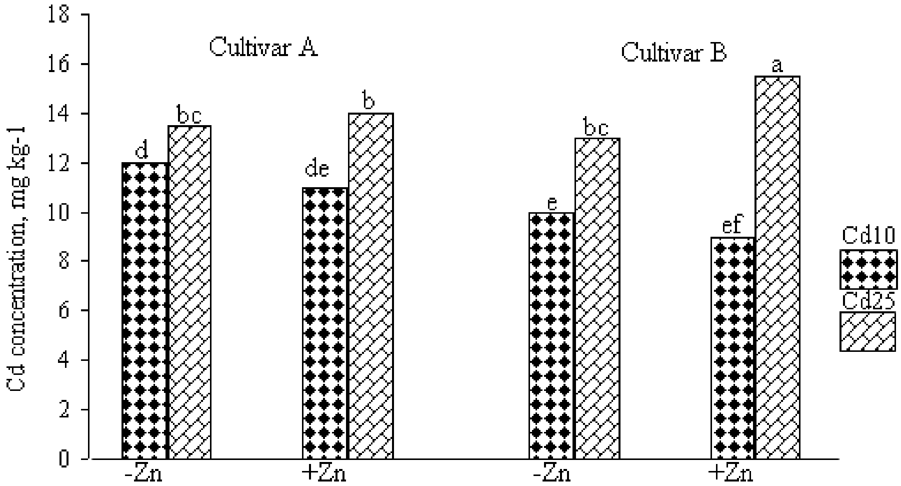

Fig. 1. Effect of increasing Cd application on shoot Cd concentration of Zea maize cultivars grown in a Zn-deficient calcareous soil, with (+Zn: 10 mg kg_1 soil) and without Zn (_Zn) treatment, different letters indicate significant difference at p<0:01; LSD test.

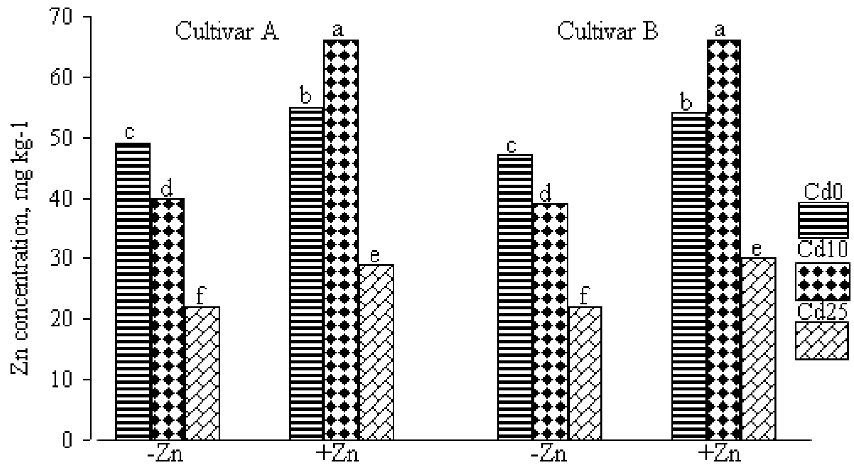

Fig. 2. Effect of increasing Cd application on shoot Zn concentration of Zea maize cultivars grown in a Zn-deficient calcareous soil, with (+Zn: 10 mg kg_1 soil) and without Zn (_Zn) treatment, different letters indicate significant difference at p<0:01; LSD test.

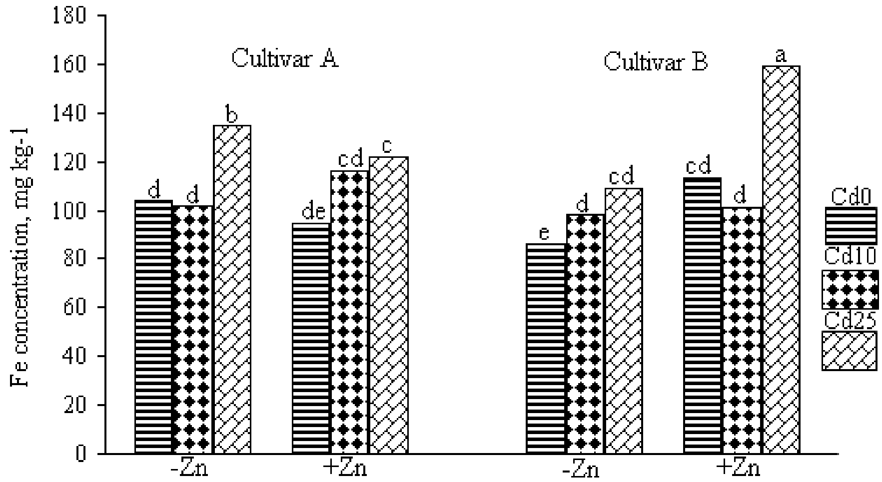

Fig. 3. Effect of increasing Cd application on shoot Fe concentration of Zea maize cultivars grown in a Zn-deficient calcareous soil, with (+Zn: 10 mg kg_1 soil) and without Zn (_Zn) treatment, different letters indicate significant difference at p<0:01; LSD test.

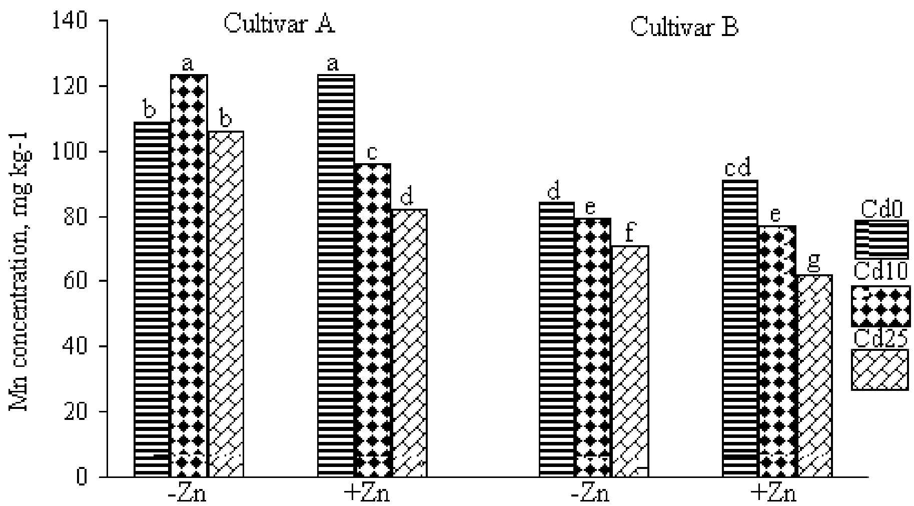

Fig. 4. Effect of increasing Cd application on shoot Mn concentration of Zea maize cultivars grown in a Zn-deficient calcareous soil, with (+Zn: 10 mg kg_1 soil) and without Zn (_Zn) treatment, different letters indicate significant difference at p<0:01; LSD test.

Discussion

Cadmium toxicity becomes more severe when plants are exposed to Zn deficiency. This may imply that an enhanced release of phytosiderophores from roots of Zn-deficient plants caused an increase in root uptake and shoot transport of Cd with the consequence of severe Cd toxicity symptoms (Oliver et al., 1997). Some authors have suggested that interactive pattern is antagonistic, whereas others argued that it is synergistic and Cd toxicity is enhanced by Zn addition (Dudka et al, 1999). Wu and Zhang (2002) found that increasing Zn application could alleviate Cd toxicity stress in barley by improving growth and reducing membrane damage. The results in Fig. 1 indicate that the greater sensitivity of Zea maize to Cd toxicity is related to plant Cd concentration. Zn addition tended to enhance Cd concentration of plants at the highest Cd treatment (Fig. 1). There is increasing evidence showing that Cd toxicity in plants reflects oxidative cell damage catalyzed by reactive oxygen species (ROS), and therefore the ability of plants to detoxify ROS under Cd toxicity is an important tolerance mechanism against Cd toxicity (Shah et al., 2001). Zinc has several protective roles in alleviation of cell damage by ROS produced under environmental stress conditions, and Zn is therefore involved in expression of plant tolerance mechanisms under stress conditions such as drought, low temperature stress, high light, salinity and Fe toxicity (Cakmak, 2000). Possibly, Zn may also protect plants from Cd toxicity by enhancing activity of antioxidative enzymes such as superoxide dismutase, a Zn-containing enzyme, and by competing with Cd for binding to eSH groups of enzymes and membrane proteins. In this regard, Wu and Zhang (2002) recently showed that Cd toxicity-induced decreases in shoot dry matter production and increases in membrane damage could be alleviated by Zn treatment. When compared to Zn-sufficient plants, Zn-deficient plants had greater Fe concentrations and lower Mn concentration at high level of Cd supply (Figs. 3). This result appears to be related to competition between Zn, Fe and Mn for uptake by roots and translocation into the shoot. Possibly, there is a common transport system on the plasma membrane of root cells for the corresponding metals (Hart et al., 2002). In the case of increasing Cd supply, Mn concentrations of plants were markedly reduced by Zn deficiency, but not affected at adequate Zn treatment (Figs. 4). Fe and Mn concentrations of shoot were much lower when compared to the plants grown under low Cd treatment and adequate Zn treatment (Figs. 3 and 4). These indicate that low levels of Mn in Zn-deficient and Cd-toxic plants might also be a consequence of greater root damage caused by combined effects of Zn deficiency and Cd toxicity. It is also well-known that under Zn deficiency plants have much higher capacity to take up Mn from soils irrespective of root or shoot growth (Cakmak, 2000).

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Evaluating the Allelopathic Potential of Decomposed Plant Residues of Some Weeds into Soil on Paddy Rice Variety

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Abstract
After the crop harvest, weed residues are major source of phytotoxins into soil. A biotic and microbial decomposition significantly affect the concentration of allelochemicals reaching other plants. To evaluate the allelopathic potential of paddy weeds like C*yperus difformis*, *Echinochloa crusgalli* and *Sagittaria trifolia*, dried powder plant parts of studied weeds at different concentrations i.e. 2, 4, 8, 16 and 32g into 250g loamy soil (six weeks) on seedling length and germination percentage of rice variety- Neda to form factorial examination in randomized complete block design with three replications were examined in Babol (Iran) at 2010. Decomposition bioassay of *Cyperus difformis* on test crop showed that seed germination was significantly decreased with an increase in amount of plant material added in the soil. Decayed plant materials of the studied weed also had inhibitory effect on both radicle and shoot length of test crop in all treatments. Results of decayed plant parts of *Echinochloa crusgalli* revealed that seedling length of Neda variety was significantly hampered by all treatments except 2g. Germination of paddy seeds of test crop was hampered with higher residues rates 16 and 32g about 26.93 and 30.77% respectively. Results of decayed plant parts of *Sagittaria trifolia* on seedling length of test crop inversely related to amount of plant material. Seed germination was not significantly affected by any treatment. The results demonstrated that the allelopathic potential of studied weeds and suggested that those weeds have inhibitory effect due to allelochemicals which are present in dried part with different values.
Keywords: Soil, Plant materials, Paddy weeds, Decomposition, Allelopathic potential.

Introduction

Allelopathy is the phenomenon in which living or dead plant material, including decaying litter, releases chemicals that inhibit (rarely stimulate) the growth of associated plants (Rice, 1984). The decomposition of plant residues potentiality provides the largest quantity of allelochemicals that may be added to the rhizosphere. After plant death, materials compartmentalized in cells are released and these processes with regard to allelopathy are the nature of the plant residues, the soil type and the conditions of decomposition. Patrick et al (1964) reported that decomposing on the decomposing condition, substances highly toxic, non toxic or stimulatory to may be found during the decomposition of similar plant residues. In general more sever and persistant toxicity has been reported from cold and wet soil (Mc Calla and Haskins, 1964 and Patrick et al. 1964). The purpose of this study was to assessing the allelopathic effect of decomposed plant residused of some weeds viz. *Cyperus difformis* L., *Echinochloa crusgalli* (L.) P. Beauv. and *Sagittaria trifolia* L. on seed of rice variety- Neda.

Material and methods

Selected weeds viz. *Cyperus difformis* and *Echinochloa crusgalli and Sagittaria trifolia* were collected from different paddy field of Babol (Iran) at maturity stage. These weeds were air-dried in shade and powdered to tiny pieces and stored in polythene bags till used. To study allelopathic potential of decomposed plant residue of selected weeds, powdered plant parts of every weed were mixed in 250 g loamy soil at the rates of 2, 4, 8,16 and 32 of plant material to wither away for six weeks and 10 ml distilled water were added daily separately (Avchar and Deokule, 2007). After six weeks, these mixtures were placed in sterilized petridish at the rate of 20g per petridish to form factorial examination in randomized of complete block design (RCBD) and were kept in triplicate at room temperature (25- 28^{o}C). These petridish were wetted with 10 ml of distilled water and 10 seeds of Neda variety were sown on moist surface of the soil. 20g of loamy soil free from decomposing plant were kept in petridish as control. After 168 hours, seed germination percentage was determined and radicle and shoot length in cm was measured. Collected data from the experiment was analyzed by SPSS package (version 17- two way analysis of variance) and the means was compared by Duncan's Multiple Rang Test (DMRT).

Results and discussion

Results of decomposition bioassay of *Cyperus difformis* showed in Table (1) indicated that seed germination of Neda variety was decreased with an increase in amount of plant material added in the soil. The greatest inhibitory effect was found at the rate of 32g plant materials in the soil over control about 50.01 %. It was also recorded that decayed plant materials into 250g of soil had inhibitory effect on both radicle and shoot length of test crop in all treatments. The effect of highest residues (32g) was more pronounced. The inhibitory effects on shoot length of seedling were greater in all treatments of plant parts mixed in the soil as compared with radicle length. This study also demonstrated that seedling growth of test crop was more hampered in all treatments as compared with seed germination of test crop. Data analysis of decayed plant parts of *Echinochloa crusgalli* (in the same Table) showed that elongation of shoot was significantly inhibited in all treatments as compared with control though level of inhibitory effects were same among all treatments in higher and lower rates of plant material added into soil statistically. On the contrary, different rates of dried plant parts of above weed did not show significant harmful effects on elongation of radicle as compared with control. In general, seedling growth and seed germination of Neda variety were affected with all treatments except 2g of plant parts into soil with same level of inhibitory effect. Results of decomposition bioassay of *Sagittaria trifolia* on growth of Neda seedling and seed germination (Table 1) revealed that shoot length of test crop was significantly diminished by all treatments except at the value of 2g decayed plant parts into soil. The plant materials at 16 and 32g had highest inhibitory effect about 66.67 and 87.83 % respectively. While radicle length of test crop significantly was hampered at value of 32g (61.68 %) followed by 16g (39.42%) incorporated plant in the soil. However, the inhibition in the overall on seedling growth was inversely related to amount of plant material allowed for decomposing. It was also observed that plant material kept in the soil did not show appreciable affect on the seed germination in higher and lower treatments of plant material. The results demonstrated that the allelopathic potential of studied weeds and suggested that those weeds have inhibitory effect due to allelochemicals which are present in dried part with different values.

Table 1. Decomposition bioassay of selected weeds on seedling growth and seed germination of Neda variety.

Weeds	Quantity of plant parts in decomposition (g/ 250g soil)	Rg (cm)	Sg (cm)	Tsg (cm)	germination percentage
Cyperus difformis	Control (D.W)	5.48±0.51*a*	3.45±0.36*a*	8.93±0.36*a*	86.67±11.55*a*
	2.00	3.77±0.18*b** (- **31.20**)	2.00±0.83*b* (- **42.03**)	5.77±0.77*b* (- **35.39**)	80.00±10.00*ab* (- 7.70)
	4.00	3.23±1.22b **(- 41.06)**	1.39±0.53bc **(- 59.71)**	4.62±1.51b **(- 48.26)**	66.67±5.77bc (- 23.08)
	8.00	3.24±0.69*b* **(- 40.88)**	1.29±0.82*bc* **(- 62.61)**	4.53±1.38*b* **(- 49.27)**	56.67±5.77*cd* **(- 34.61)**
	16.00	3.43±0.50*b* **(- 37.41)**	1.51±0.63*bc* **(- 56.23)**	4.94±1.13*b* **(- 44.68)**	56.67±5.77*cd* **(- 34.61)**
	32.00	1.88±0.43*c* **(- 65.69)**	0.60±0.44*c* **(- 82.61)**	2.48±0.85*c* **(- 72.23)**	43.33±15.27*d* **(- 50.01)**
Echinocholoa crusgalli	**Control (D.W)**	5.48±0.51*a*	3.45±0.36*a*	8.93±0.36*a*	86.67±11.55*a*
	2.00	5.52±0.30*a* **(+ 0.73)**	2.53±0.63*b* **(- 26.67)**	8.05±0.68*ab* **(- 9.85)**	86.67±15.27*a* **(0.00)**
	4.00	4.68±0.80*a* **(14.60)**	1.85±0.12*b* **(- 46.38)**	6.53±0.92*b* **(- 26.88)**	83.33±5.77*b* **(- 3.85)**
	8.00	4.64±0.39*a* **(15.33)**	1.98±0.08*b* **(- 42.61)**	6.62±0.31*b* **(- 25.87)**	83.33±5.77*b* **(- 3.85)**
	16.00	4.70±0.86*a* **(14.23)**	2.08±0.53*b* **(- 39.71)**	6.79±1.27*b* **(- 23.96)**	63.33±5.77*b* **(- 26.93)**
	32.00	4.27±0.82*a* **(22.08)**	2.24±0.47*b* **(- 35.07)**	6.51±1.29*b* **(- 27.01)**	60±17.32*c* **(- 30.77)**
Sagittaria trifolia	**Control (D.W)**	5.48±0.51*a*	3.45±0.36*a*	8.93±0.36*a*	86.67±11.55*a*
	2.00	5.38±0.50*a* **(- 1.82)**	2.98±0.06*ab* **(-13.62)**	8.35±0.50*ab* **(- 6.49)**	96.67±5.77*a* **(+ 11.50)**
	4.00	4.40±0.34*ab* **(- 19.71)**	2.25±0.86*b* **(- 34.78)**	6.64±0.67*b* **(- 25.64)**	83.33±5.77*a* **(- 3.85)**
	8.00	4.36±0.62*ab* **(- 20.44)**	2.47±0.46*b* **(- 28.41)**	6.83±1.04*b* **(- 23.52)**	83.33±15.27*a* **(- 3.85)**
	16.00	3.32±1.04*b* **(- 39.42)**	1.15±0.64*c* **(- 66.67)**	4.47±1.56*c* **(- 49.94)**	86.67±15.27*a* **(0.00)**
	32.00	2.10±0.49*c* **(- 61.68)**	0.42±0.17*c* **(- 87.83)**	2.52±0.62*d* **(- 71.79)**	73.33±11.55*a* **(- 15.39)**

. Number with different letters within column refers significant difference with confidence level 95% according Duncan's Univariete- range test. The data in parenthesis indicate % inhibition (-) or Stimulation over control.

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Volatilization bioassay method for assesing the allelopathic toxicity of paddy weeds into soil and water

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Abstract

The volatile compounds may be absorbed in vapor by surrounding plants be absorbed from condensate in dew or may reach the soil and be taken up by the roots. To test volatile compounds, are diffuse from the weeds viz. C*yperus difformis*, *Echinochloa crusgalli*, *Paspalum paspaloides* and *Sagittaria trifolia*, fresh weed species were collected from paddy fields of Babol (Iran) at 2009. From each plant species, fresh parts (150 g) were kept in surface sterilized airtight jar for 48 h. Soil (150 g) and water (150 ml) were kept inside the airtight jar separately in such a way that they are not indirect contact with plant materials, and the air inside only acts as a carrier for volatile substances. The soil and water, thus obtained were used for further bioassay using Tarom variety seeds. Bioassay of *Cyperus difformis* showed that soil kept with fresh plant material of above weed significantly hampered on both seedling growth and seed germination of the test crop about 68.91 and 52 % respectively. Water kept with above plant parts had a weak effect on mentioned parameters. Soil and water kept with fresh plant materials of the other weeds did not show significant hamper effect on seedling growth of the test crop. The weeds like *Echinochola crusgalli* and *Paspalum paspaloides* in water phase significantly decrease germination percentage of test variety. The volatile compounds like terpenoids were exhibited by preliminary phytochemical screening. The response to volatilization of fresh plant parts of studied weeds by Tarom variety varied depending on kinds of weed species.

Keywords: Volatile compounds, Fresh plant part, Paddy weeds, Soil and water, Bioassay.

Introduction

In a few case, allelochemicals may volatilize and absorbed directly from the atmosphere by neighbouring plants and absorbed from condensate in dew, be absorbed on the soil particles and subsequently taken up by contact with plants or from the soil solution (Muller, 1966). Apparently several terpenoids transfer in these way, higher plants produce a variety of essential oils of plants. The plants which is rich in such compounds, these may be released continuously to the atmosphere, with this process enhanced in hot weather; therefore, this phenomenon is observed in the arid regions of the world. Thus a number of species from several climatic regions may express allelopathy through volatilization (Rice, 1974 and Horsley, 1977). The objective of this study was to evaluate the allelopathic potential of Volatile toxins from the paddy weeds like *Cyperus difformis* L., *Echinochloa crusgalli* (L.) P. Beauv., *Paspalum paspaloides* (Michx.) Scribner and *Sagittaria trifolia* L. in soil and water on radicle, hypocotyle and seed germination of rice variety-Tarom.

Material and methods

To test volatile substance (s) from paddy weeds like *Cyperus difformis*, *Echinochloa crusgalli*, *Paspalum paspaloides* and *Sagittaria trifolia* fresh plant parts at full flowering stage were kept in airtight jar for 48 h. Soil (150 g) and water (150 ml) were kept separately inside the jar in such away that they were not in direct contact with the plant materials and the air inside only acted as a carrier. The soil and water thus obtained were bioassay using the 10 seeds of rice variety- Tarom. 10 ml water was used per petridish to moisten the filter paper and 10 g soil was used as a layer of

growth medium and irrigated with 10 ml distilled water. Radicle and hypocityl length of seedling and seed germination were recorded after 168 h (Avchar and Deokule, 2007). Preliminary phytochemical method was used for detection and confirmation of Terpenoids in root, stem and leaf of studied weeds by using standard method of Trease and Evans,1989 and Harborne,1973 c.f. Edeoga et al. 2005. Student's t- test was used for comparison of two means. Percentage of inhibition or stimulation over control
somewhere calculated according to following formula
% inhibition or stimulation: (Control - Treatment / Control) X 100

Result and discussion

Results of volatilization bioassay of *Cyperus difformis* showed in Tables (1 and 2) indicated that soil kept with fresh plant material of above weed in airtight jar significantly hampered on radicle length of paddy seedling (t- test, $p < 0.05$) than hypocotyl length of test crop which was not significantly affected. However, seedling growth of test crop was significantly inhibited by fresh plant parts of above weed. It was also recorded that soil kept in airtight jar significantly decreased seed germination (t-test, $p < 0.05$). In case of water kept with above plant parts had a weak effect on radicle and hypocotyl as well as seed germination of test crop. It indicated that soil absorbed some volatile substance(s) more from the weed plant that is responsible for allelopathic potential of above mentioned weed. The toxic chemical that inhibited the effect on the growth of crop plants like terpenoids was exhibited by preliminary phytochemical screening method in root, stem and leaves of *Cyperus difformis* and other weeds. However, seedling growth of test crop was more hampered in soil kept with fresh material as compared with seed germination about 68.91 and 52.00 % respectively.

Table 1. Effects of volatilization bioassay from *Cyperus difformis* (water) on seedling growth and seed germination of Tarom variety.

Type of bioassay	Radicle length (cm)	Hypocotyl length (cm)	Seedling growth (cm)	Seed germination (%)
Control	4.71±0.57	3.86±0.38	8.57±0.93	83.33±5.77
Treated water	5.44±1.02	3.30±1.22	8.74±1.81	86.67±11.55
p- value	0.343	0.492	0.890	0.678

Table 2. Effects of volatilization bioassay from *Cyperus difformis* (soil) on seedling growth and seed germination of Tarom variety.

Type of bioassay	Radicle length (cm)	Hypocotyl length (cm)	Seedling growth (cm)	Seed germination (%)
Control	4.66±0.65	3.06±0.97	7.72±0.64	83.33±5.77
Treated soil	1.32±0.68	1.08±0.54	2.40±1.23	40.00±10.00
p- value	0.016	0.050	0.015	0.015

During volatilization bioassay water kept with fresh plant material of *Echinochloa crusgalli* inhibited hypocotyl length of test crop (t- test, $p < 0.05$) than radicle length which was not significantly affected. However, negative impact of that collected water on hypocotyl length did not show a decrease on seedling growth of test crop. It was also recorded that seed germination of test crop was significantly hampered (t- test, $p < 0.05$). In case of soil kept with above plant parts had a weak effect on both radicle and hypocotyl as well as seed germination of test crop (Tables 3 and 4). It might be soil phase absorb less quantity of volatile substance(s) from this weed as compared with water phase.

Table 3. Effects of volatilization bioassay from *Echinochloa crusgalli* (water) on seedling growth and seed germination of Tarom variety.

Type of bioassay	Radicle length (cm)	Hypocotyl length (cm)	Seedling growth (cm)	Seed germination (%)
Control	4.71±0.57	3.86±0.38	8.57±0.93	83.33±5.77
Treated water	4.75±0.76	2.19±0.62	6.93±0.61	60.00±10.00
p- value	0.951	0.030	0.076	0.039

Table 4. Effects of volatilization bioassay from *Echinochloa crusgalli* (soil) on seedling growth and seed germination of Tarom variety.

Type of bioassay	Radicle length (cm)	Hypocotyl length (cm)	Seedling growth (cm)	Seed germination (%)
Control	4.66±0.65	3.06±0.97	7.72±0.64	83.33±5.77
Treated soil	3.07±0.76	2.19±1.38	5.26±1.52	63.33±15.27
p- value	0.065	0.114	0.074	0.111

Results of volatilization bioassay of *Paspalum paspaloides* showed that soil kept with fresh plant parts in airtight jar did not show significant effect on both radicle and hypocotyl length of paddy seedling as well as seed germination of test crop. It was also exhibited that water kept with plant parts of above said weed significantly hampered on hypocotyl length as well as seed germination of test crop (t- test, $p < 0.05$) [Table 5and 6]

Table 5. Effects of volatilization bioassay from *Paspalum paspaloides* (water) on seedling growth and seed germination of Tarom variety.

Type of bioassay	Radicle length (cm)	Hypocotyl length (cm)	Seedling growth (cm)	Seed germination (%)
Control	4.71±0.57	3.86±0.38	8.57±0.93	83.33±5.77
Treated water	4.08±0.39	2.19±0.62	6.27±0.94	60.00±10.00
p- value	0.198	0.030	0.053	0.039

Table 6. Effects of volatilization bioassay from *Paspalum paspaloides* (soil) on seedling growth and seed germination of Tarom variety.

Type of bioassay	Radicle length (cm)	Hypocotyl length (cm)	Seedling growth (cm)	Seed germination (%)
Control	4.66±0.65	3.06±0.97	7.72±0.64	83.33±5.77
Treated soil	2.99±1.22	1.38±0.57	4.37±1.79	80.00±10.00
p- value	0.115	0.073	0.051	0.644

Data analysis of volatilization bioassay of *Sagittaria trifolia* showed in Tables (7 and 8) revealed that water kept with plant parts in airtight jar significantly hampered on hypocotyl length of test crop only (t- test, $p < 0.05$). It was also recorded that soil kept with fresh plant parts of above selected weed had a weak effect on radicle and hypocotyl length as well as seed germination of Tarom variety.

Table 4.28. Effects of volatilization bioassay from *Sagittaria trifolia* (water) on seedling growth and seed germination of Tarom variety.

Type of bioassay	Radicle length (cm)	Hypocotyl length (cm)	Seedling growth (cm)	Seed germination (%)
Control	4.71±0.57	3.86±0.38	8.57±0.93	83.33±5.77
Treated water	4.24±1.41	2.05±0.45	6.29±1.82	73.33±14.53
p- value	0.619	0.019	0.135	0.540

Table 4.29. Effects of volatilization bioassay from *Sagittaria trifolia* (soil) on seedling growth and seed germination of Tarom variety.

Type of bioassay	Radicle length (cm)	Hypocotyl length (cm)	Seedling growth (cm)	Seed germination (%)
Control	4.66±0.65	3.06±0.97	7.72±0.64	83.33±5.77
Treated soil	3.89±0.31	2.62±0.29	6.51±0.60	86.67±15.27
p- value	0.147	0.494	0.086	0.111

This study showed that the response to volatilization of fresh plant parts of studied weeds by Tarom variety varied depending on kinds of weed species.

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The Effect of Humic Acid Application on Soil Properties and *Brassica napus* L. Yield in Nickel-contaminated Soil

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Abstract
It is well known that some elements such as nickel among other heavy metals are essential for plant growth in low concentrations. Nevertheless, beyond certain threshold concentrations, these elements become toxic for most plants. In addition, soil with low organic matter in arid regions decreases the productivity of soil. The present study was conducted to investigate the effect of humic acid (HA) application on the soil properties, Ni accumulation in roots and shoots and the growth of rapeseed (*Brassica napus* L.) in Ni-contaminated soil (125 $mg.kg^{-1}$). Rapeseed (*B. napus*) was grown in the framework of a pot experiment with three replications in a greenhouse. Pots were filled with 7 kg of artificially contaminated soil and then were treated with 125 gr per pot humic acid. After 8 weeks, the plants were harvested and soil, shoot and root of plants were collected. Then, the amount of shoot and root dry weight and Ni concentration in plant were measured. The results showed significant ($p<0.01$) effect of humic acid to reduce acidity and increase the electrical conductivity and organic matter percent, that obviously these parameters have a significant impact on plant growth. Rapeseed shoot dry weight showed significant ($p<0.05$) enhancement in humic acid treatments. Moreover, Ni concentration in roots increased significantly ($p<0.05$) with the usage of humic acid. In conclusion, application of humic acid in Ni-contaminated soil enhanced the plant production due to providing better soil qualification and could possibly be an economical method of cultivation in Ni-contaminated soils in arid regions.
Keywords: humic acid, soil properties, growth, rapeseed (*Brassica napus* L.), nickel**.**

Introduction

Human activities release pollutants in the environment; heavy metals, in particular, originate from industrial emissions, mining activity, disposal of wastes and fertilizers and pesticides use (Marchiol et al., 2004). Plants have a natural propensity to take up metals. Some metals, such as Cu, Co, Fe, Mo, Mn, Ni, and Zn, are essential mineral nutrients. Others, however, such as Cd and Pb, have no known physiological activity (Lasat, 2002). This propensity is a drawback for human health when contamination of food crops is too high. Nickel is abundant in the crust of the Earth, comprising about 3% of the composition of the earth. Nickel averages 50 mg Ni $.kg^{-1}$ in soils and commonly varies from 5 to 500 mg Ni $.kg^{-1}$ soil. In sewage-amended soils or in contaminated soils, it is often difficult to relate total nickel load with plant productivity, as factors such as the chemical properties of the contaminant and base soil, pH, and oxidation-reduction state affect results (Kukier and Chaney, 2001). Plant species also differ in their ability to obtain nickel from soils and hence any measurement of soil nickel must be interpreted with consideration of the plant species of interest. However, it is not always clear that poor plant growth can be ascribed to any single factor concerning nickel. The nickel content of a plant is determined by the nickel availability in the soil, plant species, plant part, and season. Plants growing on serpentine soils (derived from ultramific rocks) or contaminated soils can accumulate high levels of nickel and other heavy metals (Salt et al., 1995). Species growing on the same soil can also vary dramatically in nickel content and within plant distribution. In general, nickel is transported preferentially to the grains, particularly under conditions of marginal nickel supply. Therefore, methods to minimize adverse effects of the pollutants toxicity on crop production have consequently gained a good deal of attention (Marchiol et al., 2004) All plants can extract metals from soil; however, some plants have demonstrated ability to extract, accumulate and tolerate high levels of heavy metals that are toxic to other organisms (Evangelou et al., 2004). Today, phytoextraction with *Brassica napus* L. has the potential to become a profitable enterprise when combined with biofuel production, especially in view of the increasing oil prices over the coming years. Rapeseed varieties are mainly used in food applications, but to a growing extent also in the production of biofuel (Veerle et al., 2006).

Regardless of the plants used, availability of heavy metals to plant roots is considered the key factor limiting the efficiency of phytoextraction (Felix, 1997). The degree of availability for uptake i.e., the phytoavailability of metals is affected by numerous soil factors, such as cation exchange capacity (Moore et al., 1995), pH (Hornburg et al., 1995; Reddy et al., 1995; Schmidt, 2003) and organic matter content (Li and Shuman, 1996). In addition, the speciation of the metal, which is correlated to the factors mentioned above (Reddy et al., 1995), and the metal species (Atanassova, 1999) itself, play an important role. Soil OM has been of particular interest in studies of heavy metal retention in soils due to the tendency of transition metals to form stable complex with organic ligands (Chen, 1996). When using organic materials, the effect of their organic matter on heavy metals availability cannot be separated from other collateral effects on the soil properties, such as pH, Eh and the presence of phosphate and soluble salts (Walker et al., 2003). Humic substances also promote the formation of macro aggregates in mine tailings, increasing the pH, cation exchange capacity and organic carbon content of the soil (Ibrahim and Goh, 2004). Therefore composts and humic acid rich materials are adequate for the field-scale remediation of heavy metal polluted soils (Clemente et al., 2003), it being a cost effective method given the low price of residues and by-products that can be used as soil amendments. Humic acids (HA) represent the fraction of humic substances insoluble in water under acidic conditions, which becomes soluble and extractable at higher soil pH. Molecules of HA are characterized by acidic groups such as carboxyl and phenol OH functional groups (Hofrichter and Steinbuchel, 2001) and, when applied to the soil, play an important role in the transport, bioavailability and solubility of HM (Lagier et al., 2000). It has been demonstrated that HA contribute to reducing the physical mobility (diffusion, mass flow) of various metal species (e.g. Cu, Pb, Zn, Ni) in the soil, and thus reduce the consequent risk of lateral or vertical contamination of water bodies, as acetic acid extraction of metals is generally reduced with HA (Halim et al., 2003). Nickel, unlike many other divalent cations, is readily retranslocated within the plant likely as a complex with organic acids and amino acids (Cataldo et al., 1988). Nickel rapidly retranslocates from leaves to young tissues in the phloem, particularly during reproductive growth. Indeed, up to 70% of nickel in the shoots was transported to the seed of soybean (Tiffin, 1971).

In this study, the effects of applying humic acid was evaluated in terms of the soil properties, also both shoot and root growth in *Brassica napus* L., cultivated in soil artificially polluted by nickel. The ability to accumulate nickel in the shoots and roots was also investigated, aiming at identifying the relation between the humic acid usage and nickel accumulation in plant.

Materials and Methods

A pot experiment was conducted in the greenhouse of Islamic Azad University of Khorasgan (Isfahan). The soil was air-dried at room temperature, then sieved through a 2 mm sieve and characterized as is shown in table 1.

Table1. The physicochemical properties of the soil used in experiment before contamination

Ni DTPA	Ni total	P available	K available	N total	O.M	C.E.C	EC	pH $CaCl_2$	Sand	Clay	Silt
mg.kg^{-1}				%		cmolc.kg^{-1}	dS.m^{-1}		%		
0.28	51	46.05	449.7	0.13	0.25	10.7	2.4	7.9	16.17	20.83	63

For preliminary contamination of the soil, nickel sulfate ($NiSO_4 .6H_2O$), was used at concentrations of 335.64 gr.kg^{-1} soil. Nickel sulfate was applied by spraying to the soil which then saturated with de-ionized water and air-dried before using. Plastic pots (24 cm in diameter holding 7 kg of dry soil) were filled with soil and half of them mixed with 125 gr humic acid per pot. Then soils were brought to 75% of the ultimate field water capacity. Pots were arranged in a randomized complete block design with three replications and seeds of rapeseed (*B. napus*) were sown. No fertilizer was needed due to adequate amount of N, P and K in soil as is shown in table 1. All rapeseed plants were grown under controlled environmental conditions with about 15-hour light period, an average temperature about 32 ˚C and 65% relative humidity. After the first pair of true leaves appeared, seedlings were thinned to six plants per pot and kept constant until the end of the experiment. The plants were harvested 8 weeks after germination and separated into shoots (aboveground parts) and roots. The plant samples were washed free of all adhering soil in distilled water and oven dried at 65 °C for 48 hours to reach a constant weight. The dry weight was measured and the samples were grounded and homogenized. Ground shoot and root samples (1g) were digested in a mixture of concentrated HNO_3 and $HClO_4$ (4:1, by volume) and the nickel concentration in the solutions was determined (Chen et al., 2004) using the atomic absorption spectrophotometry method. Soil samples were collected after harvesting and pH, EC, and OM% of each sample were measured. Statistical analysis of the data was performed and the difference between specific pairs of means was identified by the Duncan test.

Results

Table 2 indicates the mean values of acidity (pH), electrical conductivity (EC) and organic matter percent (OM %) of soil samples, collected from each pot after harvesting the plants.

Table2. The mean table of measured parameters of soil after harvesting

Treatment	pH $_{CaCl_2}$	EC	OM
		$dS.m^{-1}$	%
HA_0	7.91 a	2.7 b	0.32 b
HA_1	7.75 b	4.0 a	0.41 a

The results showed that usage of humic acid significantly ($p<0.01$) reduced the acidity (7.91 to 7.75) and increased the electrical conductivity (2.7 to 4 $dS.m^{-1}$) also organic matter percent of soil (0.32 to 0.41%).

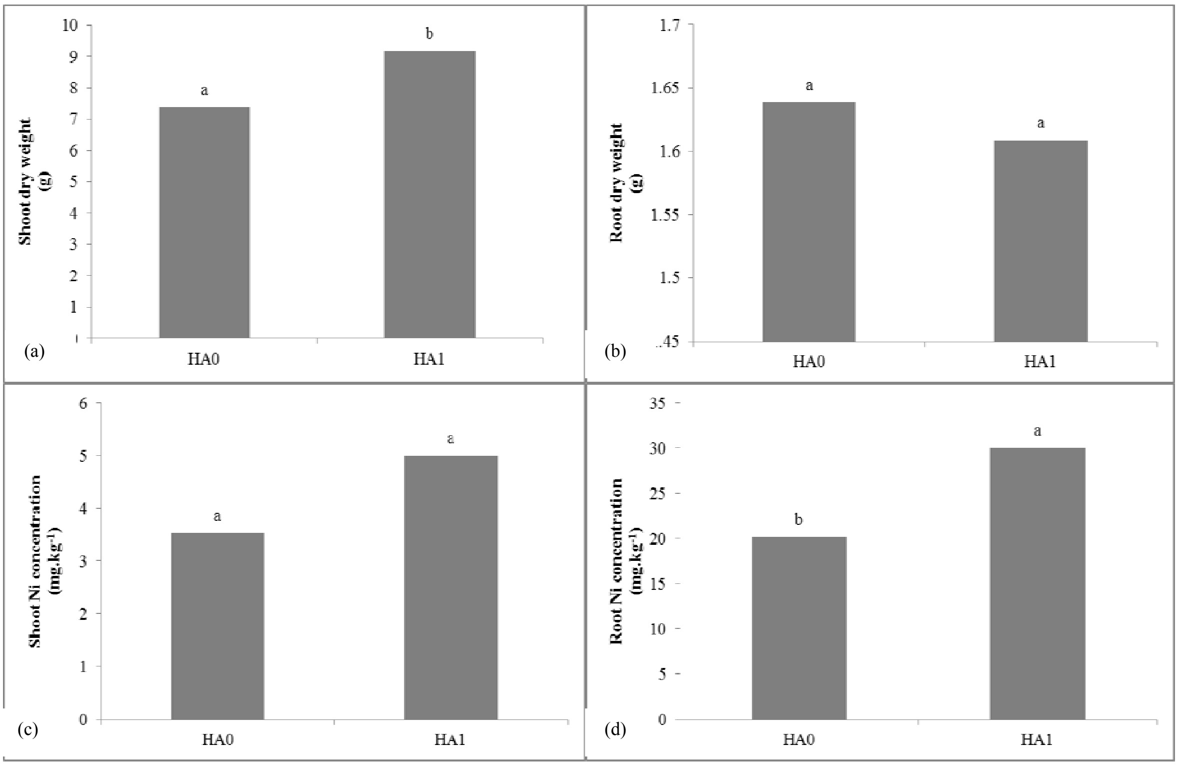

Fig1. The effect of humic acids application on shoot dry weight (a), root dry weight (b), shoot Ni concentration (c) and root Ni concentration (d) of Rapeseed (*B. napus*).

Figure 1 shows the dry matter yield of rapeseed and nickel accumulation in shoots and roots grown with (HA_1) and without (HA_0) the humic acid treatments. Our results showed that the differences between the shoot dry weight and root nickel concentration of the control treatment plants and the plants cultivated with the addition of humic acid were found to be statistically significant ($P<0.05$). However, there were no statistical significant differences among the root dry weight of the treatments, although the plants grown at humic acid treatments had the lowest weights (1.6 g). Likewise, there were no statistical significant differences between the shoots nickel concentration of plants with addition of humic acid, although humic acid treatments had the highest shoot nickel concentration (5 $mg.kg^{-1}$).

Discussion

The statistical analysis revealed that all measured parameters of soil changed significantly ($p<0.01$) by addition of humic acid. Humic acid may reduce the soil pH due to having phenol and carboxyl functional groups in its structure (Hofrichter and Steinbuchel, 2001). The role of pH in nickel availability was illustrated by Van de Graaff et al. (2002), who observed that long-term irrigation with sewage effluent increased heavy metal loading in soil, but that plant metal contents did not increase, apparently owing to the increased soil pH, iron complexation and coprecipitation, and precipitation of phosphorus–metal complexes. Indeed, the importance of considering soil pH is well illustrated by Kukier and Chaney (2001), who demonstrated that addition of limestone to raise soil pH, is highly effective in immobilizing nickel in situ and in reducing phytotoxicity. Wang et al. (2010), also stated addition of acid humic acid to sediments reduced soil pH from 7.7-7.6 to 7.2-7.1. According to Khan et al. (2006), soil pH was affected by adding humic acid to the soil through the irrigation water.

Soil electrical conductivity (EC) increased about 0.3 and 1.6 $dS.m^{-1}$ in humic acid and without humic acid (control) treatments respectively. Humic acid based products are used to promote plant growth and mineral nutrition (Lulakis and Petsas, 1995; Arancon et al., 2004). Consumption of humic acid initiates large amounts of salts into the soil, which increases the electrical conductivity of soil. Ke et al. (2003) also showed that sediments with addition of humic acid have higher salinity. In addition, EC enhancement in control soils may be due to accumulation of salts during irrigation.

İn this study, application of humic acid results in increasing OM% up to 0.41. Enhancement of OM% in control soil is probably because of roots exudations. Humic substances are naturally present in all agricultural soils and involve 80 percent of soil organic matter. Humic materials are the large sources of organic carbon for the global carbon cycle and are known as the major facilitators in transport of metal ions in the environment (Rashid, 1985). Applications of humic substances to soils with low content of clay and organic matter have produced significant growth responses (Lulakis and Petsas, 1995), especially in soils originates from arid regions which are poor at organic matter (OM%) content.

Humic acid usage increased shoot (above ground) dry weight significantly ($p<0.05$), but slightly decreased root dry weight. Humic acid decreased soil pH which results in more bioavailability of micro nutritions such as nickel and also release some essential nutriants and finally promoted rapeseed plants growth. Bandiera et al. (2009), showed that very little use of humic acid (0.1 $g.kg^{-1}$) regardless of the method of application, has had a positive effect on shoot mass weight, although compared to controls, the effect was not significant. Atiyeh et al. (2002) showed that plant growth gradually increased by adding humic acid from 1000 to 100 $kg.ha^{-1}$ to the soil. In case of root dry weight, the rapeseed plants growing in the humic acid treated soil received more amounts of nickel due to higher bioavailability of nickel in the soil and revealed more nickel concentration in root (up to 30 $mg.kg^{-1}$). The accumulation of nickel in roots results in appearing toxicity symptoms such as decreasing in root dry weight. Nickel concentrations above the toxicity levels of >10 mg.kg-1 dry weight in sensitive species, and >50 mg.kg-1 dry weight in moderately tolerant ones results in impaired root and shoot growth without any remarkable defining characteristics (Barker and Pilbeam, 2006). Previous studies have been reported that the addition of high concentrations of humic substances to soils would have an inhibitory effect on plant growth (Ke et al., 2003). The plant analyses indicated that although nickel concentration in the roots was considerable, but in case of shoots, it was comparatively low in plants. The accumulation of nickel in plant roots is very high in comparison with shoots which showed that in contaminated soils, the main part of the nickel taken up by the rapeseed, accumulated in the roots. According Bandiera et al. (2009), concentrations of heavy metals such as lead, copper, zinc, nickel, chromium, cadmium and manganese in forage radish roots was several times higher than their concentrations in the shoots.

In conclusion, this study has shown that rapeseed (*B. napus*) could possibly be used prosperously in nickel-polluted soil where its growth would be acceptable. Humic acid could have considerable influences on pH, EC and OM% in contaminated soil and also positive effects on the above ground growth although inhibited root growth due to toxicity effects of nickel accumulation.

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The Effects of Cadmium Contaminations on the Properties of *Chenopodium botrys* L. Weed

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Abstract

The aim of phytoremediation is remove heavy metals from environment or reduce pollution. It is a green revolution. In this research, Cadmium was added to soil as cadmium chloride in concentration (0, 25, 50, 75 and 125). The plant was maintained in greenhouse under controlled circumstances. Then its effects were evaluated on *chenopodium botrys* L. After harvest, cadmium content of the plant and soil was measured by ICP. Results of the experiment indicated that toxicity of cadmium was lead to reduce the plant growth in cadmium concentrations more than 10 mg/ kg soil. The Cd concentration was increased with cadmium contents in soil and the maximum accumulation occurred in the roots. High accumulation of Cd by this halophyte reveals that *chenopodium botrys* L. can be considered as a suitable plant for phytoremediate Cd-contaminated soils, despite the fact that its biomass decreases by increasing soil cadmium.

Evaluate the Effect of Rock Fragments and Flow Discharge on Distance of Rill Erosion Initiation

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Abstract
Rock fragments on the soil surface protect the soil against erosive agents. However, the effect of rock fragments cover on rill initiation is not well documented. The objective of this study was to evaluate the effect of rock fragment cover and the rate of flow discharge on rill initiation of erodible loess soils of Golestan province, Iran. The investigation was conducted using a flume with 6 m length, 0.5 m width, and 3% gradient. The treatments included rock fragment cover (0%, 10%, 20%, and 30%), and three levels of flow discharge (3, 6, and 9 lit. min^{-1}). The results showed that the surface cover of rock fragment had significant effect on different erosion processes, as well as rill initiation. Moreover, results indicated that velocity of water flow and the Froude number decreased but the Manning roughness coefficient increased (0.012 to 0.115 $m^{-1/3}$ s) with an increase in rock fragment cover, whereas the Reinhold's number remained nearly the same with a small variation among different rock fragment cover percent. This variable was increased with increasing flow discharge. In addition, distance of rill initiation increased with a rise in rock fragment cover. In general, rock fragment cover percentage was more effective than flow discharge rate to determine the distance and time of rill initiation.
Keywords: Loess soils, Rill erosion, Flume, Golestan province, Iran.

Introduction

Rill erosion is a major participant to soil loss from crop land area. Other erosion types, such as inter-rill, splash, or tillage erosion often lead to translocation of soil within the field (Parsons et al., 2004). Soil surface conditions (roughness, vegetation, and rock fragments cover) play important roles to control soil water infiltration, runoff generation, and reduction of soil erosion. A number of studies have showed that rock fragments cover has a significant effect on runoff and soil erosion. All these studies reported that runoff and erosion decrease with increasing rock fragments cover (Nyssen et al, 2001; Martinez-zavala and Jordan 2008; Tailong et al, 2010). The soil surface can be protected by rock fragments cover against the impacts of raindrops, surface sealing, detachment, and transport of soil particles (Martinez-zavala and Jordan 2008). So, the rock fragment cover on the soil surface reduces the total sediment yield (Rieke-Zapp et al 2007). The effects of rock fragments on eroding environment are: (1) protection of soil surface from direct impact of raindrops, and soil particles detachment, (2) decreasing the physical degradation of the soil surface, and (3) increasing the surface roughness and delaying overland flow and thus reducing detachment and transport capacity of the run off (Poesen and Lavee 1994).

Rock fragment surface cover can influence rill initiation, because of its effects on soil erosion processes. Studying the distance and time of rill initiation can be useful in soil conservation practices, such as determining of the appropriate intervals in plant works, terracing, and other conservation structures. The distance to rill initiation, on bare soil, decreases with increasing the slope steepness (Yao et al 2007). Rieke-Zapp et al. (2007) indicated without rock fragments in the soil, rill incision continued over time and headcutting increased for experiments with few or no rock fragments in the soil. A few studies have focused on surface cover and how it affects rill formation. The purpose of present study was to investigate the effect of rock fragment surface cover on the distance and time of rill formation, and understanding the relationship between the flow discharge and rock fragments cover in rill initiation.

Material and Methods

The experiment was conducted in the Laboratory of rainfall and runoff simulation, Soil Conservation and Watershed Management Research Institute, Tehran, Iran. A runoff simulator with a sloped plot (6 × 0.5 m) was used. The plot was initially prepared in a horizontal position. A 10 cm layer of coarse sand was uniformly placed in the bottom of the plot box; drainage holes in the bottom provided free drainage. On the top of the sand layer, a silty-loam soil (loess) was packed

loosely and evenly to a depth of 20 cm. The soil texture was 20% clay, 69% silt, and 11% sand. The soil was obtained from the root layer of a cultivated field located in the Loess Plateau area in Golestan province, Iran. The soil was air-dried, crushed and sieved with a 10 mm screen and then was packed in the plot to reach a bulk density of about 1.3 g cm^{-3}. During the packing process, a Rollers method was used to pack the soil uniformly in the plot. After packing, the soil surface was smoothed manually with a rake. Then rock fragments with 7 to 8 mm diameter were randomly distributed on the soil surface. The soil was saturated from below and allowed to equilibrate for 24 h, while the plot remained in a horizontal position to ensure a uniform initial soil moisture profile.
The treatments included rock fragment cover (0%, 10%, 20%, and 30%), each with three levels of flow discharge (3, 6, and 9 $Lit.min^{-1}$) that were tested at the 3% slope (the slope was same as the field). Each test was conducted 24 h after the saturation and pre wetting. The time of rill initiation was from runoff entry to plot until Primary rill formation moment. In each experiment, runoff and sediment samples were collected every minute until rill formation time. Runoff volumes and sediment mass were determined. Flow velocity was measured using a dye-tracing technique (potassium permanganate). The surface velocities (V_m) were converted to average velocity of flow profile (V) using the formula:

$$V = aV_m \qquad (1)$$

where a is a coefficient equal to 0.67 (Li et al. 1996).
Rill widths were measured with a ruler during the experiments. Each experiment ended, When a rill channel (at least 5 cm long, 0.5 cm deep, and 1 to 2 cm wide was formed (Torri et al, 1987)). After each test, the rills locations were determined throughout the profile accurately, by Laser Distance Meter. The average distances of all rills in a test were used as distance to rill initiation for subsequent calculations.
Flow depth was calculated as:

$$h_i = \frac{q}{\vartheta_i} \qquad (2)$$

Where h_i is flow depth (m), q is unit flow discharge (m^2 s^{-1}), and v_i is average surface flow velocity (m s^{-1}).
The Reynolds (Re) and Froude numbers (Fr) were calculated by Eqs. (3) and (4), respectively:

$$\mathrm{Re} = \frac{Vh}{\vartheta} \qquad (3)$$

$$Fr = \frac{V}{\sqrt{gh}} \qquad (4)$$

Where ν is kinematic viscosity (1.01×10^{-6} m^2 $s^{-1)}$ and g is the gravity acceleration (m s^{-2}).
The Manning roughness coefficient (n) was calculated as follow:

$$n = V^{-1} S^{1/2} R^{2/3} \qquad (5)$$

Where n is manning roughness coefficient ($m^{-1/3}$s), V is mean surface flow velocity (ms^{-1}), S is average slope steepness (sine of slope angle), and R is hydraulics radius (m).

Results and Discussion
Water and sediment is trapped by rough surface because it contains many barriers that decrease the flow velocity. Increasing the percentage of rock fragment cover in this study increased the surface roughness and Manning roughness coefficient (Table 1). Poesen et al (1990) and Tailong et al (2010) reported similar results at flume and field experiments, respectively. Increasing rock fragment cover, decreased the Froude number, but had not significant effect on Reynolds number (Table 1). Also increasing the rate of flow discharge, decreased the Froude number and increased the Reynolds number.

Table. 1 The flow hydraulic properties at different rock fragment cover and flow discharge

Flow discharge (Lit min^{-1})	Rock fragment cover (%)	Average flow depth (mm)	Flow velocity (cm s^{-1})	Reynolds number	Froude number	Manning roughn. coef. ($m^{-1/3}$ s)
	0	0.6	16.1	83.4	2.24	0.012
3	10	1.2	8.3	82.6	0.84	0.033
	20	1.5	6.4	79.6	0.58	0.049
	30	2	5	82.9	0.39	0.074
	0	1.2	16.4	162.5	1.66	0.017
6	10	2.2	9.1	163.5	0.68	0.042
	20	3	6.7	165.8	0.43	0.071
	30	3.8	5.3	166.3	0.3	0.104
	0	1.5	19.6	253.2	1.73	0.016
9	10	2.9	10.4	256.8	0.66	0.046
	20	4.2	7.2	247.9	0.39	0.081
	30	5.3	5.7	241.6	0.28	0.115

Distance to rill formation values are presented in Figure 1. The rills developed when soil critical shear stress was exceeded the surface roughness induced shear resistance. Figure 1 shows that distance to rill initiation increased with increasing rock fragment cover and decreased with increasing flow discharge rate. These results are similar to those reported by Renard et al (1997) which assumed that rill erosion is insignificant in slope lengths shorter than 4.5 m at a field.

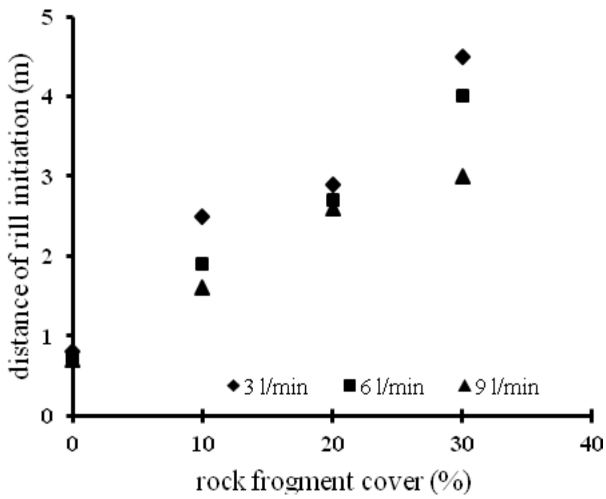

Fig. 1 Relationship between distance to rill initiation and rock fragment cover.

Figure 1 shows quantitatively that the changes in distance to rill initiation were different for varied rock fragment percents and flow discharge rates, and that the changes due to the rock fragments were more than that of the flow discharge rates, within the ranges of this experiment. This can be more clearly demonstrated using the relative change rate, defined as:

$$r = \frac{\Delta l_i}{l_0} \times 100 \qquad (6)$$

Where, r is the percent of distance change to rill formation, Δl_i is the change of rill initiation distance because of rock fragment and flow discharge ranges (0% to 30% rock fragment covers in a determined flow discharge rate, or 3 to 9 Lit. min^{-1} flow discharge rates in a determined rock fragment cover), and L_0 is minimum value (belong to distance to rill formation of 0% Rock fragment cover in different flow discharge rates, or 3Lit. min^{-1} flow discharge rate in different rock fragment covers). The results of this sensitivity analysis are given in table 2. The distance to rill formation has a greater sensitivity to rock fragment covers relative to flow discharge rates, so the effect of rock fragment covers on distance to rill initiation (L_i) is more significant than that of flow discharge rates.

Table. 2 Distance to rill initiation at different rock fragment covers and flow discharge rates

Factor	Rock fragment cover Change (0% to 30%)			Flow Discharge change (3 to 9 l min^{-1})			
Level	3	6	9	0	10	20	30
Li change range	0.8 to 4.5	0.74 to 4	0.7 to 3	0.7 to 0.8	1.6 to 2.5	2.6 to 2.9	3 to 4.5
r change rate (%)	462.5	440.5	328.5	12.5	36	10.3	33.3
r average change rate	410.5%			23.1%			

Conclusion

The general results of this investigation show that flow discharge rate and rock fragment cover percent, both affect the distance to rill initiation, but the effect of rock fragment is more significant. Increased rock fragment cover decreases runoff velocity, because of increasing the soil surface roughness. The rill initiation was retarded with increasing the rock fragment cover, because of flow velocity and erosivity power.

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Estimate of Scaling Parameter in Arya-Paris Equation to Achieve the Soil Water Retention Curve

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Abstract: Arya-Paris method is one of the indirect methods which use particle size distribution curve data to estimate the soil water retention curve. Regarding the equations in this model, scaling parameter, α, is used in order to estimate the average size of pore volume radius. The value of this parameter is considered either constant or obtained from linear and logistic methods in various soil textures. In this research, 58 soil samples were used to evaluate this parameter. Using measured soil water retention curves, particle size distribution curves and inverse solving methods, the average values of α was obtained by 1.49, 1.42, 1.37 and 1.32 for sandy, loamy sand, sandy loam and silt loam soil texture, respectively. These values were compared to the calculated values from both linear and logistic methods. Results show that, logistic method is appropriate for sandy, loamy sand, and sandy loam textures while linear equation has a better approximation only for silt loam texture.

Keywords: Scaling Parameter, Soil Retention Curve, Particle Size Distribution

Introduction

Survey Behavior of unsaturated soils is required to determine the soil water characteristics curve. This curve shows the relationship between soil suction and moisture content in it. Soil moisture condition and then the amount of plant available water can be realized in the field with determine this curve. As regards, laboratory estimates of soil moisture curve is very time consuming and costly, thus much effort has been to be achieved easier and less costly methods with mathematically and physically basis to predict this curve. there is an increasing interest for models that estimate this property from simple taxonomic data (texture or complete particle size distribution, bulk density, particle density, organic **as** matter) and other basic properties (Arya et al., 1999; Pachepsky and Rawls, 1999). Particle-size distribution data have been widely used as a basis for estimating soil hydraulic properties. Significant contributions were made by Arya and Paris (1981) to predict h (θ) curves using the PSD. Their physic empirical approach is based mainly on the similarity between shapes of the cumulative PSD and h (θ) curves. As a physico-empirical model, the AP procedure contains an empirical parameter, α, used to estimate pore radius (r_i) from particle radius (R_i). Arya and Paris (1981) assumed that r_i is determined by scaling pore length, calculated from the packing of spherical particles of size Ri to natural pore length using the scaling factor α. Originally, arya and paris (1981) introduced α as constant (α=1.38) and the model proved to work relatively well for sand soils. Later investigations by Arya et al. (1982) showed that the average a varied among textural classes and ranged in value from 1.1 for finer textures to 2.5 for coarse textured materials. A similar range of values was reported by Tyler et al. (1989) for the fractal dimension. Yoshida et al. (1985) also reported higher values of α for coarse-textured materials. Later, a value of α= 0.938 was proposed by Arya and Dierolf (1992), but it did not affect the results in any substantial way (Basile and D'Urso, 1997). However, Schuh et al. (1988) have shown the varyation of α for a wide range of soil matric potentials and Basile and D'Urso (1997) derived an expression for α as a function of the soil matric potential (α=f(ψ)).
The value of this parameter is considered either constant or obtained from linear and logistic methods in various soil textures. In this research, 58 soil samples were used to evaluate and verifying usual methods that estimate this empirical parameter. Using measured soil water retention curves, particle size distribution curves and inverse solving methods, the average values of α were

obtained from inverse solution and compared with values calculated from linear and logistic methods.

Materials and Methods

Field data

in this paper, we used The measured data from 30 sites that were studied. The sites were scattered over about 400 km^2 area surrounding the Hyytiälä Forestry Field Station of the University of Helsinki in central Finland (61º48' N, 24º19' E). The soil samples were taken during the summer 1987 and 1988. The forest site types were Calluna type (CT), Vaccinium type (VT), Myrtillus type (MT) and Oxalis-Myrtillus type (OMT) according to the Finnish forest site type classification (Cajander 1926). A total of seven CT sites, seven VT sites, eleven MT sites and five OMT sites were selected for soil sampling. In each stand a 10 m * 30 m rectangular area was chosen and three soil pits dug at each. A single sample (150 cm3 cylinder, 5.8 cm diameter and 5.7 cm height) was taken. Altogether 360 mineral soil samples were collected. 360 WRCs were measured and PSDC was available from 108 samples. Measured particle size distribution curve, bulk density and loss of ignition are shown for 108 soil samples in App. Volumetric water content was computed on the basis of the total volume of the soil. For preparing the water retention characteristic curves water contents had been measured at pressure heads of 0.01, 0.10, 0.32, 0.63, 1.0, 10.0 and 152.0 m. The porosity calculated from the bulk density of the mineral soil samples of this 36 study was in good correspondence with the measured saturated water content (Sahlberg 1992). The classification of the particle size fractions was according to the Finnish system: clay < 2 µm, fine silt 2-6 µm, coarse silt 6 – 20 µm, fine sand 20 – 60 µm, coarse fine sand 60 – 200 µm, medium sand 200 – 600 µm and coarse sand 600 – 2000 µm. in this research we used 58 soil samples and then they were classified into four Categories based on soil texture (table 1).

Table 1: Average values of some physical parameters of soils in each category

Soil texture	**void ratio** ($m^3/m^{3)}$)	**Bulk density** (gr/cm^3)	**sand** (%)	**silt** (%)	**clay** (%)
sand	0.99	1.36	90.78	6.43	2.75
Loamy sand	0.94	1.37	79.11	16.34	4.54
Sandy loam	1.12	1.26	62.83	31.32	5.81
Silty loam	1.09	1.29	35.30	59.78	4.9

Arya and Paris Model

In the following we will review pertinent aspects of the model. The particle-size distribution curve is divided into n size fractions, and the solid mass in each fraction is assembled to form a hypothetical, cubic close-packed structure consisting of uniform size spherical particles. The pore volume in each assemblage is calculated from the bulk density and particle density measured on the natural structure soil.

The pore volume, V_{pi} (cm^3. g^{-1}), associated with the solid mass in the ith particle-size fraction, is represented as a single cylindrical capillary tube, and is given by equation (1):

$$V_{pi} = (\frac{W_i}{p_s})e = \pi r_i^2 l_i \qquad (1)$$

where w, is the fraction solid mass ($g.g^{-1}$), ps is the particle density (g. cm^{-3}), e is the void ratio, r_i is the pore radius (cm), and l_i, is the pore length (cm. g^{-1}). The void ratio, e, is given by equation (2):

$$e = \frac{\rho_p - \rho_s}{\rho_s} \qquad (2)$$

where p_b is the bulk density of the natural soil (g. cm^{-3}). The water content, $\theta_]$, (cm^3. cm^{-3}), is obtained from successive summations of water-filled pore volumes according to equation (3):

$$\theta_i = (\phi . S_w)\sum_{i=0}^{i=1} w_i \qquad (3)$$

where Ø is the total porosity (cm^3. cm^{-3}), and S_w is the ratio of measured saturated water content to theoretical porosity. The number of spherical particles, n_i (g^{-1}), for each fraction of the particle-size distribution is calculated from equation (4):

$$n_i = \frac{3w_i}{4\pi R_i^3 \rho_p} \qquad (4)$$

where R_i is the mean particle radius (cm) for the ith particle size fraction. For an ideal soil consisting of uniform size spherical particles in a cubic close-packed assemblage, l_i, can be arestimated by $l_i = 2n_iR_i$ and the pore radius is related to the particle radius by equation (5):

$$r_i = 0.816 R_i \sqrt{e} \qquad (5)$$

For a natural soil, made up of the same solid mass but with no spherical particles that are arranged randomly, l_i, can be estimated by $l_i = 2n_i^{\alpha} R_i$ and the corresponding pore radius is related to the particle radius by equation (6):

$$r_i = 0.816 R_i \sqrt{e n_i^{1-\alpha i}} \qquad (6)$$

where α is the scaling parameter. Note that $n_i^{(1-\alpha i)}$ is dimensionless. Calculated pore radii, r_i are converted to equivalent pressure heads, h (cm water), using the capillary equation (7):

$$\varphi_i = \frac{2\sigma \cos\theta}{\rho_w g r_i} \qquad (7)$$

where γ is the surface tension at the air-water interface (g.s^{-2}), ϑ is the contact angle, p_w is the density of water (g. cm^{-3}), and g is the acceleration due to gravity (cm. s^{-2}). The model assumes perfect wet ability and, hence, $\theta = 0$.

Procedure to Derive α

The scaling factor is obtained using measured soil water retention curves, particle size distribution curves and inverse solving method. pore radius is is calculated from equation (8):

$$r_i = 1 - \frac{\log\left[\frac{3}{2e}\left(\frac{2\gamma}{\rho_w \psi_i g R_i}\right)^2\right]}{\log(n_i)} \qquad (8)$$

Through the adjustment of measured soil water retention data to the model for 58 soil samples using inverse solution method and compared with the recommended values provided by Arya and Paris for the soil textures that used in this research. Also, the scaling factor values were calculated using Linear and Logistic methods. Arya et al. (1999) explored formulations for a based on a linear relationship between log N, and log (wi/R^3_i). The generalized form of this relationship is:

$$\alpha_i = \frac{a + b \log \frac{W_i}{R_i^3}}{\log n_i} \tag{9}$$

The relationship between log N_i and log n_i closely follows the logistic growth equation (Thornley, 1990):

$$Y + \Delta Y = \frac{Y_f\, Y_{in}}{Y_{in} + \left(Y_f - Y_{in}\right)\exp\left\{-\mu\left(X + \Delta X\right)\right\}} \tag{10}$$

Required empirical coefficients for these methods are given in table 2. Finally, for each category of soil samples, we calculated soil water retention curve with α values calculated by different methods and then results of each method were evaluated with standard error parameter by equation (11):

$$SE = \sqrt{\frac{1}{n_p - 1} \sum \left(\theta_m - \theta_p\right)^2} \tag{11}$$

Table 2: Empirical coefficients of linear and logistic methods for each soil texture category

Soil texture	**Linear method**		**Logistic method**				
	a	**b**	**$(\log Ni)_{in}$**	**$(\log Ni)_f$**	**μ**	**$\Delta \log N_i$**	**Δlogni**
sand	-2.748	1.49	0.966	16.602	0.609	1.734	0.00032
Loamy sand	-3.073	1.631	0.777	16.792	0.581	2.113	0.924
Sandy loam	-3.398	1.773	0.559	16.983	0.553	2.492	1.849
Silt loam	-2.48	1.353	0.719	19.686	0.457	1.902	0.684

Results and Discussion

Values of scaling parameters recommended by Arya and paris, calculated from linear, logistic and inverse solution methods for all soil textures categories in this research are presented in the table 3. In the sand and loamy sand textures, α values obtained from the linear method are closer than other methods to values obtained from inverse solution method. Also In the sandy loam and silt loam textures, α values obtained from the logistic method are closer than other methods to values obtained from inverse solution method. Recommended values of α by Arya and Paris in the soils with loamy sand and sandy loam textures are close to values obtained from inverse solution method. But in the other soil textures there were significant difference between recommended values of α and values calculated from inverse solution. Results obtained by the method of inverse solution showed, constant α hypothesis (α =1.38) in the soils with loam texture is acceptable.
The next step, soil water retention curve were obtained with Arya and Paris method using different scaling parameters that calculated from methods mentioned above and were compared with actual soil water retention curves.

Table 3: Values of scaling parameters obtained by different methods

Soil texture	**Scaling parameter(α)**			
	Linear method	**Logistic method**	**α (constant)**	**Inverse solution**
sand	1.292	1.488	1.285	1.531
Loamy sand	1.463	1.394	1.380	1.442
Sandy loam	1.456	1.422	1.459	1.374
Silt loam	1.193	1.153	1.150	1.295

Figure 1 show soil water retention curves obtained from different methods of calculating α compared with actual curves. The curves obtained from inverse solution method must be closer than other methods to actual curves. Because these values were calculated from actual soil water retention curve data and these are the optimal values of α. In addition, for the same values of soil volumetric moisture, values of suction were calculated from different method of calculating α and then compared with actual values and Standard error values were calculated for them. These values are presented in table 4. Results show that, in sandy soil textures linear and constant methods had the similar and better than logistic method. In the loamy sand soil textures, constant method had the best results and logistic method has been worst. In the sandy loam, constant method had the highest accuracy but linear and logistic methods are similar and had the lowest. In the silt loam soil textures, constant and linear methods had similar accuracy and better than logistic method.

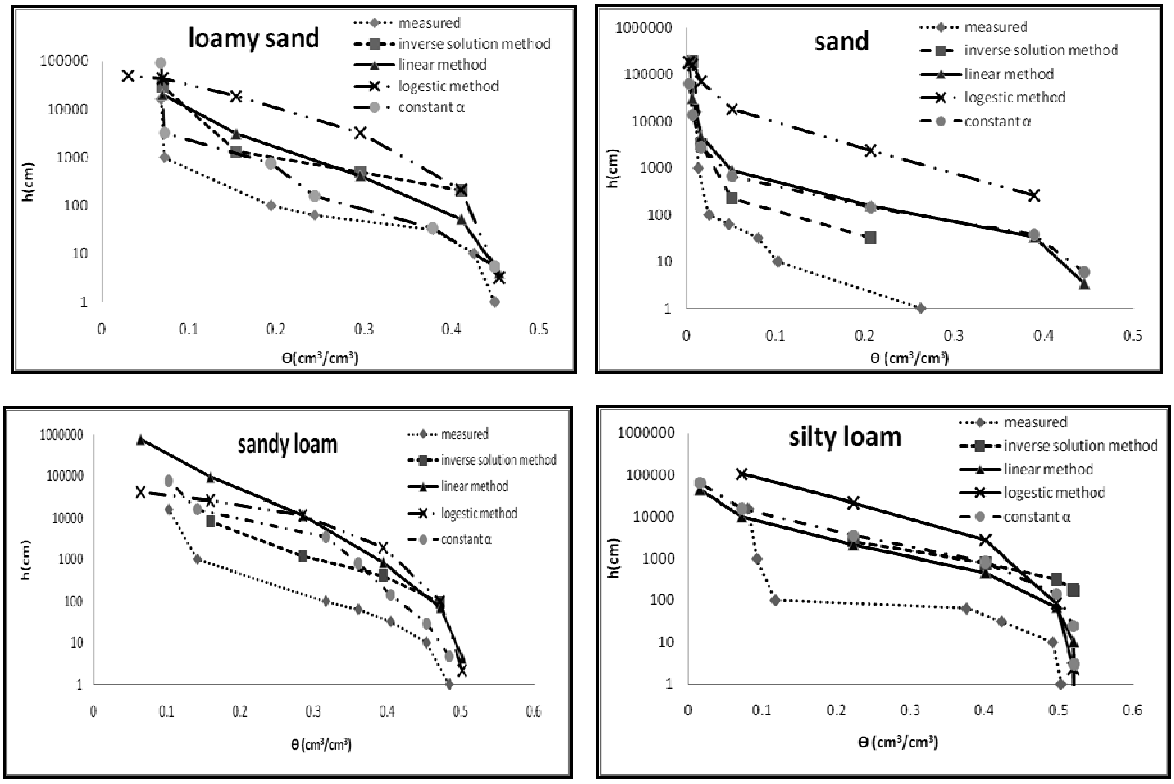

Figure 1: soil water retention curves obtained from different methods compared with actual curves

Table 4: Standard error values calculated in the estimation of soil suction

Soil texture	Estimated methods (α)			
	constant	linear	logistic	Inverse solution
sand	0.28	0.22	0.40	0.17
Loamy sand	0.16	0.11	0.18	0.10
Sandy loam	0.10	0.14	0.16	0.20
Silt loam	0.23	0.17	0.23	0.14

Finally, we can be stated constant and linear methods had acceptable results and Logistic method had the lowest accuracy in estimate of scaling parameter. As a result, Arya and Paris method has good accuracy in estimate of soil water retention curve in soils with texture close to loam compared to other soil textures. Survey the actual values of scaling parameter in the fine and coarse textures and relationship between soil physical characteristics with this empirical parameter recommended.

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Residual Effects of Boron Applied to Sugar Beets on Wheat

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Konya Toprak, Su ve Çölleşme ile Mücadele Araştırma İstasyonu Müdürlüğü
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Abstract
This study was carried out determining boron requirement of sugar beet, resistant to high boron toxicity and susceptible to boron deficiency, and to investigate the residual effect of boron following crop, wheat. Although the yield of sugar beet increased by boron rates in soils where boron level was 0.5 ppm, boron application did not increase yield in soils where boron level was 1 ppm or higher. Regression that between boron content in soil and yield of sugar beet is different significantly. The yield of bread wheat and durum wheat were affected significantly by boron rates applied to sugar beet. It was shown that boron applied to sugar beet was also sufficient for the wheat. Furthermore, increased boron level decreased sterile flower number in wheat. After harvesting sugar beet soil samples taken applied boron plots showed according increasing boron doses level, soil boron content increased
Key words: Boron, Sugar beet, Wheat, soil pollution, residual effect

Introduction
Boron is one of the vital nutrition elements for plant growth. The useful amount of boron that is required in the soil changes depending on the type of plants. Sugar beet which is the topic of this study is among the boron-resistant, wheat on the other hand is among the boron-sensitive. A study investigated the useful boron content useful for plants on farm lands in Middle-south Anatoilan region. The results obtained from the analysis of 898 soil samples indicated that 26.6% of the soil in the region is inadequate for plants like sugar beet which necessary high boron and 18% of the soil is toxic for plants like wheat which is boron-sensitive. Thus in Konya region it is necessary to use fertilizers including boron in the sugar beet farm land with 63 820 ha, which is the second largest area following the wheat production area, to increase yield. This study aimed at determining boron requirement of sugar beet and investigating the residual effect of boron following crop, wheat.

Material and Method
The experiments were carried out in two different regions, Konya-Karaaslan and Çumra. Field experiments were conducted in a completely randomized block design for two years at same area Depending on experiment topics the determined boron amounts were applied to sugar beet and planting was carried out. After the sugar beet harvest, bread wheat was planted in the same plot with no boron application. In the research borax ($Na_2B_4O_7 10H_2O$) was used as boron source. The fertilizer application was conducted in accordance with Ülgen and Yurtsever (1995).The analysis mentioned below for the soil samples of the experimental farm lands obtained from 0-20 and 20-40 cm. under the surface before fertilizer application was determined according to Tüzüner (1990). Besides boron useful for plants were determined as Kacar (1998) mentioned. By applying variance analysis to the yield obtained from the experiments the difference among boron applications was controlled. The equation $Y=a+bx-cx^2$ was used to determine the relationship between boron quantity and yield (Yurtsever,1984).

Results and Discussion
The effects of boron applications on sugar beet tuber yield
Average sugar beet yield obtained from experiments by applying boron to soils with different boron contents is presented in Table 1. As can be seen in the table, boron applications in the fields where boron content is low (0.26 ppm – 0.42 ppm) caused considerable increase in the sugar beet tuber yield. Every four experiments conducted in Konya, 2.0 kg/ha boron application caused statistically significant increase in the sugar beet tuber yield. Although higher amount of boron applications caused increase in the yield, they were not found to be statistically significant. In the experiments in Çumra in which the boron content is 1.14 ppm boron application did not increase the sugar beet tuber yield.

Table 1 Sugar beet tuber yield (kg/ha)

Deneme konusu (kg/ha B)	experiment location year boron content				
	Konya 2001 (0.26 ppm)	Konya 2002 (0.42ppm)	Çumra 2002 (1.14 ppm)	Konya 2009 (0.40 ppm)	Konya2010 (0.30 ppm)
0.0	54986	66274	75139	51793	62847
2.0	59626*	72576*	75762	54111*	68750
4.0	60319	76593	74377	55091	69792
6.0	61773	77632	72992	55120	69931
8.0	61080	77978	75623	53986	70694

*there are significantly difference to control

In the experiments where boron content varies between 0.26 ppm and 0.42 ppm the boron application above 2.0 kg/ha increased sugar beet tuber yield, although it was not statistically significant. In this case it is not correct to claim that 2.0 kg/ha is enough to obtain maximum yield. To determine the amount of boron to be applied more accurately, it is necessary to determine the relationship between sugar beet tuber yield and amount of boron applied. To do this, regression analysis was used over average yield obtained from four experiments in which boron content is low and it is suggested that the relationship between sugar beet tuber yield and amount of boron applied could be expressed by using $Y=59228+2378.7x-195.66x^2$ (R=0.9921equation. According to this equation in the fields in Konya region where boron content varies between 0.26ppm and 0.42 ppm, boron application with 6.1 kg/ha calculation is enough. The higher amount of boron application did not cause any extra increase in sugar beet tuber yield,

The effects of boron application on soil boron content

After the sugar beet harvest before wheat was planted the boron content of the soil samples obtained from 0-20 and 20-40 cm under the surface from each parcels was presented in Table 2.

Table 2 The average boron content of experimental plot after sugar beet harvest (ppm)

Boron Applied (kg/ha)	Depth of Soil (cm)	Boron Content After Harvest		
		Konya 2001	Konya 2002	Çumra 2002
0.0	0-20	0.26	0.53	1.16
	20-40	0.22	0.52	1.15
2.0	0-20	0.38	0.82	1.30
	20-40	0.39	0.79	1.26
4.0	0-20	0.55	1.10	1.46
	20-40	0.66	1.00	1.30
6.0	0-20	1.11	1.31	1.70
	20-40	0.79	1.03	1.69
8.0	0-20	1.13	1.71	2.14
	20-40	0.97	1.43	2.03

As can be seen in the table, depending on boron application, the boron contents in the experimental fields increased. Although the boron content in the upper layer of soil is higher than the one in the lower layer, no considerable difference was observed. This data shows that some amount of boron leaching to deep

The residual effect of boron applied on sugar beet on wheat

In order to determine the residual effect of boron applied on sugar beet on wheat, bread wheat was planted in experimental blocks where sugar beet was planted. The obtained grain yield is given in Table 3.

Table 3 The grain yield at different boron levels (kg/ha)

Treatment	Experiment Location Boron Content			
	Konya 2002 (0.26 ppm)	Konya 2003 (0.42ppm)	Konya 2010 (0.40 ppm)	Konya2011 (0.30 ppm)
0.0	4058	3610	3236	5750
2.0	4479*	3756	3753*	6450*
4.0	4771	3733	3778	6467
6.0	4825	3802	3830	6550
8.0	4829	3565	3693	6433

*there are significantly difference to control

As can be seen in Table 3 the residual effect of boron applied on sugar beet increased wheat grain yield. To determine the relationship between the amount of residual boron and yield of wheat variety regression analysis was used. It is suggested that the relationship between the amount of residual boron and yield of wheat could be expressed by using $Y=4190+217.7x-20.50x^2$ (R=9829*) equation. According to this equation 6.1 kg boron application to sugar beet is enough to obtain highest yield in wheat which is planted after sugar beet.

The effect of boron applied to sugar beet on wheat sterility

Table 4 summarizes the values obtained from the increasing amount of boron application to sugar beet, which determines the sterility ratio in wheat which was planted after sugar beet

Table.4 The sterility ratio (%) in wheat at different boron levels

(kg/ha B)	Locations and years		
	Konya 2002	Konya 2010	Konya 2011
0.0	5.5	5.8	5.4
2.0	3.5*	3.8*	3.8*
4.0	3.5	2.8	1.6*
6.0	3.2	3.4	2.2
8.0	3.0	3.8	2.6

* there are significantly difference to control

The table indicates that depending on the increasing amount of boron application to sugar beet, sterility ratio decreases in wheat which was planted after sugar beet.

Discussion

This study indicated that in fields in Konya region with 0.26-0.42 ppm or less boron content, boron application incased sugar beet yield significantly; in the fields with boron content above 1 ppm boron application did not incase sugar beet yield. At boron levels applied in experiments conducted in fields with different boron content, toxic signs were not observed in sugar beet. According to the relationship between the amount of boron applied and sugar beet tuber yield, 6.1 kg/ha calculation is enough to obtain maximum yield. The higher amount of boron applications did not increase sugar beet tuber yield, but it can also cause loss in the yield. Thus before boron application to sugar beet, the boron content of soil should be determined and only in soils with low boron content boron fertilizer should be applied. The amount to be applied should not exceed 6.1 kg/ha level.

This amount of boron applied to the sugar beet is enough to provide the boron need of wheat which is planted after sugar beet. In this study the residual effect of boron applied to sugar beet was investigated in only a trial with low boron content. As it is known sugar beet is resistant to high boron. Hence it may not be affected negatively as much as boron sensitive plants from high boron applications. Consequently in Çumra experiment whose boron content is higher than 1.1 ppm, boron application did not cause any increase in yield, but it did not show any clear toxic effect. It should be kept in mind that the unnecessary and careless application of boron which is more than necessary may accumulate in soil and cause toxic effects in semi-dry climate conditions especially in regions like Konya. In this study after sugar beet in 0-20 cm depth of control plot, boron contents which were 0.26 ppm, 0.53 ppm, and 1.16 ppm increased 1.13 ppm 1.71 ppm, and 2.14 ppm levels respectively after boron application with 8.0 kg/ha. Although reliable results could not be obtained from wheat experiments conducted in Çumra and they were not included in analysis, the boron content which is over 2.0 pmm is not desirable for wheat production (Ayers and Westcot, 1976).

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Effect of Natural and Modified Montmorillonite on Mobility of Cd in Soils

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Abstract
Mobility of Cd in a sandy soil and a clay loam soil amended with 10% natural or modified montmorillonite were investigated in a batch experiment. Modification of montmorillonite was carried out by direct polymerization of polyacrylamide in suspensions of montmorillonite. The sorption of Cd on the amended soils was studied as a function of the metal concentrations in the range of 1.5-10 mgL^{-1} using a 24h batch equilibration experiments. Then different sorption isotherms including Freundlich, Langmuir and Koble-Corrigan were fitted to the data. The results of equilibrium test (isotherms) revealed that removal of Cd from solutions was affected by the amendments for the both soils. The Koble-Corrigan isotherms was described satisfactorily equilibrium data. The values of Freundlich (K_F) and Langmuir (K_L) bonding constants were greater in soils containing modified montmorillonite as compared to soils containing the natural ones. Maximum sorption capacity calculated from the Langmuir model for the sandy soil amended with natural and modified montmorillonite were 0.124 and 0.251 mgg^{-1}, and the corresponding values for the clay loam soil were 0.651 and 0.715 $mg.g^{-1}$, respectively. Based on the results, modified montmorillonite can be considered as a Cd immobilization agent in the polluted soils.
Keywords: Cd, montmorillonite, polyacrylamide, removal, sorption.

The Influence of Horizontal Drain Pipe Spacingon Quantity of Leached Nitrogen from Soil Type Pseudogley

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Abstract
The research was conducted on the experimental drainage field Varna of the Institute of Soil Science, Belgrade, in the village near Šabac (44°41'38" N;19°39'10" E), formed on the soil type pseudogley, with installed horizontal drain pipes with spacing of 20,25 and 30 m.
The experimental crop was wheat variety Renaissance, grown using standard agro-technical measures.
The research was conducted during 2008 and 2009, by taking samples of drain water in tree replicates, from all tree variants of drainage experiment and from well, located in experimental field, isolated from the influence of applied agro-technical measures.
From collected samples of water it was determinate the quantity of nitrites nitrogen (NO_2 – N), nitrate nitrogen (NO_3-N), and ammonia nitrogen (NH_4-N).
Collected results indicate that the nitrogen in drainage and groundwater mainly is present in the nitrate nitrogen form. Highest average concentration of nitrate nitrogen form registered in drainage water was 43 mg NO_3-N/l in variant with drain spacing of 20 m, while the concentration of nitrate nitrogen form was in range up to 0,5mg NO_3-N / l.
The processed results show that the level and distribution of rainfall including the type and amount of fertilizer applied have a dominant role in the amount of nitrogen leached in the drainage water.
Key words: drainage and groundwater, nitrites, nitrate, and ammonia nitrogen.

Introduction

Presented results have intention to present up to which the extent of the spacing of installed horizontal drainage pipes on soil type pseudogley -stagnosol (WRB, 2006), affects the amount of leached nitrogen by cultivating crops; wheat and corn. Examinations were conducted in the area of village Varna near Sabac in Mačva, where the experimental drainage field of Institute of Soil is located.
Of the total amount of nitrogen fertilizers, which are entered into the soil, the plant adopts about 50%, 25% is lost by flushing and 15% is lost due to de-nitrification processes (Azem et al. 1985; Klačić et al., 1998; Čoga et al., 2003). Part of the nitrogen that is washes out in the form of nitrate can affect the eutrophication and pollution of watercourses.
The amount of nitrogen that will be washed out with drainage waters from agricultural areas depends on many factors. The most important factors are: the type of applied fertilizer, water physical characteristics of soil, species of crops, climate characteristics of the region, especially the rainfall, and if the horizontal pipe drainage is applied, also on drainage spacing.

Materials and Methods

The investigation was conducted at experimental drainage field of the Institute of soil science, Varna, (44°41′38″N ; 19°39′10″E) located on the tenth kilometer road which leads from Šabac, southeast to Loznica and Valjevo, at the entrance to the village Varna (Pivić R., 2005), established on pseudogley soil, which is characterized by adverse air and water physical and chemical characteristics in the period from September 2008 to October 2009.

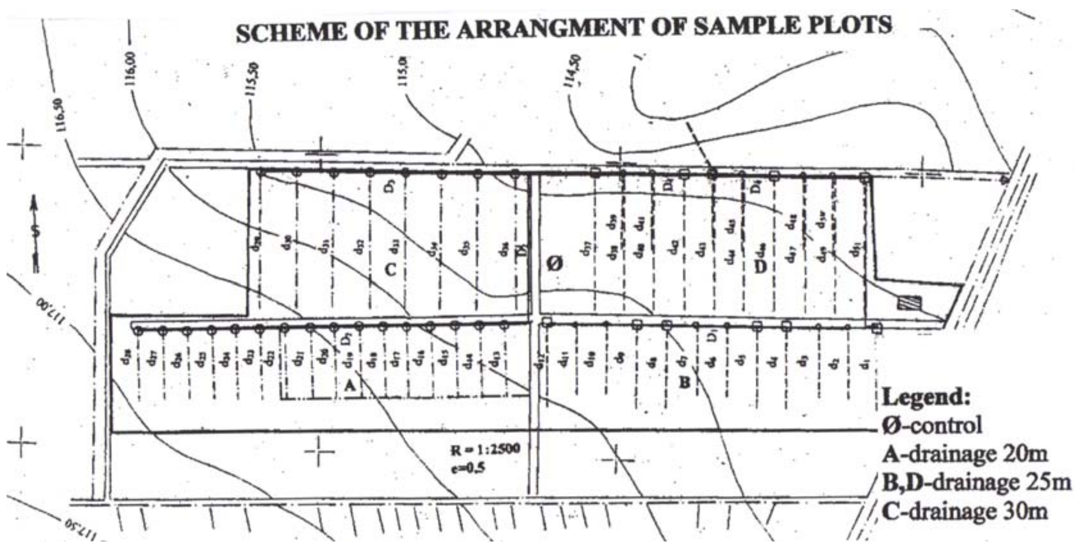


Figure 1.- Scheme of the arrangement of sample plots Varna

In 1978 the main project of the drainage of a part of sample plot was constructed (Pivic, 2005). Drainage sample plot consists of two separate parts of rectangular form, separated by a road for mechanization. One part consists of three plots: A,B,C, and the other part consists of six plots designated and to VI. All the plots are of the same size 75.0x52.0m, individual area 0.39ha. The basis for dewatering of the nine sample plots is flexible perforated PVC drainage pipes, spacing 25 metres. Within the plots there are two drains ∅80mm, at the depth of 0.95m. Drain length is equal to plot length and amounts to 52m, minimal design slope is 0.25%. The experiment was amended in 2002 by adding two additional variants of drain spacing treatments: 20m (field A) and 30m (field C), at the same depth of 90 cm, and perforated PVC pipes Ø80mm.

Monitoring of the main climatic parameters (precipitation, air temperature, relative humidity, insolation, cloudiness) in the trail period (1964-2009) was carried out at the nearest meteorological stations: Sabac, Varna, Loznica and Valjevo.

Water-physical and chemical condition of soil is obtained by standard laboratorial methods in the laboratory of the Institute of Soil Science of Belgrade.

During experiments it was applied a standard agricultural practices. Test crop was wheat variety "Renaissance" seeded on 20th October with the previous crop - corn. With crop residues it was plowed 250kg/ha NPK 15:15:15 and during the growing season fertilization was carried out in February with 90kg/ha NPK 15:15:15 and in April with 120kg/ha KAN.

Since the automatic equipment for the measurement of drainage leakage was not installed, measurements were performed manually. Drainage leakage was registered using volumetric method, whenever, it was shown and presented in observation series.

In the period of appearance of the drain leakage, the average sample was collected from all three variants of drainage spacing (20, 25 and 30m) and also from the well within the experimental field. Observation points are presented in Table 1.

Table 1. Points of observation

Plot	Drainage distance(m)	Drain outflow		
A	20	d_{25}	d_{26}	d_{27}
B	25	d_3	d_7	d_9
C	30	d_{32}	d_{33}	d_{34}

For collecting and transporting of water samples, which were preserved by using toluol, it was used plastic bottles, previously washed with water. Bottles were transported in refrigerators to the laboratory.

The collected samples were analyzed for their content of ammonia nitrogen (NH_4-N), using distillation method and Devard's alloy (Jakovljević M. et al., 1985), nitrate nitrogen (NO_3-N) and nitrites nitrogen (NO_2 – N) using Lachat Quik Chem 8500 (USEPA, 1983).
The criteria for classification of the water samples depending on the content of the mineral forms of nitrogen are presented in Table 2.and adopted from the Regulative of European Union (EU), Environmental Protection Agency (EPA, SAD), and World Health Organization (WHO).

Table 2. Criteria for classification of the water samples

Parameter	EU (mg l^{-1})	EPA, SAD (mg l^{-1})	WHO (mg l^{-1})
NO_3-N	50.0	45.0	50.0
NO_2-N	0.2	-	0.2
NH_4-N	1.0	1.0	1.0

Results and Discussion
The values of the main climate elements are presented in the Table 1.
Average annual precipitation for study locality is 703mm, it has mainly a uniform distribution throughout the year per months, except in June and July when somewhat higher values of this climatic element were recorded. Precipitation frequency is expressed through high-intensity precipitation in short periods, which is, together with mean annual air temperature 11.5 ^oC, the main characteristic of the climate in this region.

Table 3. Average monthly value of major climatic indicators in the 1964 -2009 period

	I	II	III	IV	V	VI	VII	VIII	IX	X	XI	XII	$\bar{x}$
Air temperature (^oC)	0.1	2.5	6.8	11.8	17.2	20.3	22.0	21.4	16.9	11.8	6.1	1.7	**11.5**
Relative air humidity (%)	87.0	84.0	77.0	74.0	74.0	77.0	76.0	77.0	81.0	84.0	86.0	89.0	**80.0**
Precipitation (mm)	44.5	50.1	54.4	58.7	64.1	86.8	75.0	59.7	57.8	59.4	63.7	62.6	**61.4**
Insolation (%)	65.1	91.7	143.3	177.7	231.6	252.0	298.8	269.9	199.5	151.3	82.9	53.2	**168.1**
Cloudiness (0-10)	7.2	6.7	6.2	6.2	5.7	5.3	4.2	4.1	5.1	5.6	6.9	7.5	**5.9**

Table 4 presents distribution and amount of precipitation during the study.

Table 4. Distribution and amount of precipitation

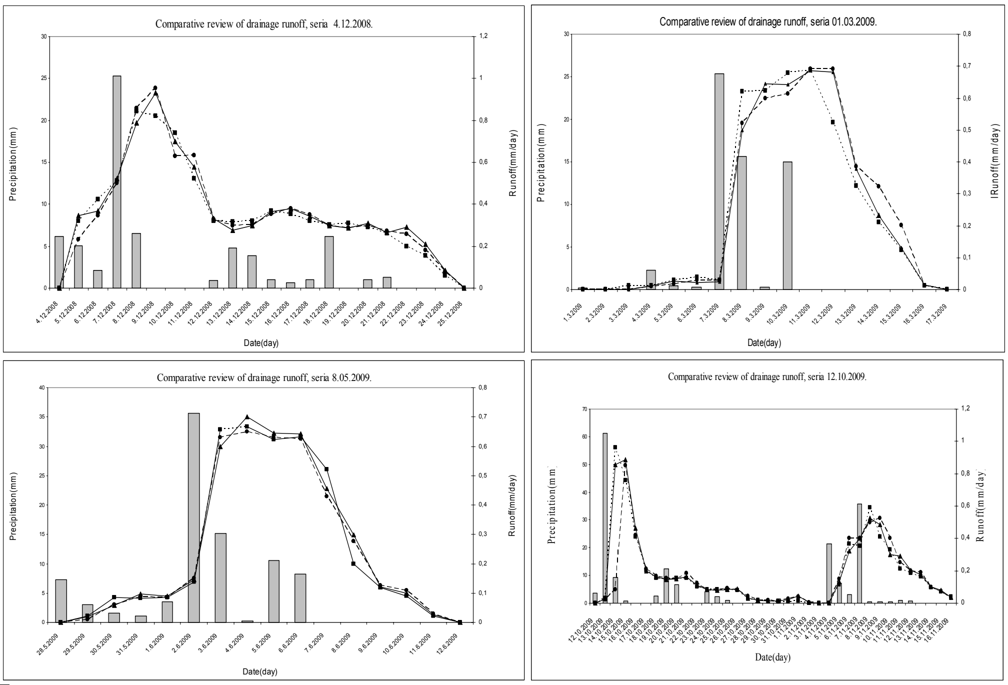
Year	**2008**			**2009**										**Sum**
Month	**X**	**XI**	**XII**	**I**	**II**	**III**	**IV**	**V**	**VI**	**VII**	**VIII**	**IX**	**X**	
Precipitation (mm)	19,9	69,7	58,1	76,1	47,1	104,5	16,5	18,2	194,9	60,6	70,9	4,2	124	864,7

Major soil physical and chemical properties are given in Table 5.

Table 5. Major soil physical and chemical properties

Depth (cm)	**Content of particles (%)**		**Porosity (%)**	**Capacity (%)**		**pH**		**Humus (%)**
	Silt	**Clay**		**Water**	**Air**	**H_2O**	**1M KCl**	
0-27	38,7	26,5	43,39	43,20	4,33	5,5	4,2	2,21
27-45	36,3	26,2	41,30	41,66	0,34	5,3	4,1	1,25
45-75	33,6	34,6	40,07	40,30	0,42	5,7	4,5	1,00
75-120	31,3	39,4	40,28	40,10	0,36	5,6	4,4	0,70

Soil of experimental drainage field Varna, can be based on granulometric composition belongs to the group of light clay. Participation of sand fraction is quite high for this type of soil, and its content decreases with depth. Participation of colloidal clay is slightly lower from the surface to the depth of 45 cm and then it is increasing to the 120 cm. The content of clay fraction and the mineralogical composition of the clay determined many characteristics of this type of soil.
Disposition of the colloidal clay particles clearly indicates flushing of colloidal particles and their transition to the Btg horizon, which is one of the main characteristics of the tested soil. Closely related to the clay and silt contents are the total soil porosity, water and air capacity. pH increases with soil depth. As regards the humus content, the soil is only slightly humous in the surface horizon and poor in humus horizon in the subsurface. The amount of nitrogen leached is directly proportional to the amount of drainage leakage. The highest amounts were registered on the variant of drainage spacing of 20 meters.
Figure 2 is presenting precipitation and drainage leakage during the period of monitoring on variants of drainage spacing



▌-Precipitation(mm); ■-drainage 20m; ▲-drainage 25m; ●-drainage 30 m

Figure 2.- Comparative review of drainage outflow

Collected results indicate that the nitrogen in drainage and groundwater mainly is present in the nitrate nitrogen form. Highest average concentration of nitrate nitrogen form registered in drainage water was 43 mg NO_3-N/l in variant with drain spacing of 20 m, while the concentration of nitrate nitrogen form was in range up to 0,5mg NO_3-N / l.
Collected data correspond to the statements of Skaggs and Gill, 1981; Schuch and Jordan, 1985, Soskic et al., 1987, Klačića et al., 1998, Čoga et al., 2003 indicating that one of the important factors for flushing of nitrate in drained soil is drain spacing. Nitrogen in drainage and in well water is present mainly in the nitrate form.
Figure 3. is representing the concentration of nitrate, ammonium and nitrite nitrogen by a series of monitoring in relation of the drain spacing and the content of the tested parameters in the well (groundwater). The content of the parameters that were followed did not deviate significantly from the legally permissible concentration.

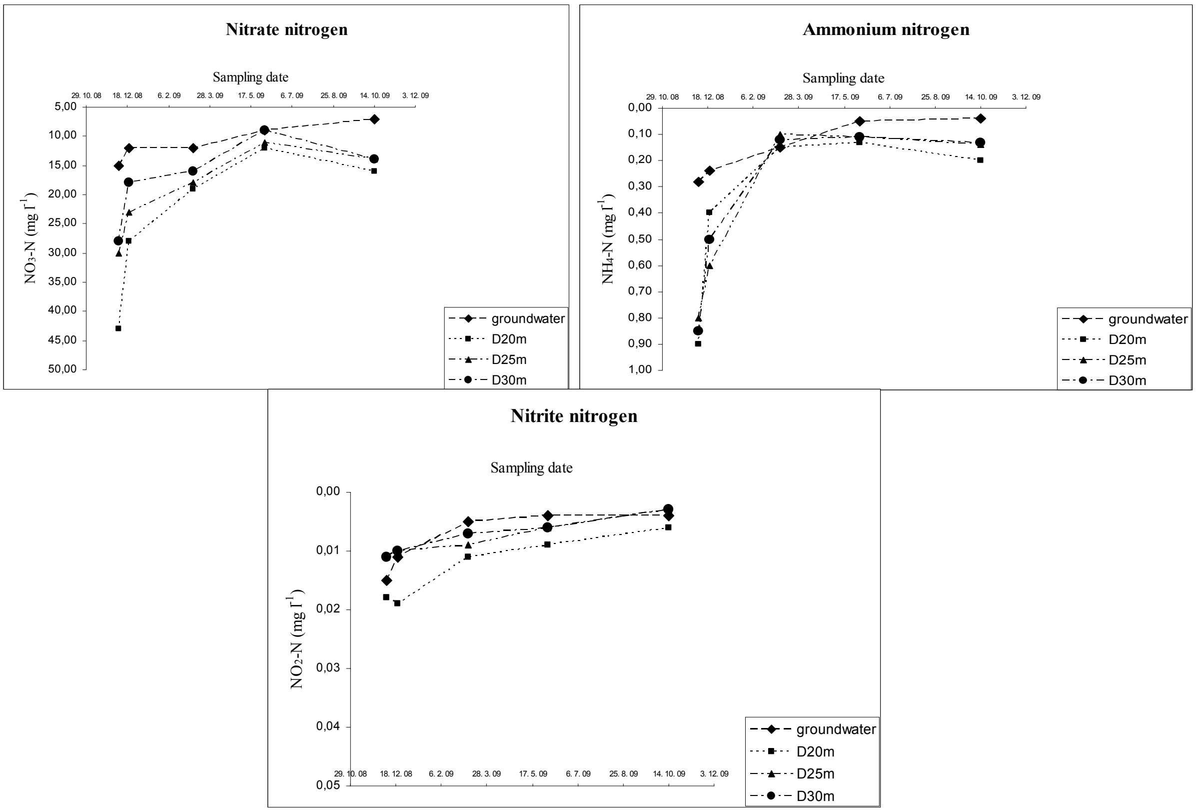


Figure 3. Concentration of nitrate, ammonium and nitrite nitrogen by a series of monitoring

The concentration of nitrate and ammonium nitrogen in drainage and ground water varied depending on the time of sampling and drain spacing. The highest concentrations are registered in the cold part of the year when the quantity of drain leakage was also highest.
Results of analysis of drainage and draw well waters from tested location, do not detect pollution with mineral semblance of nitrogen, which could be expected as a side effect from agro technical measures of fertilization of soil.

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Effect of soil texture and organic matter on the accuracy of Time domain reflectometry method for estimating soil moisture content

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Abstract: Determination of soil water content using the Time Domain Reflectometry (TDR) method is recently considered as a modern method and is widely used. This method is based on dielectric constant measurement which is affected by the soil characteristics. Therefore, calibration and determination of the relationship between dielectric constant and volumetric soil moisture is required for the practical application of this device. The purpose of this research is the evaluation of the effects of soil texture and organic matter on the accuracy of volumetric water content measured by TDR. For this purpose The experiments were carried out on five types of soil texture (sandy, sandy loam, loam, sandy clay loam and clay loam), and three organic matter levels (2, 3.5 and 5 percent) which contain litter fertilizer in three repetitions under quite random conditions. In order to evaluate and calibrate the results of the TDR system, soil volumetric water content was also measured by direct (weight) method. Also, Estimated Values of soil moisture by the TDR system were compared with Topp et al (1980) and mixed models. The results showed that the volumetric water content values obtained from TDR system, decreased with increasing the percentage of clay and soil organic matter, as TDR system in sand with 2 percent and clay loam textures with 5 percent organic matter respectively, with the percent relative error (RE) 3.97 and 11.79 has been most and least accurate. Comparing the results of topp et al (1980) model and mixed method In all samples tested was indicating good precision in top model. Finally, Relations have been calibrated with a high correlation (R^2) between weight and TDR moisture measurements were presented.

Keyword: Dielectric constant, soil water content, soil texture, TDR,RE.

Introduction

Having knowledge of soil moisture condition is necessary in order to optimize crop yield and flood control. Recently, Time-domain reflectometry (TDR) method has been popular in measuring volumetric soil moisture. TDR became known as a useful method for soil water content and bulk electrical conductivity measurement in the 1980s through the publication of a series of papers by Topp, Dalton and others (Topp et al., 1980; Dalton et al., 1984). Commercial systems became available in the late 1980s and continue to evolve with TDR instruments, probes, and multiplexers (e.g. Evett, 1988) available from a few companies. Smith and Tice (1988) and Dasberg and Hopmans (1992) showed that the K_a/θ_v ratio is also different for fine- textured mineral soils. Few calibration equations have been published for organic soils (Herkelrath et al. 1991, Pepin et al. 1992, Roth et al. 1992, Borner et al. 1996, Shibchurn et al. 2005), but are required if TDR is to be used to monitor changes in the amount of plant available water in the forest floor, the layer in which much of the fine root biomass is located (Pietikainen et al. 1999). Schaap et al. (1996) calibrated TDR in woodland areas. In their study, using a regression between soil moisture and refraction index, they observed that decomposition of soil organic maters has a significant impact on measured values of soil moisture and reflected time from TDR. Jacobsen and Schjonning (1993) in a laboratory study, analyzed the effect of soil texture and bulk density (ρb) on the calibration curve of TDR using soil samples from 5 regions in Denmark (ρb of the samples were between $1.38\text{-}1.78\times10^3$ kg.m^{-3}). They found a third order relationship between volumetric soil content and the calibration dielectric as follow:

$$\theta_v = -3.41\times10^{-2} + 3.45\times10^{-2}k_a - 11.4\times10^{-4}k_a^2 - 11.4\times10^{-4}k_a^2 + 17.1\times10^{-6}k_a^3 - 3.7\times10^{-2}\rho + 7.36\times10^{-4}\%clay + 47.7\times10^{-4}\%OM \quad (1)$$

Where OM is the percent of organic matter in soil and ka is dielectric constant. In current study, the effects of soil texture and organic maters on the accuracy of a TDR equipment in measuring volumetric soil moisture has been studied and calibrated relationships have been proposed for different soil textures at different levels of organic maters.

Methods and Materials

TDR Theory

In the TDR method, a very fast rise time (approx. 200 ps) step voltage increase is injected into a waveguide that carries the pulse to a probe placed in the soil or other porous medium. The velocity of the pulse in the probe is measured and related to soil water content, with smaller velocities indicating wetter soils. In a typical field installation, probes are connected to the instrument through a network of coaxial cables and multiplexers. Although most TDR instruments display the horizontal axis in units of length (a holdover from the primary use of these instruments in detecting the location of cable faults), the horizontal axis is actually measured in units of time. The TDR instrument converts the time measurement to length units by using the relative propagation velocity factor setting, v_p, which is a fraction of the speed of light in a vacuum. For a given cable, the correct value of v_p is inversely proportional to the permittivity, ε, of the dielectric (insulating plastic) between the inner and outer conductors of the cable:

$$v_p = \frac{v}{c_o} = (\varepsilon\mu)^{-0.5} \tag{2}$$

Where v is the propagation velocity of the pulse along the cable, c_o is the speed of light in a vacuum, and μ is the magnetic permeability of the dielectric material. For a TDR probe in a soil, the dielectric between the probe rods is a complex mixture of air, water and soil particles that exhibits a variable apparent permittivity; ε_b. Water is the largest determinant of permittivity in soils. It has a permittivity of approximately 80, whereas the permittivity of soil minerals varies in the range of 3 to 5; the permittivity of organic matter is likewise low; and the permittivity of air is unity. Also, soil water is the only rapidly changing determinant of ε_b. Thus, we are able to usefully calibrate soil water content vs. measured ε_b. The measured property in the TDR method is the travel time, t_t, of the electronic pulse along the length (L) of the probe rods that are exposed to the soil. The velocity of the pulse can be calculated as $v = 2L/t_t$. Assuming μ=1, one sees that an apparent permittivity, ε_b, may be determined for a probe of known length, L, by measuring t_t :

$$\varepsilon_b = \left[\frac{c_o t_t}{2L}\right]^2 \tag{4}$$

Two general methods have been used for stating the relationship between the dielectric constant and
volumetric soil content. The first methods are empirical equations among which, Topp's model is a well-known model. Topp et al. (1980) found that a single polynomial function described the relationship between volumetric water content, θ_v, and values of ε_b determined from Eq. 5 for four mineral soils.

$$\theta_v = 5.3 \times 10^{-2} + 2.92 \times 10^{-2}\varepsilon_b - 5.5 \times 10^{-4}(\varepsilon_b)^2 + 4.3 \times 10^{-6}(\varepsilon_b)^3 \tag{5}$$

The second methods which have physical basis are based on a mixture of the dielectrics of all phases forming soils. Since 1980, other researchers have noted that the quantity $[t_t/(2L)]$ in Eq. 5 is quadratic, and have shown that the relationship between θv and $t_t/(2L)$ is practically linear (e.g. Ledieu et al. 1986). Several attempts have been made to predict ε_b of soils from theoretical considerations using dielectric mixing models that consider the volumetric proportions of soil mineral, organic, water, and air constituents, as well as soil mineralogy and particle shape and packing considerations (e.g. Roth et al. 1990). Dobson et al. (1985) and Roth et al. (1990) proposed this mixing model:

$$\theta_v = \frac{\varepsilon_b^{\beta} - (1-\eta)\varepsilon_s^{\beta} - \eta\varepsilon_a^{\beta}}{\varepsilon_w^{\beta} - \varepsilon_a^{\beta}} \tag{6}$$

Where η is the soil porosity, ε_a, ε_s and ε_w are the dielectric constants for air, soil and water (respectively) and β is the shape parameter which can be set to 0.5 for a two-phase medium. Dielectric constant can be affected by soil texture, bulk density, electric conductivity of pore water and the percent of organic maters in soil layers. Specific area of soil particles is one of the most important factors affecting the soil dielectric. This property increases the adsorption of ions on soil colloids and causes the increase of diffused double layer. Gouy (1910) and Chapman (1913) proposed a relationship between the inverse of the thickness of diffused double layer (d) and the dielectric constant:

$$d = \left(\frac{1000 \; e^2 N_A \sum_i Z_i^2 M_i}{\varepsilon k T} \right)^{1/2} \tag{7}$$

Where N_A is Avogadro's value (6.02×10^{23}), Zi is ion capacity, Mi is the molar concentration of ion i, ε is the dielectric constant, k is Boltzman's constant (1.38×10^{-23}) and T is the absolute temperature (k^o). Since any increase in specific area of soil particles decreases diffused double layer, according to this equation, increases dielectric constant. In this study, the effects of soil texture and percent of organic maters on the accuracy of TDR measurements were analyzed because of their great impacts on soil bulk density and specific area of soil particles. Soil moisture measurements were carried out using a TDR model TRASE (6050X1) in the Agricultural Research Center of East Azerbaijan (Iran). Two-wing waveguides of 15-cm length were used. This set has three windows for soil moisture measurements (10, 20 and 40 nanosecond), all were used in this study. 5 soil samples (Sandy, Sandy Loam, Loam, Sandy Clay Loam and Clay Loam) at 3 levels of organic maters (2%, 3.5% and 5%) were analyzed in a complete random design (Table 1). The samples were prepared in buckets of 23-cm height and with uniform density. Then, the buckets were saturated from the bottom and TDR probes were fixed. Soil moisture measurements were carried out using TDR and also using weighting (direct) method. Also, in each measurement, the soil dielectric constant were recorded for all the samples. The values of 1, 81 and 4 were chosen for air, water and soil dielectric (according to previous studies) and the value of 0.5 was selected for β.

Table1- Physical and chemical characteristics of the samples.

PH	Ec (ds/m)	ρb (gr/cm^3)	Organic matter (%)	Soil texture
7.2	0.95	2.76	2	Light(sand)
7.7	0.94	2.72	3.5	
7.5	0.95	2.70	5	
7.7	2.00	2.65	2	Light(sandy loam)
7.5	1.99	2.63	3.5	
7.9	2.00	2.60	5	
7.1	2.40	2.46	2	Medium(loam)
7.3	2.40	2.40	3.5	
7.4	2.45	2.37	5	
8.3	1.88	2.30	2	Heavy(Sandy Clay Loam)
8.5	1.80	2.25	3.5	
8.1	1.87	2.20	5	
8.4	2.00	2.21	2	Heavy(Clay Loam)
8.2	2.10	2.15	3.5	
8.3	2.00	2.12	5	

In order to assess the performance of the models, RMSE and RE statistics were used:

$$RMSE = \sqrt{\frac{\sum_i^n di^2}{n}} \tag{8}$$

$$RE = \frac{\sum_i^n \left|\theta_v - \theta_{TDR}\right|/n}{\bar{\theta}_v} \times 100 \tag{9}$$

Where RMSE: the root mean square error (cm^3/cm^3), RE: relative error (%), di : the difference between measured and TDR soil moisture (cm^3/cm^3), n: the number of samples, θv: the volumetric soil moisture (by weighting method)(cm^3/cm^3), $\overline{\theta_v}$: the average value of θv and θ_{TDR}: the volumetric soil moisture measured by TDR(cm^3/cm^3).

Results and Discussion

Using the measurement window of 10 nanosecond (ns) outperformed the 20 and 40 ns windows. Although, differences between 10 and 20 ns windows were not significant and can be related to the difference between TDR measured travel time and the length of measurement window. The results are presented in Table 2.

Table 2- Values of Relative error (RE) and Root means square error (RMSE) in 10, 20 and 40 ns windows.

Soil texture	organic matter (%)	windows					
		10		20		40	
		RE	RMSE	RE	RMSE	RE	RMSE
sand	2	0.002	3.97	0.012	5.65	0.035	17.00
	3.5	0.009	4.11	0.010	4.67	0.033	15.71
	5	0.010	4.13	0.012	4.48	0.026	10.87
sandy loam	2	0.019	6.90	0.023	8.49	0.052	21.58
	3.5	0.026	9.99	0.032	12.32	0.021	21.28
	5	0.025	9.82	0.036	14.30	0.058	23.46
loam	2	0.028	7.44	0.027	7.08	0.049	15.32
	3.5	0.025	7.78	0.028	8.84	0.067	21.94
	5	0.032	8.89	0.033	8.96	0.049	14.86
Sandy Clay Loam	2	0.032	7.74	0.036	7.75	0.060	17.85
	3.5	0.034	9.47	0.036	9.88	0.071	20.99
	5	0.035	10.19	0.037	10.80	0.075	23.08
Clay Loam	2	0.037	10.70	0.037	10.82	0.058	17.03
	3.5	0.040	11.69	0.042	11.98	0.073	22.25
	5	0.042	11.79	0.043	12.02	0.096	26.60

The difference between the soil moisture values measured by TDR and weighting method was limited to 0.5-1% for light-textured samples (Sandy and Sandy Loam). These differences decreased with the reduction of soil moisture. For Loam texture and at high levels of moisture, TDR overestimated the soil moisture, while the differences decreased with the reduction of soil moisture. The difference between the values measured by TDR and the values measured by weighting method was limited to 1-2%. In heavy-textured samples (Sandy Clay Loam and Clay Loam), TDR overestimated the values of weighting method, in general (max. 3%). It can be concluded that by the increase of the soil clay and organic maters percent, specific area of soil particles increases which causes the diffused double layer to decrease, the dielectric constant to increase and resulted in overestimation of soil moisture. The results of the comparisons made for Sandy and Clay Loam samples were shown in Figure 1. RMSE and RE statistics of Topp’s model are lower than the statistics of mixing model (Table 3). The increase of the percent of soil clay and organic maters resulted in the increase of the errors for all models. The measured values of soil moisture were depicted versus the values estimated by all three models as a function of the dielectric constant in Figures 2 and 3 (Only for Sandy and Clay Loam samples). These graphs show that for a given value of dielectric, the differences between measured values and mixing model are more than the differences of TDR and Topp’s model. Tables 4 through 8 show the calibrated relationships for the TDR method. As it can be seen, the correlation coefficient of these equations decreases with the increase of soil organic maters and clay percent.

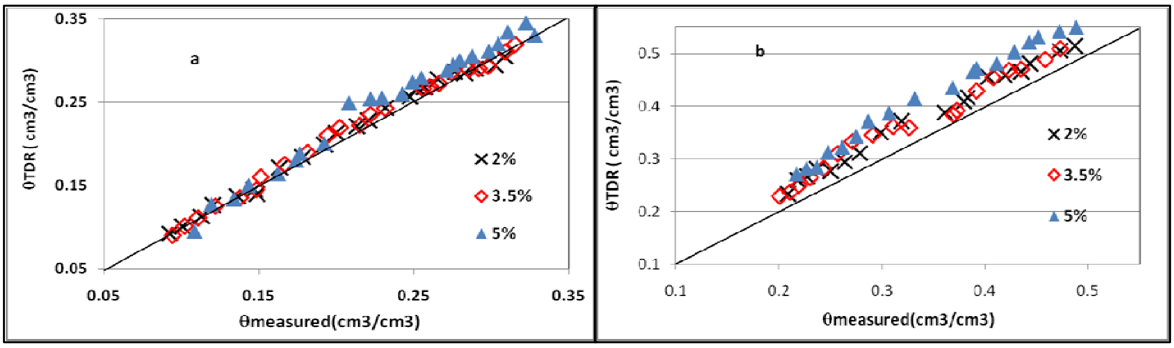

Figure 1- Comparison of soil moisture measured with weight and TDR methods for different level 2, 3.5 and 5 percent of organic matter: (a) sand, (b) clay loam

Table 3- Values of Relative error (RE) and Root means square error (RMSE) in models.

Soil texture	organic matter (%)	Mixed model		Topp et al (1980) model	
		RE	RMSE	RE	RMSE
sand	2	0.019	7.73	0.009	3.66
	3.5	0.012	5.41	0.005	2.00
	5	0.013	5.19	0.009	3.28
sandy loam	2	0.033	12.26	0.019	4.48
	3.5	0.041	14.83	0.027	5.30
	5	0.042	15.98	0.026	5.19
loam	2	0.045	13.73	0.031	5.98
	3.5	0.044	14.41	0.021	6.57
	5	0.053	15.74	0.032	7.78
Sandy Clay Loam	2	0.054	13.24	0.029	7.23
	3.5	0.053	15.41	0.029	7.63
	5	0.055	16.38	0.031	7.95
Clay Loam	2	0.058	16.65	0.033	8.49
	3.5	0.061	17.88	0.036	9.23
	5	0.064	18.06	0.039	10.13

Table 4- Equation fitted to different level of organic matter in sandy soil.

organic matter (%)	Equation of line	R^2
2	$\theta_v=0.939\theta_{TDR}+0.006$	0.991
3.5	$\theta_v=0.986\theta_{TDR}+0.001$	0.981
5	$\theta_v=0.956\theta_{TDR}+0.012$	0.977

Table 5 Equation fitted to different level of organic matter in sandy loam soil.

organic matter (%)	Equation of line	R^2
2	$\theta_v=0.962\theta_{TDR} - 0.006$	0.991
3.5	$\theta_v=1.014\theta_{TDR} - 0.023$	0.981
5	$\theta_v=0.990\theta_{TDR} - 0.021$	0.977

Table 6- Equation fitted to different level of organic matter in loam soil.

organic matter (%)	Equation of line	R^2
2	$\theta_v=0.998\theta_{TDR} - 0.035$	0.930
3.5	$\theta_v=0.992\theta_{TDR} - 0.035$	0.930
5	$\theta_v=1.080\theta_{TDR}+0.072$	0.870

Table 7- Equation fitted to different level of organic matter in sandy clay loam soil.

organic matter (%)	Equation of line	R^2
2	$\theta_v=0.896\theta_{TDR}+0.021$	0.951
3.5	$\theta_v=0.978\theta_{TDR} - 0.014$	0.979
5	$\theta_v=0.973\theta_{TDR} - 0.018$	0.970

Table 8- Equation fitted to different level of organic matter in clay loam soil.

organic matter (%)	**equation of line**	**R^2**
2	$\theta_v=0.878\theta_{TDR}+0.020$	0.955
3.5	$\theta_v=0.971\theta_{TDR} - 0.020$	0.964
5	$\theta_v=0.993\theta_{TDR} - 0.030$	0.922

Summary

The results of this study showed that the estimated soil moisture using TDR method for 10 and 40 ns windows have the highest and lowest accuracy, respectively. Also, with the increase of soil clay percent and the percent of organic maters in soil samples, the dielectric constant increases and the accuracy of TDR method decreases significantly. For all the samples (textures) and at all levels of organic maters, Topp's model outperforms the mixing model. Both models are more accurate in light textures and lower percent of organic maters. Moreover, by the decrease of soil moisture, the accuracy of TDR increases in measuring volumetric soil moisture (in comparison to the weighting method).

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Experimental Study of the Performance of Some Models in the Time Domain Reflection Measuring Soil Moisture Estimation

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Abstract: Soil moisture identification using time domain reflectometry method has recently been introduced as a modern and useful method. The soil dielectric constant is the basis of the soil moisture measurement using TDR system. On the other hand, the value of this character directly depends on some physical and chemical soil features, so the practical application of this machine needs calibration and identification of calibrated relations between dielectric and soil moisture. Several experimental relations between the dielectric constant of machine and volumetric soil moisture have already been demonstrated such as Topp et al (1980), mixture (1990), Malicki et al (1996), Roth et al (1992), Herkelrath et al (1991), Oleszczuk et al (2004), Pepin et al (1992) and white et al (1994) models. But the effects of all effective soil moisture features on the precision of machine application were not considered in all mentioned researches. The purpose of this research is the evaluation of precision in all mentioned models for some kinds of soil with various texture which contain various organic matter. Therefore, soil moisture identification has been experimented using TDR system for 5 various soil tissues (soft, mean and heavy), 3 organic matter levels (2, 3.5 and 5 percent) which contain litter fertilizer. These experiments have been carried out under random conditions. Results showed that Topp et al (1980), white et al (1994), mixture, Roth et al (1992) and Oleszczuk et al (2004) models with Root Mean Square Error (RMSE) 0.03, 0.035, 0.042, 0.07 and 0.08 (m^3/m^3), respectively are the best applications and Malicki et al (1996), Herkelrath et al (1991) and Pepin et al (1992) with Root Mean Square Error (RMSE) 0.23, 0.2 and 0.19 (m^3/m^3) are the less application in soil moisture assessment.

Keywords: TDR, Calibration, Dielectric Constant, RMSE.

Introduction

Soil moisture is one of most important factors affecting crop growth. It has impacts on many of soil properties as well as soil air content, transportation of gases, soil micro organisms and soil chemical processes. Recently, the time domain-reflectometry method (TDR) is being used increasingly for measuring the moisture content of soils. The determination of soil water content using the TDR method requires the knowledge of the relationship between the apparent dielectric constant (Ka) and the volumetric moisture content of the soil (θv). The TDR is based on the delay produced on an electromagnetic wave that travels along a probe inserted in the soil. The dielectric constant of most mineral soils varies between 3 and 10, that of air is approximately 1 and is 81 for water. Thus, small variations in the water content of an unsaturated soil lead to significant changes in the bulk air–soil–water dielectric constant, Ka. The Ka constant is determined from the transit time (t) that a voltage pulse takes to travel forward and backward along a wave-guide of length L, using the expression Ka =(ct/2L)2, where c is the velocity of light in vacuum. The first more general calibration results were published by Topp et al. (1980). They found a third order polynomial relationship between (Ka) and (θv) as the form of calibration equation to be valid for mineral soils ranging from sandy loam to heavy clay soils.Topp et al. (1980) also established a calibration equation (Ka (θ)) for organic soils. A number of empirical relationships between the dielectric constant and soil water content for mineral soils were established later (Roth et al., 1992). However, only a few relationships for organic and peat soils were developed. The calibration equations for organic soils published in the literature together with a ‘universal’ equation, was proposed by Topp et al. (1980). Considering that the dielectric constant is under the influence of soil texture, density and percent organic matter. therefore, get to a precise relationship That can consider the effect these factors together to estimate the soil dielectric constant has great importance. In this paper examined operation some of provided calibrated models by different researchers in organic and mineral soils for estimation soil moisture based on dielectric constant (Table 1) .

Table 1- Presented empirical equations by different researchers for organic and mineral soils

Authors	Type of soil	Presented empirical equations
Topp et al (1980)	Mineral soil	$\theta_v = \left(-530 + 292K - 5.5K^2 + 0.043K^3\right)\times 10^{-4}$
Mixing model (1990)	Mineral and organic soil	$\theta_v = \frac{K^{\beta} - (1-n)K_s^{\beta} - nK_a^{\beta}}{K_w^{\beta} - K_a^{\beta}}$
Malicki et al (1996)	organic soil	$\theta_v = \frac{(\sqrt{K} - 0.819 - 0.168\rho - 0.159\rho^2)}{17.89 - 9.045\rho}$
Roth et al (1990)	Mineral and organic soil	$\theta_v = \left(-233 + 285K - 4.3K^2 + 0.03K^3\right)\times 10^{-4}$
Herkelrath et al. (1991)	Typic Haplorthod	$\theta_v = \left(-0.051 + 0.127K\right)\times 10^{-4}$
Oleszczuk et al. (2004)	Peat soil	$\theta_v = \left(-27.6 + 247.7K - 3.15K^2 + 0.02K^3\right)\times 10^{-4}$
Pepin et al. (1992)	Sphagnum peat	$\theta_v = \left(850 + 192K - 0.95K^2\right)\times 10^{-4}$
White et al (1994)	Mineral soil	$\theta_v = \frac{\left(\sqrt{K} - 1.451\right)}{8.979}$

Methods and Materials

This research was carried out using existing soil in some parts of East Azerbaijan, Iran. Some of properties of soil samples showed in table 2.

Table 2- Physical and chemical characteristics of the samples.

PH	EC (ds/m)	ρb (gr/cm^3)	Organic matter (%)	Soil texture
7.2	0.95	2.76	2	
7.7	0.94	2.72	3.5	Light(sand)
7.5	0.95	2.70	5	
7.7	2.00	2.65	2	
7.5	1.99	2.63	3.5	Light(sandy loam)
7.9	2.00	2.60	5	
7.1	2.40	2.46	2	
7.3	2.40	2.40	3.5	Medium(loam)
7.4	2.45	2.37	5	
8.3	1.88	2.30	2	
8.5	1.80	2.25	3.5	Heavy(Sandy Clay Loam)
8.1	1.87	2.20	5	
8.4	2.00	2.21	2	
8.2	2.10	2.15	3.5	Heavy(Clay Loam)
8.3	2.00	2.12	5	

Soil moisture measurements were carried out using a TDR model TRASE (6050X1). Two-wing waveguides of 15-cm length were used. This set has three windows for soil moisture measurements (10, 20 and 40 nanosecond), that In this study was used of 10 nanosecond window because of its high accuracy data recorded. The samples were prepared in buckets of 23-cm height and with uniform density. Then, the buckets were saturated and TDR probes were fixed. Soil moisture measurements were carried out using TDR and weighting (direct) method. Also, in each measurement, the soil dielectric constant were recorded for all the samples. The values of 1, 81 and 4 were chosen for air, water and soil dielectric (according to previous studies) and the value of 0.5 was selected for β. In order to assess the performance of the models, RMSE and RE statistics were used:

$$RMSE = \sqrt{\frac{\sum_i^n di^2}{n}} \tag{1}$$

$$RE = \frac{\sum_i^n \left|\theta_v - \theta_{TDR}\right|/n}{\bar{\theta}_v}\times 100 \tag{2}$$

Where RMSE: the root mean square error (cm^3/cm^3), RE: relative error (%), di : the difference between measured and TDR soil moisture (cm^3/cm^3), n: the number of samples, θv: the volumetric soil moisture (by weighting method)(cm³/cm³), $\overline{\theta_v}$: the average value of θv and θ_{TDR}: the volumetric soil moisture measured by TDR(cm³/cm³).

Results and Discussion

Values obtained from RE and RMSE is showed in table 3. It was revealed that Topp et al (1980), white et al (1994), mixture, Roth et al (1992) and Oleszczuk et al (2004) models with the mean RMSE values of 0.03, 0.035, 0.042, 0.07 and 0.08, respectively, outperformed the Malicki et al (1996), Herkelrath et al (1991) and Pepin et al (1992) with the mean RMSE values of 0.23, 0.2 and 0.19, respectively. This trend was common between all the studied soil textures and in all levels organic matter. On the other hand results showed that the increase of the percent of soil clay and organic maters resulted in the increase of the errors for all models.

Table 3- The mean RE and the mean RMSE values in a from models

texture	OM (%)	model															
		Topp		White		Mixing		Malicki		Roth		Oleszczuk		Pepin		Herkelrath	
		RE	**RMSE**	**RE**	**RMSE**	**RE**	**RMSE**	**RE**	**RMSE**	**RE**	**RMSE**	**RE**	**RMSE**	**RE**	**RMSE**	**RE**	**RMSE**
sand	2	0.011	4.28	0.010	3.90	0.014	5.19	0.201	65.26	0.054	15.56	0.042	11.86	0.069	28.91	0.129	54.10
	3.5	0.013	4.72	0.011	4.27	0.016	5.41	0.212	66.58	0.058	16.41	0.046	13.39	0.081	34.03	0.193	60.71
	5	0.019	6.77	0.017	5.93	0.027	7.70	0.242	74.55	0.068	20.67	0.062	17.05	0.082	35.96	0.198	61.70
sandy loam	2	0.012	5.30	0.011	4.07	0.043	12.26	0.211	66.52	0.059	16.67	0.043	12.40	0.083	35.91	0.203	64.34
	3.5	0.015	5.30	0.029	8.01	0.048	14.41	0.242	74.15	0.067	24.59	0.067	19.76	0.081	37.83	0.211	66.49
	5	0.026	7.70	0.031	8.95	0.054	15.74	0.250	75.72	0.071	30.77	0.068	20.47	0.084	39.52	0.221	68.81
loam	2	0.014	4.90	0.029	8.08	0.044	13.24	0.270	79.74	0.066	23.80	0.066	22.56	0.085	41.17	0.220	69.01
	3.5	0.036	9.01	0.041	10.17	0.049	14.83	0.272	80.68	0.069	26.85	0.067	23.82	0.086	42.50	0.230	70.08
	5	0.039	10.65	0.041	11.60	0.056	15.98	0.291	86.19	0.072	32.02	0.069	26.84	0.088	43.87	0.235	70.76
Sandy Clay Loam	2	0.042	11.14	0.040	11.52	0.042	13.73	0.274	81.39	0.070	27.97	0.069	26.77	0.089	44.68	0.243	73.56
	3.5	0.044	12.39	0.042	12.25	0.053	15.41	0.295	86.74	0.072	31.32	0.070	29.74	0.087	44.51	0.245	74.72
	5	0.051	15.07	0.063	17.01	0.057	16.38	0.307	94.39	0.088	43.55	0.081	33.08	0.129	50.38	0.281	82.13
Clay Loam	2	0.044	12.81	0.064	21.97	0.059	16.65	0.290	85.70	0.071	31.24	0.082	38.20	0.199	61.29	0.345	101.6
	3.5	0.046	13.26	0.068	24.24	0.063	17.88	0.340	105.7	0.081	37.71	0.086	40.73	0.206	64.61	0.351	106.5
	5	0.061	17.67	0.070	27.00	0.065	18.06	0.383	114.3	0.090	46.18	0.088	44.01	0.221	68.71	0.372	111.1

Also, by comparing the mean RE values for different ranges of soil moisture to the conclusion reached that the Topp et al (1980), white et al (1994) and mixture models in Sandy textures and in lower ranges of soil moisture (less than 16%) and in higher ranges of soil moisture (more than 17%) were more accurate with the mean RE values of 3% and 13%, respectively. In Sandy Loam textures and in lower ranges of soil moisture (less than 18%) and in higher ranges of soil moisture (more than 19%), these models had the mean RE values of 14% and 10%, respectively. In Loam, Sandy Clay Loam and Clay Loam soil textures, the Topp et al (1980), white et al (1994) models in the soil moisture content of less than 30% and more than 31% had the mean RE values of 25% and 14% and outperformed the other models (Table 4).

Table 4- Values of the mean RE obtained from models.

Soil texture	Moisture range	model				
		Topp et al (1980)	White et al (1994)	Mixing (1990)	Oleszczuk et al. (2004)	Roth et al (1990)
Light(sand)	16 – 10	2.08	2.80	2.91	15.04	15.78
	31 - 17	8.21	7.14	9.85	13.06	21.32
Light (sandy loam)	18 - 14	7.40	8.53	14.15	20.19	28.01
	33 - 19	4.71	4.64	10.19	18.49	23.30
Medium (loam)	30 – 14	10.90	11.50	15.08	22.84	26.25
	45 - 31	5.82	6.95	13.87	19.84	21.05
Heavy(Sandy Clay Loam)	28 - 15	16.42	18.10	16.65	31.09	32.91
	46 - 29	8.56	9.66	12.10	24.55	26.99
Heavy (Clay Loam)	30 - 20	18.84	25.25	17.44	46.67	45.87
	54 - 31	9.16	13.62	15.90	29.17	33.75

Tables 5 through 9 show the calibrated relationships for the TDR method. As it can be seen, the correlation coefficient of these equations decreases with the increase of soil organic maters and clay percent. With heavy and increasing soil organic matter, specific surface of soil particles increases that causes Reduction of the double layer and the dielectric constant. The model of Topp et al. (1980) has the highest values of correlation coefficient in light soil textures. Figures 1 through 5 show the values of measured dielectric constant and the estimated values of soil moisture using all the models in all the soil samples. It can be seen that with the calibration of the equations of all the studied models, their accuracy increase significantly.

Table 5- Equations of calibrated models for estimation soil moisture in sandy soil.

model	**equation lines**	R^2
Topp et al (1980)	$\theta v = 0.937\ \theta m - 0.054$	0.99
Pepin et al (1992)	$\theta v = 0.964\ \theta m - 0.069$	0.92
Roth et al (1990)	$\theta v = 0.083\ \theta m + 0.006$	0.94
Mixing (1990)	$\theta v = 0.902\ \theta m + 0.001$	0.97
Oleszczuk et al. (2004)	$\theta v = 0.897\ \theta m - 0.003$	0.95
Herkelrath et al (1991)	$\theta v = 0.860\ \theta m + 0.109$	0.88
Malicki et al (1996)	$\theta v = 0.602\ \theta m - 0.003$	0.87
White et al (1994)	$\theta v = 0.981\ \theta m + 0.005$	0.99

Table 6- Equations of calibrated models for estimation soil moisture in sandy loam soil.

model	**equation lines**	R^2
Topp et al (1980)	$\theta v = 0.905\ \theta m - 0.092$	0.98
Pepin et al (1992)	$\theta v = 0.803\ \theta m - 0.066$	0.91
Roth et al (1990)	$\theta v = 0.727\ \theta m - 0.012$	0.93
Mixing (1990)	$\theta v = 0.830\ \theta m + 0.012$	0.95
Oleszczuk et al. (2004)	$\theta v = 0.744\ \theta m - 0.018$	0.94
Herkelrath et al (1991)	$\theta v = 0.770\ \theta m - 0.121$	0.86
Malicki et al (1996)	$\theta v = 0.631\ \theta m - 0.033$	0.86
White et al (1994)	$\theta v = 0.879\ \theta m - 0.018$	0.97

Table7- Equations of calibrated models for estimation soil moisture in loam soil.

model	**equation lines**	R^2
Topp et al (1980)	$\theta v = 1.309\ \theta m - 0.091$	0.96
Pepin et al (1992)	$\theta v = 0.874\ \theta m - 0.053$	0.88
Roth et al (1990)	$\theta v = 0.846\ \theta m - 0.006$	0.92
Mixing (1990)	$\theta v = 0.874\ \theta m + 0.002$	0.94
Oleszczuk et al. (2004)	$\theta v = 0.844\ \theta m - 0.009$	0.92
Herkelrath et al (1991)	$\theta v = 0.792\ \theta m - 0.073$	0.85
Malicki et al (1996)	$\theta v = 0.647\ \theta m - 0.035$	0.84
White et al (1994)	$\theta v = 1.002\ \theta m - 0.008$	0.95

Table 8- Equations of calibrated models for estimation soil moisture in Sandy Clay Loam soil.

model	equation lines	R^2
Topp et al (1980)	$\theta v= 1.025\ \theta m - 0.110$	0.95
Pepin et al (1992)	$\theta v= 0.816\ \theta m - 0.049$	0.87
Roth et al (1990)	$\theta v= 0.829\ \theta m - 0.020$	0.90
Mixing (1990)	$\theta v= 0.887\ \theta m - 0.006$	0.94
Oleszczuk et al. (2004)	$\theta v= 0.853\ \theta m - 0.019$	0.92
Herkelrath et al (1991)	$\theta v= 0.860\ \theta m + 0.137$	0.84
Malicki et al (1996)	$\theta v= 0.615\ \theta m - 0.038$	0.83
White et al (1994)	$\theta v= 0.\ 980\ \theta m - 0.022$	0.95

Table 9- Equations of calibrated models for estimation soil moisture in Clay Loam soil.

model	equation lines	R^2
Topp et al (1980)	$\theta v= 1.084\ \theta m - 0.152$	0.95
Pepin et al (1992)	$\theta v= 0.852\ \theta m - 0.079$	0.86
Roth et al (1990)	$\theta v= 0.908\ \theta m - 0.068$	0.91
Mixing (1990)	$\theta v= 1.013\ \theta m - 0.064$	0.94
Oleszczuk et al. (2004)	$\theta v= 0.923\ \theta m - 0.063$	0.88
Herkelrath et al (1991)	$\theta v= 0.929\ \theta m - 0.189$	0.81
Malicki et al (1996)	$\theta v= 0.665\ \theta m - 0.083$	0.81
White et al (1994)	$\theta v= 0.\ 980\ \theta m - 0.022$	0.92

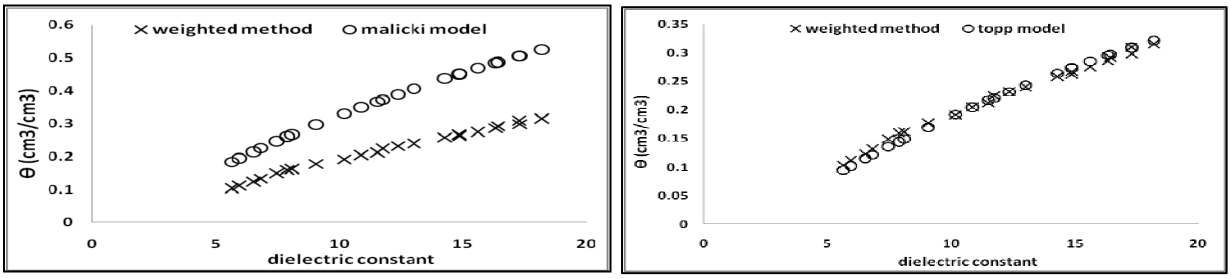

Figure 1- Comparison of soil moisture measured by weight method and models in sandy soil.

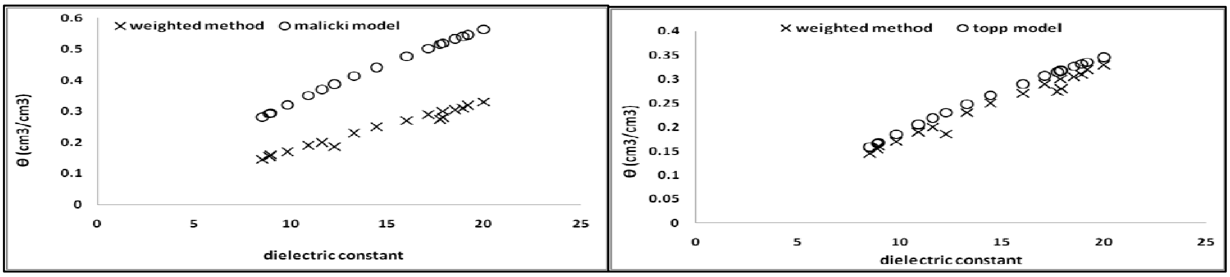

Figure 2- Comparison of soil moisture measured by weight method and models in sandy loam soil.

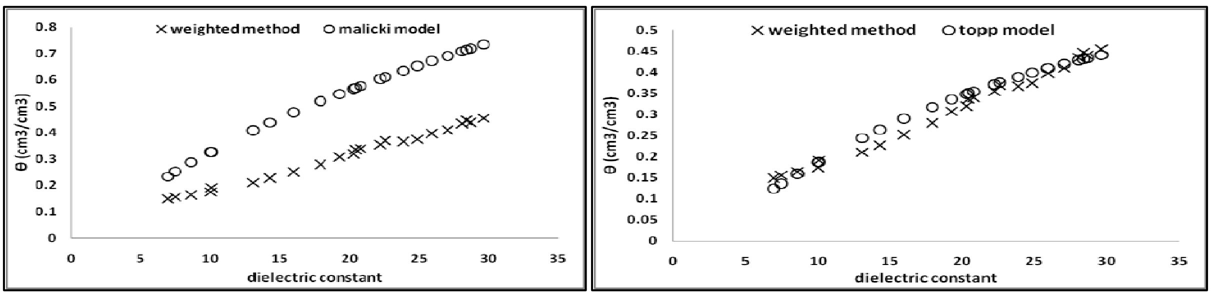

Figure 3- Comparison of soil moisture measured by weight method and models in loam soil.

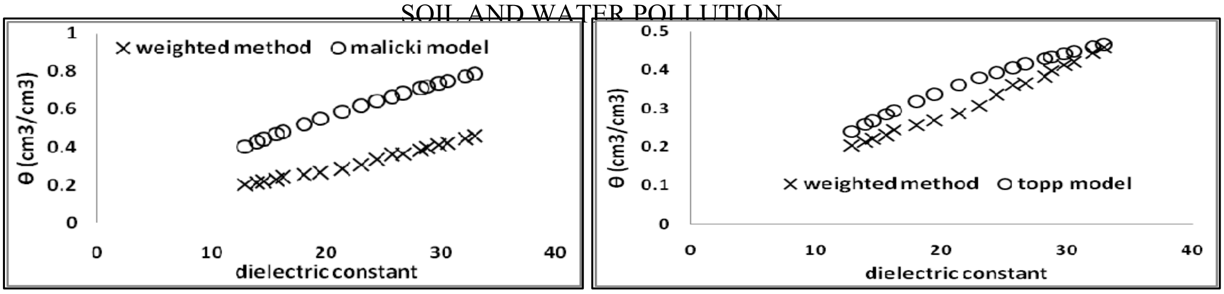

Figure 4- Comparison of soil moisture measured by weight method and models in sandy clay loam soil.

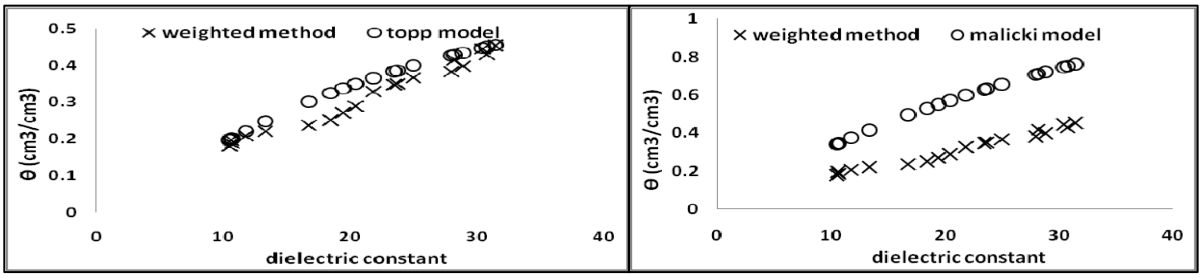

Figure 5- Comparison of soil moisture measured by weight method and models in clay loam soil.

Summary

The results of this study showed that with the increase of soil clay percent and the percent of organic maters in soil samples, the dielectric constant increases and the accuracy of TDR method decreases significantly. The accuracy of the studied models decreased With heavy and increasing soil organic matter. Also, it was revealed that the models of Topp et al (1980), White et al(1994) and the mixing (1990) model outperformed the other models. It is suggested that the effects of soil bulk density, soil salinity and temperature on the accuracy of these models should be studied.

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Residual Zn-Cd Interaction on Cadmium Uptake of Spinach

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Abstract

This Experiment was performed as completely randomized block design with three replicates. The experiment was a 20×3×2 factorial with 3 levels of Zn (0, 5 and 10 µg/g soil as ZnSO4, 7H2O), 2 levels of Cd (0 and 3 µg/g soil as 3CdSO4, 8H2O) remaining from the previous experiment "Effect of Zn and Cd on growth and chemical composition of rice" and 20 soils in the greenhouse.

The results showed that application of 3 µg Cd/g soil that were remained from previous experiment increased crop tissue Cd concentration as much as 7.4 µg /g in relation to Cd control treatment (0.65 µg Cd /g plant). Zn residual treatments that were remained from previous experiment decreased Cd concentration of crop tissue in all soils, so that mean concentration of Cd in control of Zn treatment was 4.49 µg Cd/g decreased to 4.06 and 3.60 µg Cd/g respectively in treatments 5 and 10 µg Zn /g soil that were remained from previous experiment.

Interaction of Cd-Zn showed that in any levels of residual Cd treatment, soil residual Zn treatments reduced the Cd concentration of crop tissue, so that this reduction was negligible in control of Cd treatment where as substantial in treatment 3 µg Cd /g soil.

Keywords: Cd, Zn, interaction, spinach

Introduction

High ability of spinach in cadmium absorption reported by several researchers. Vaseghi and et al (2002) in a study of sewage sludge on heavy metal uptake by spinach and lettuce in four soils, observed that the mean of Cd concentration in all four soils was 1.32 µg Cd/g that increased up to 2.56 µg Cd/g in dry matter of spinach with application of sewage sludge. Rahimi (1993) reported that Cd uptake in both check and the soil which have treated by Esfehan compost was very low. Hooda and et al (1997) reported that the Cd absorption decreased in the soil which have treated with sewage sludge in fallowing plants, spinach>> carrot> wheat. Antagonistic effect of Zn on Cd concentrations may be caused of dilution effect or antagonism effect of Zn on transportation of Cd from root to leaves. Low integrity of cell membrane coz of nutrient deficiency and may be facility of Cd and other ions by mass flow (Cakmak and Marschner, 1988). Alloway (1990) believed that Zn in low level up to medium has antagonistic effect with Cd but in high level it has synergistic effects. Mclaughlin and et al (1996) in a field study reported that application of 100 Kg Zn/ha had negative interaction between Zn and Cd in potato tuber.

Material and methods

This Experiment was performed as completely randomized block design with three replicates. The experiment was a 20×3×2 factorial with 3 levels of Zn (0, 5 and 10 µg/g soil as ZnSO4, 7H2O), 2 levels of Cd (0 and 3 µg/g soil as 3CdSO4, 8H2O) remaining from the previous experiment "Effect of Zn and Cd on growth and chemical composition of rice" and 20 soils in the greenhouse.

There was four Kg soils in any pots from first step of trial, after screening and separating the roots the soils and has got back to the pots again. It was added 50 µg N/g, 5 µg Fe/g and 10 µg Mn/g soil from urea, FeEDDHA and MnSO4, H2O respectively in solution form. Then distilled water added to the soils up to field capacity (F.C), which after moisture decreasing the soil of the pots mixed and refilled in the pots. Ten seeds of spinach from domestic variety cultivated in 0.5 cm depth. When plants were grown up to 5 cm height, half of them has omitted. Ordinary operation in plant growth stage, such as, irrigation has been carried out until harvesting. 50 µg N/g as a urea added to any pots by top dressings in plant 8 cm heights. The aerial parts of plants has cut from soil surface an after washing dried in 65 °C duration 48 h and its dried matter weighted. The samples milled into the powder and digested by dry ashing in electrical furnance (650 °C) then measured by AAS.

Results

The results showed that application of 3 µg Cd/g soil that were remained from previous trial increased crop tissue Cd concentration as much as 7.4 µg /g in compare with to Cd check (0.65 µg Cd /g plant) treatment (Table 1). Zn residual treatments that were remained from previous trial decreased Cd concentration of crop tissue in all soils, so that mean concentration of Cd in control of Zn treatment was 4.49 µg Cd/g plant decreased to 4.06 and 3.60 µg Cd/g plant respectively, in treatments 5 and 10 µg Zn /g soil that were remained from previous trial (Table 1). Interaction of Cd-Zn showed that in any levels of residual Cd treatment, soil residual Zn treatments reduced the Cd concentration of crop tissue, so that this reduction was negligible in control of Cd treatment where as substantial in treatment 3 µg Cd /g soil.

Table 1. Effect of residual Zn on the growth (aerial part of plant) and [Zn and Cd] in spinach

Zn levels (µg /g soil)	Dry matter (g)	Zn concentration (µg /g plant)	Cd concentration (µg /g plant)
0	9.55 a	53.4 c	4.5 c
5	9.40 a	66.2 b	4.1 b
10	9.49 a	81.3 a	3.6 a
Cd levels (µg /g soil)			
0	9.35 a	29.60 a	0.65 b
3	9.38 a	28.70 a	5.6 a

*Means in each columns and individual plant response , fallowed by at least one letter in common are not significantly different at 5 % level of probability- using by Duncan's multiple range test.

Table 2. Residual Zn and Cd interaction on Cd concentration (µg /g plant) in spinach

Cd levels (µg /g soil)	Zn levels (µg /g soil)			Mean
	0	5	10	
0	0.73 d	0.62 d	0.6 d	0.65 B
3	8.30 a	7.50 b	6.6 c	5.60 A
Mean	4.51 A	4.06 B	3.6 C	

*Means, fallowed by at least one letter in common (small letters for interaction and capital letters for main effects) are not significantly different at 5 % level of probability- using by Duncan's multiple range test.

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The Leaching Potential of Heavy Metals From Compost, Sewage Sludge, Fly Ash and Slag

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Abstract
The aim of this study is to determine the leaching potential of heavy metals from several materials. The materials used in this study were fly ash and slag taken from a site close to the Muğla Yatağan Thermal Power Plant and sewage sludge taken from İzmit Advanced Biological Treatment Plant, compost taken from ISTAÇ A.Ş. For this purpose, after determining the physical and chemical characteristics of materials, the Toxicity Characteristic Leaching Procedure (TCLP) (US EPA Method 1311) and the Synthetic Precipitation Leaching Procedure (SPLP) (EPA 1312) were applied to all materials to observe the risk of heavy metal leackage to soil and groundwater. The heavy metal content of materials and their leaching potential has been evaluated. According to the results, the SPLP (pH 5), SPLP (pH 4,2) and TCLP values for sewage sludge for example were found to be 37,6 mg Zn kg^{-1}, 175,8 mg Zn kg^{-1} and 379,7 mg Zn kg^{-1}, respectively. Considering heavy metal content of sludge, the measured values are lower then the limit values given by the Turkish Regulation on "The Use of Domestic and Urban Sewage Sludge in Soil." Regarding the findings, there is no drawback to apply this sludge to soil. In general, heavy metal levels measured in the TCLP and SPLP solutions did not exceed 75.94 mg kg^{-1}.
Keywords: Heavy metals, leaching potential, TCLP, SPLP.

Introduction
Generators of solid waste frequently try to find alternative disposal methods and thus request permission from regulatory agencies to use their waste materials in a more beneficial way. One form of beneficial use often proposed for some types of solid wastes are their use as a soil conditioner. Compost, sewage sludge, fly ash, industrial waste and slag are frequently used as soil conditioners. Addition of organic conditioners such as compost, sewage sludge, bark chips and woodchips to soil has been used for many centuries to improve soil fertility, enhance revegetation and decrease the plant availability of toxic metals (Williamson and Johnson, 1981; Abbott et al., 2001; Gadepalle et al., 2007). The addition of organic amendments to soils can reduce the heavy metal bioavailability by changing them from bioavailable forms to the fractions associated with organic matter or metal oxides or carbonates (Walker et al., 2004). Organic amendments can also improve soil aeration, water and nutrient holding capacities. The use of compost (municipal solid waste compost, biosolid compost, mature compost, cow manure, etc.) for restoring heavy metal and arsenic contaminated soils has also been reported by various researchers (Cao et al., 2003; Cao and Ma, 2004; Clemente et al., 2003; Cala et al., 2005; Walker et al., 2004; Kiikkila et al., 2002; Rate et al., 2004; Gadepalle et al., 2007). The results represented that the uptake of heavy metals and arsenic by plants was reduced, however an increase in risk to the groundwater due to accumulation of metals in the soil. Fly ash and slag have been regarded as problematic inorganic solid wastes all over the world, Many possible beneficial applications of this materials are being evaluated to minimize waste, decrease cost of disposal and provide valueadded products. The conventional disposal methods for fly ash and slag lead to degradation of arable land and contamination of ground water. However they are useful ameliorant that may improve the physical, chemical and biological properties of problemic soils and may be regarded a source of readily available plant macro and micro nutrients. In conjunction with organic manure and microbial inoculants, fly ash and slag can enhance plant biomass production from degraded soils.

When examining the suitability of a proposed use of waste materials, possible impacts on human health and the environment must be evaluated. The impact is generally evaluated by assessing the risk arising from the used waste. In most cases the total concentration of a given pollutant in the waste is assumed to contribute to the risk. The bioavailability and/or mobility of a pollutant in the waste is generally incorporated into the risk assessment process. Mobility of a pollutant increases the risk of groundwater pollution and thus the risk from the exposure to contaminated groundwater. It is well established that only a fraction of a pollutant will leach from a granular waste and have a

potential to migrate to groundwater. Thus, an estimate of the fraction that leaches must be made. There are several leaching tests which are used for estimating the leachability of contaminant. The TCLP was adopted by the U.S. EPA to identify wastes that have the potential to contaminate groundwater with a number of regulated elements based upon disposal in a municipal solid waste landfill. The SPLP does not have a specific regulatory application, but simulates the extraction of elements from wastes by percolating rainfall.
The aim of this study is to determine the leaching potential of heavy metals from several materials. The materials used in this study were fly ash and slag taken from a site close to the Muğla Yatağan Thermal Power Plant and sewage sludge taken from İzmit Advanced Biological Treatment Plant and the compost taken from ISTAÇ A.Ş. For this purpose, the Toxicity Characteristic Leaching Procedure (TCLP) (US EPA Method 1311) and the Synthetic Precipitation Leaching Procedure (SPLP) (US EPA Method 1312) were applied to all materials.

Materials and Methods

The heavy metal contents and leaching potential of these metals from fly ash and slag taken from a site close to the Muğla Yatağan Thermal Power Plant, sewage sludge taken from İzmit Advanced Biological Treatment Plant and compost taken from ISTAÇ A.Ş were determined.

Characteristics of Materials

Organic matter content of materials was determinated according to the Walkley-Black method (Walkey and Black, 1934). pH and electrical conductvity (EC) values was determinated using a pH-meter and electrical conductivity probe. The properties of materials are given in detail in Table 1.

Table 1**.** Some physical and chemical properties of the materials

Sample	Organic Matter Content (%)	pH	Electrical Conductivity (mS/cm)
Compost	46.97	7.05	3.20
Sewage sludge	55.88	6.43	3.58
İndustrial waste	5.30	8.52	1.73
Slag	1.53	11.40	4.18
Fly ash	4.14	9.64	0.95

Heavy Metal Content of Materials

The digestion of sewage sludge and compost were accomplished using combinations of HNO_3-H_2O_2 solutions as described in the U.S. EPA Method 3050B (U.S. Environmental Protection Agency, 1996). Fly ash, slag and industrial waste were digested using HF-$HClO_4$-HCl solutions as described by Tessier et al. (1979).

Leaching Tests

Tests on leaching characteristics of materials were conducted according to the standard EPA methodologies, i.e., TCLP and SPLP (Methods 1311 & 1312). Respectively, the TCLP (U.S EPA, 1997) is a test used to determine toxicity of wastes in a highly buffered environment. Extraction fluid No. 1 (pH = 4.93 ± 0.05) was used for wastes with pH 5 or under, and extraction fluid No. 2 (pH = 2.88 ± 0.05) for wastes with pH above 5. The SPLP (U.S EPA, 1997) is a test designed to simulate acid rain. The conditions pH = 4.2 ± 0.05 are less stringent than in the TCLP test. For both tests, extraction was carried out at 1:20 solid to liquid ratio. Diluted acetic acid was used as the extraction fluid for TCLP and a 60/40 weight percent mixture of sulfuric and nitric acid was used for the SPLP method. Leachates collected from each test were analyzed by AAS to determinate Cu, Zn, Pb and Cd concentrations.

Results and Discussion

Heavy metal content of materials

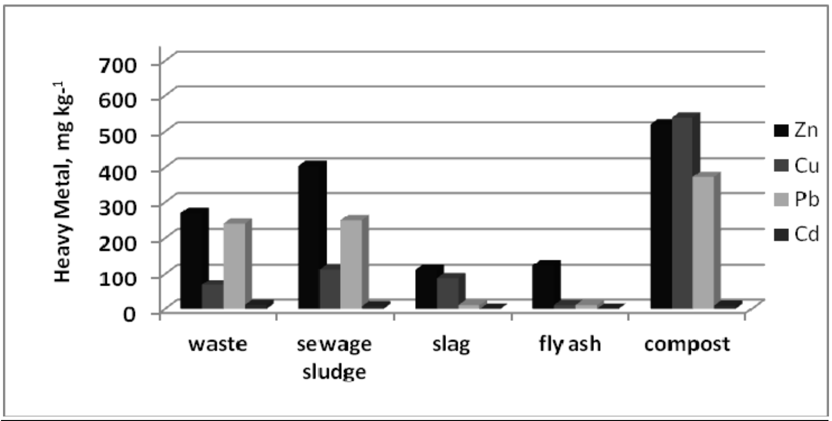

Fig.1. Heavy metal content of materials

As can be seen from Figure 1, the metal content of materials were variable. Zn was almost the highest measured metal in all materials. The highest Zn content was observed in compost, reaching a value of 500 mg kg^{-1}. Similarly, the highest copper and lead concentrations were determined in compost, reaching levels of 540 mg kg^{-1} and 375 mg kg^{-1}. Copper levels in the other materials were below 113 mg kg^{-1}. The lead content of waste and sewage sludge were close about 240 mg kg^{-1} and 250 mg kg^{-1}, respectively. The lead level in fly ash and slag was about 10 mg kg^{-1}. Fly ash and slag did not contain cadmium and the cadmium content of waste and sewage sludge was below 12 mg kg^{-1} and 8 mg kg^{-1}, respectively.

Regarding the Turkish Regulation on “The Use of Domestic and Urban Sewage Sludge in Soil” (official gazete no:27661, 2010) he metal content of analysed sewage sludge did not exceed the limit values, which were given as 2500 mg Zn kg^{-1}, 1000 mg Cu kg^{-1}, 750 mg Pb kg^{-1}, 10 mg Cd kg^{-1}. The lead and cadmium content of compost was slightly higher than the common reported values for municipal solid waste compost (Orkun et. al., 2010).

Leaching Potential of Heavy Metals

The heavy metal concentration determinated in the SPLP solution with pH=4.2 and pH=5 are given in Figure 2 and 3. The results are given as mg kg^{-1} to enable the comparison of the level of metal leached with the heavy metal content of all mateials.In general, Zn present at highest levels in the materials, presented a high leaching potential. The potential of copper to leach was especially found to be related to the organic matter content of materials, sewage sludge and compost representing the highest concentrations when these concentrations are compared with the metal content of materials, it can be stated that the proportion of cadmium determined in the SPLP solutions of waste and sewage sludge was about 21-22% and 13-14%, respectively. The propotion of zinc leached was between 0.05-8.7% in general. There was no copper and cadmium leakage from fly ash and slag.

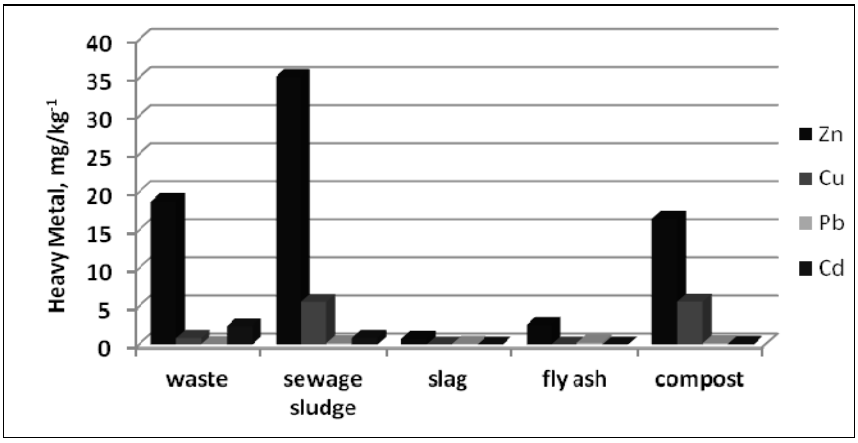

Fig.2. Heavy metal concentration leached with the SPLP pH=4.2 solution

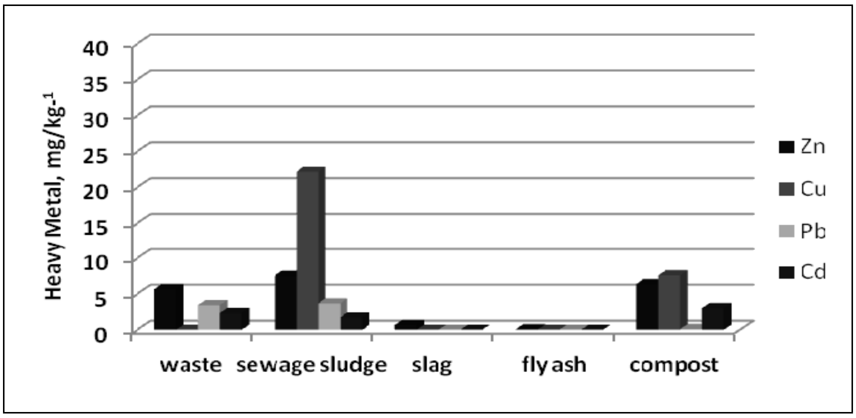

Fig.3. Heavy metal concentration leached with the SPLP pH=5 solution

According to Figure 2 and Figure 3, when the concentrations in SPLP solutions are considered, waste, sewage sludge, slag, fly ash and compost can be ranked in the following order; sewage sludge > waste > compost > fly ash > slag for SPLP tests. pH is an important parameter affecting the leaching rate of metals from materials to leaching solutions (Baba, 2000). Generally, leaching potential and velocity of inorganic compounds from fly ash and slag are increasing at lower pH values (Topal et. Al, 2011). In this study, we determined that values of SPLP pH=4.2 were higher than SPLP pH=5 values for fly ash and slag, too.

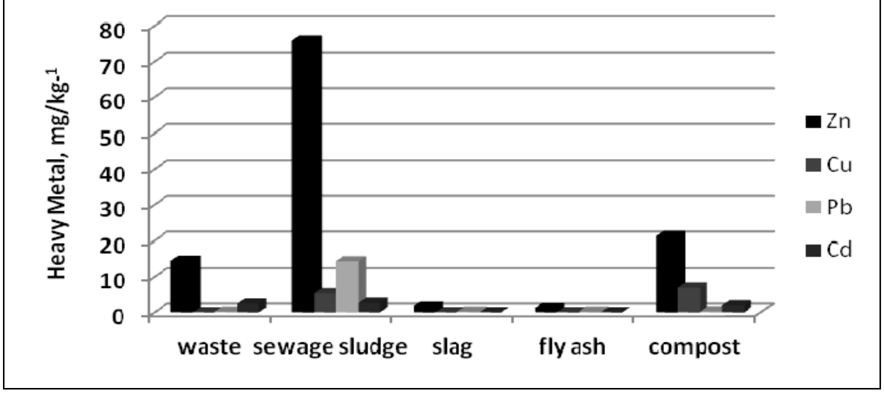

Fig.4. Heavy metal concentration leached with the TCLP solution

The heavy metal concentrations measured in the TCLP solution (pH=5) are presented in Figure 4. Similarly, among the heavy metals Zn concentration were very high, reaching levels of 75mg kg^{-1} for sewage sludge. Copper was not determined in the TCLP solutions of waste, fly ash and slag. A higher leaching potential of metals was generally found fort he sewage sludge. On the other hand the lowest leaching potential was found for fly ash and slag. Considering the heavy metal contents of material, cadmium similarly reflected the higher proportion, which was 22%, 38% and 23% for the waste, sewage sludge and compost, respectively. In general, the leaching potential with respect to the metal content followed an order of Cd > Zn > Pb > Cu for TCLP test. SPLP values were lower when the TCLP pH=5 and SPLP pH=5 results were compared. Cao and Dermantas (2008) explained this case with less aggressive characteristic of SPLP solutions.

Conclusion

In this study, leaching behaviour of some heavy metals from different materials was investigated with TCLP and SPLP tests. Results of analyses showed that TCLP method is more effective than SPLP method to determine the heavy metal leachability. In general, the heavy metal levels observed in the TCLP and SPLP solutions did not exceed 75.94 mg kg^{-1}. The heavy metal leaching potential of materials followed an order of waste > compost > fly ash > sludge in terms of pollution.

Considering the disposal of the materials in a landfill, several limit values have to be met. These limit values are given in the Turkish Regulation of Waste Disposal in Sanitary Landfills. (Official Gazette No: 27533)(2010).

Table 2**.** Comparison of hazardous wastes landfill criterias with some heavy metal concentrations which came into by wastes leaching

Metal	Limit Values (mg l^{-1})	Waste		Sewage Sludge		Slag		Fly ash		Compost	
		TCLP	SPLP	TCLP	SPLP	TCLP	SPLP	TCLP	SPLP	TCLP	SPLP
Zn	0.40	0.72	0.94	3.80	1.75	0.08	0.03	0.06	0.1	1.07	0.82
Cu	0.20	0.00	0.04	0.27	1.10	0.00	0.00	0.00	0.00	0.35	0.37
Pb	0.05	0.01	0.17	0.72	0.18	0.01	0.005	0.01	0.01	0.01	0.01
Cd	0.004	0.12	0.12	0.13	0.08	0.00	0.00	0.00	0.00	0.10	0.15

As can be seen in Table 2, some heavy metal concentrations seem to exceed the limit values of regulation. However, it should be taken into account that these values are considered for a leaching test with a solid to liquid ratio of 1:10. Stil, some of the observed heavy metal concentrations for some materials are significantly high indicating to the risk of ground and surface water pollution. So, short-long term precautions should be taken before disposal or storage of these materials, usage as soil amendment.

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Evaluation of Air Pollution Tolerance Index (APTI) In Some Plant Species in an Industrial Area

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Abstract
Air pollution has harmful effect on human health and vegetation. Plant species absorb pollutants and accumulate them in their leaves. Different plant species have various reactions to air pollution. In the recent years, using of plant species to reduce air pollution has been increased significantly. The aim of this study is to determine tolerant species to air pollution. To approach this goal, four biochemical parameters of four plant species, growing in an industrial area, such as leaf relative water content (RWC), ascorbic acid (AA) content, total leaf chlorophyll (TCh), and leaf extract pH, was calculated to compute air pollution tolerance index. The results showed that *Cupressus arizonica, Robinia pseudoacacia, Pinus eldarica, Quercus brantii* respectively are tolerant, moderately tolerant, intermediate and sensitive tree species to air pollution.
Keywords: Air pollution tolerance index- Relative water content -Total chlorophyll content –pH- Ascorbic acid

Introduction

Air pollution is a main problem ascending mainly from industrialization (Odilora et al., 2006). Trees performance as a sink of air pollutants and this reduce their concentrations in the air (Prajapati and Tripathi, 2008). When exposed to airborne pollutants, Most of plants experienced physiological changes before presenting visible injury to leaves (Dohmen et al., 1990). Plants vary significantly in their responses to pollutants, some being highly sensitive and others tolerant (Singh and Rao, 1983; Shannigrahi et al., 2003). Lack of enough knowledge about tolerant and sensitive species to air pollution has caused the studies to identify sensitive and tolerant species to air pollution (Prajapati and Tripathi., 2008).
Air pollution tolerance index (APTI) based on four biochemical parameters including leaf relative water content (RWC), ascorbic acid (AA) content, total leaf chlorophyll (TCh) and leaf extract pH has been used for categorizing tolerance of plant species to air pollution (Escobedo et al., 2008; Singh et al., 1991).
In this study, we target to determine tolerance of 4 tree species growing in the botanical garden of Isfahan Mobarekeh steel company of Iran by using the APTI method.

Materials and Methods

Leaf samples were collected from 4 tree species growing in botanical garden of Isfahan Mobarekeh steel company of Iran. 4 biochemical parameters such as leaf extracts pH (Sing and Rao, 1983), relative water content (RWC) (Sen and Bhandari, 1978), total chlorophyll (TCh) (Arnon, 1949), ascorbic acid (AA) (Keller and Schwager, 1977**)** were estimated in the leaf samples.
The air pollution tolerance index (APTI) was determined by using the following formula (Singh and Rao, 1983).
$APTI = [A (T+P) + R] \div 10$
A = Ascorbic acid content (mg/g f.w)
T = Total chlorophyll (mg/gf.w)
P = pH of leaf extract
R = Relative water content of leaf %
APTI value was divided into four grades of Air pollution tolerance referring to study of Liu and Ding (2008).

Results and Discussion

4 biochemical parameters in 4 tree species showed significantly difference at $\alpha = 0.05$. The average RWC varied from 73.82 mg/g FW for *Cupressus arizonica* to 31.84 mg/g FW for *Quercus brantii*. The lowest average of pH recorded by *Quercus brantii and* the highest by *Cupressus arizonica*. *But Cupressus arizonica* didn't has significance difference with *Robinia pseudoacacia.* It was

reported that Plants with lower pH are more susceptible, while those with pH around 7 are more tolerant (Singh and Verma, 2007).
The highest Total chlorophyll content recorded for *Cupressus arizonica.* Higher chlorophyll content in plants might favor tolerance to pollutants (Joshi et al., 1993).
Robinia pseudoacacia had Maximum content of Ascorbic acid. Ascorbic acid; a natural antioxidant in plants has been shown to play an important role in pollution tolerance (joshi and swami, 2007).

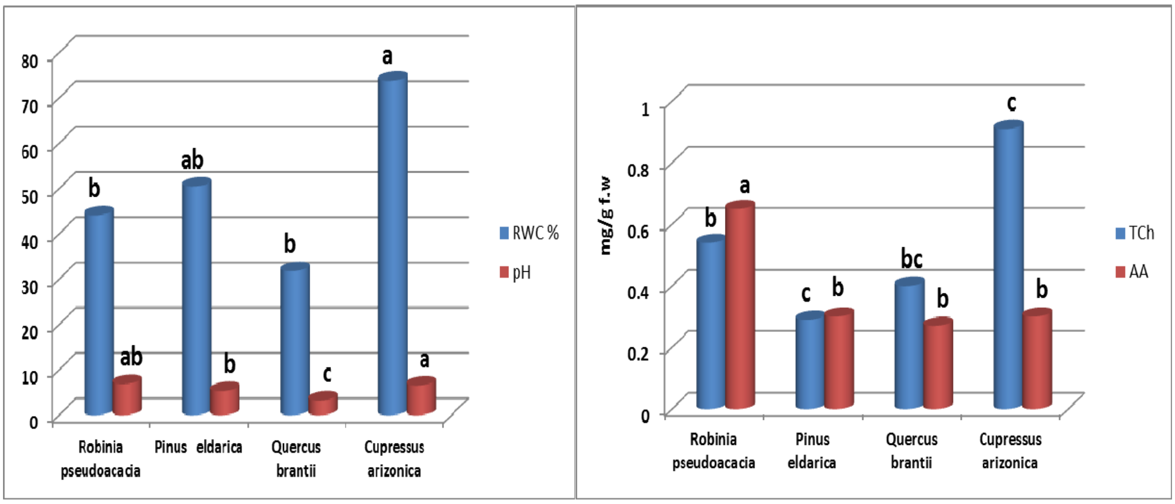

Fig. 1 leaf relative water content (RWC), ascorbic acid (AA) content, total leaf chlorophyll (TCh), and leaf extract pH in leaves of tree species

APTI value in 4 tree species showed significantly difference at $\alpha = 0.05$.The average APTI ranged from 9.67 mg/g FW for *Cupressus arizonica* to 4.18 mg/g FW for *Quercus brantii*. The gradation of APTIs was calculated referring to section 2.7. *Cupressus arizonica, Robinia pseudoacacia*, *Pinus eldarica, Quercus brantii* respectively were categorized as tolerant, moderately tolerant, intermediate and sensitive tree species to air pollution. Tolerant species can be used for expansion of greenbelt in urban and industrial areas.

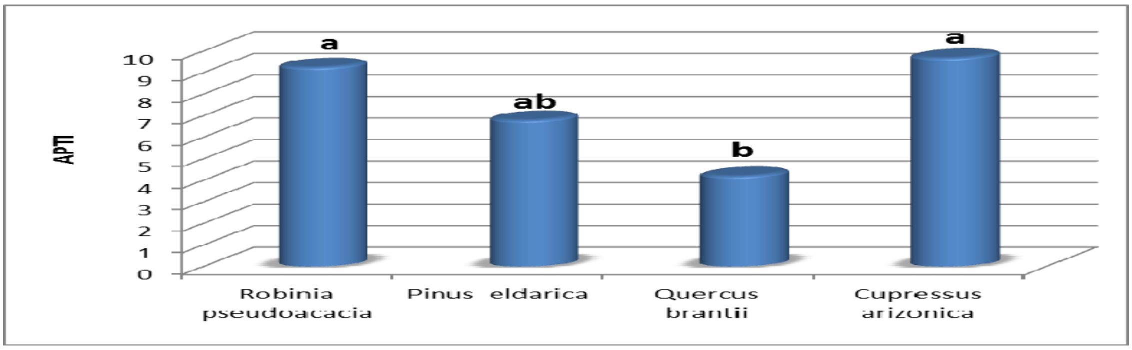

Fig. 2 air pollution tolerance index (APTI) in tree species

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The authors wish to thank *Islamic Azad University, Khorasgan Branch, College of Agriculture, Department of Soil Science* for providing necessary laboratory facilities.

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Evaluation of Zn, Ni, Cu and Pb Contamination in Some Tree Species in an Industrial Area in Esfahan

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Abstract
Industrial development leads to increase concentration of heavy metal in air and soil of industrial areas. Heavy metal components are unique toxicant because of their sustainability in the environment. So heavy metals have a potential of threat for many years. The effective method for cleaning contaminated sites is phytoremediation. It is an innovative technique that utilizes plant species to remove contaminants from the environment. Plants have ability to take up metals from air and soil and accumulate them in above ground tissues .The aim of this study is to estimate the concentration of zinc, copper, nickel and lead in leaves of eleven plant species of an industrial area in Esfahan city. The highest accumulation for Zn was found in Berberis vulgaris. Ni concentration in all plant species were in the range of Pollution level. Pb concentrations in plant species except for Robinia pseudoacacia were higher than normal limit of Pb.
Keywords: Plant species -Heavy metals-Phytoremediation-contaminant

Introduction
Contamination of soil, water, and the atmosphere with heavy metals has impact on plants in addition to all the organisms in their habitat (Monaci et al., 2000; Onder and Dursun , 2006). Plants growing in the nearby industrial areas show increased concentration of heavy metals (Mingrance et al., 2007). Furthermore to roots, heavy metals are taken up by plants from the air or foliar precipitation (Onder and Dursun , 2006). Biological methods can substitute with physical methods in the places that are limited to detect air pollution (Aksoy and Ozturk , 1997). Bio monitoring with plants is low-cost and valuable method to assess the influence of different air and environment pollutants (Oliva et al., 2007). Leaves of higher plants have been used for bio monitoring of heavy metals since the 1950s (Al-Shayeb et al., 1995). Higher plants can be used as bio monitors in areas that do not have these species (Sawidis et al., 1995; Aksoy and Ozturk, 1997; Rossini Oliva and Mingorance, 2006). Chemical foliar analysis has also been used to study the impact and level of air pollutants (Djingova et al., 1999; Ericsson et al., 1995; Hüttl and Fink, 1991). The aim of this present work was to determine plant species with highest concentration of heavy metals.

Materials and Methods
Leaf samples were collected from 4 tree species growing in botanical garden of Isfahan Mobarekeh steel company of Iran. The climate of the area is arid and the annual precipitation is 140 mm.
To determine the total heavy metal content, both that taken up by the roots and translocated to the leaves and that found on the leaf surface, leaf samples were not washed. Then All samples were dried in an oven at 70°C during 48 h, then milled, crushed to pass a 1.5 mm sieve and one gram digested with HCl (2 M).The concentration of Zn, Ni, Cu and Pb in leaf samples were determined by flame atomic absorption spectrophotometry.
All statistical analyses were performed using the Statistical Package for the Social Sciences (SPSS) version 19 software (SPSS Inc.). Data were also subjected to analysis of variance (ANOVA) and differences between means were determined using the Duncan test.

Results and Discussion
Leaf Zn concentration in plant species ranged from 23.42 to 236.5 mg kg^{-1} DW. Zn concentration was the highest in *Berberis vulgaris* which differed significantly from other species. This concentration was higher than the standard levels of Zn in plant dry matter (5–100 mg kg-1 dry matter) (Herrick and Friedland,1990).

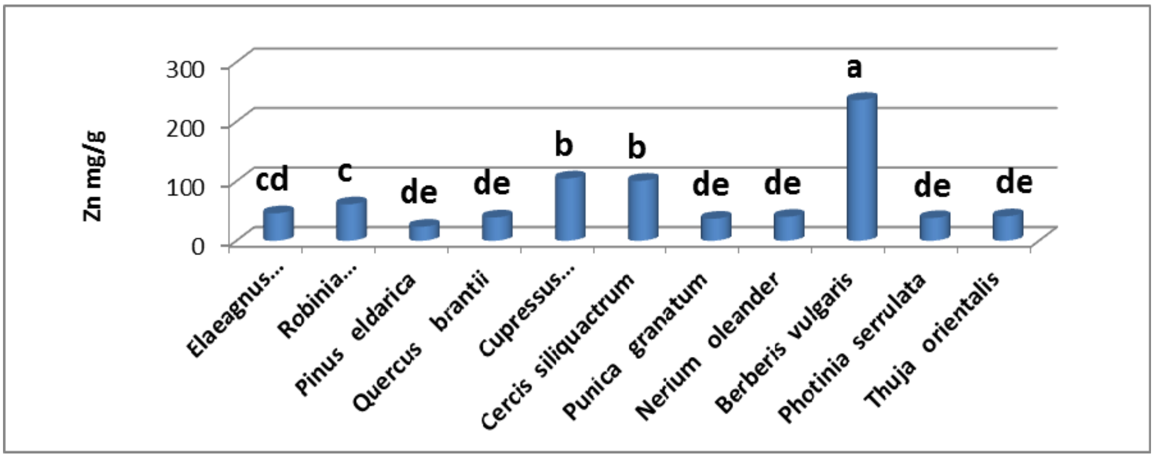

Fig. 1 Concentrations of Zn (mg kg−1 DW) in dry tissues of leaves

For Cu concentrations in leaves, ANOVA analysis did not revealed a significant species variation. Cu concentration ranged from 12.2 to 30.67 mg kg−1 DW. Cu concentrations in *Robinia pseudoacacia, Thuja orientalis, Cercis siliquactrum, Punica granatum, Quercus brantii, Elaeagnus angustifolia were* slightly *higher* than the standard levels of Cu in plant dry matter (2–20 mg kg^{-1} dry matter) (Herrick and Friedland, 1990).

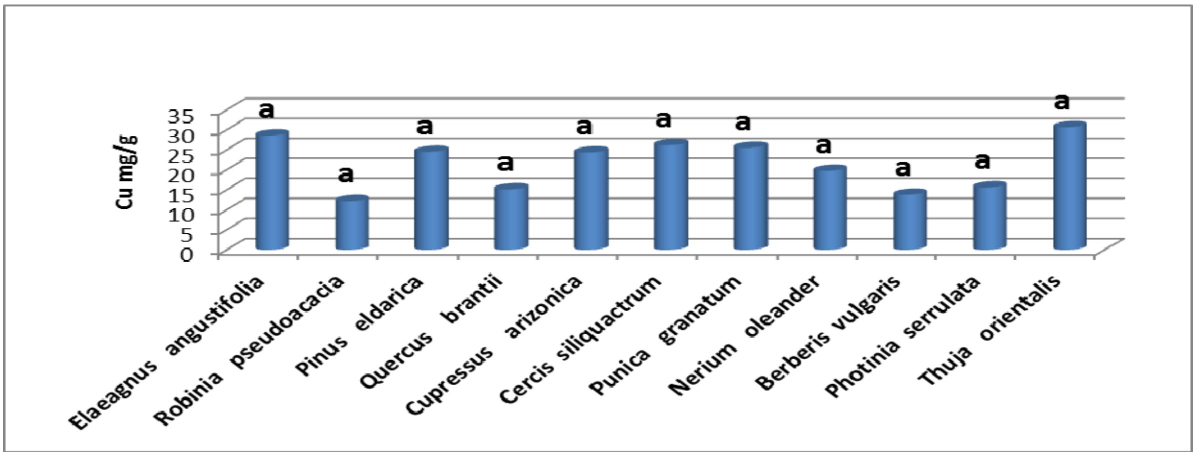

Fig. 2 Concentrations of Cu (mg kg^{-1} DW) in dry tissues of leaves

Pollution level of nickel was 3-50 in plant dry matter (Herrick and Friedland, 1990).Leaf Zn concentration in plant species ranged from 18.39 for Punica granatum to 40.65 mg kg−1 DW for Nerium oleander .So these concentration were in range of Pollution level of nickel.

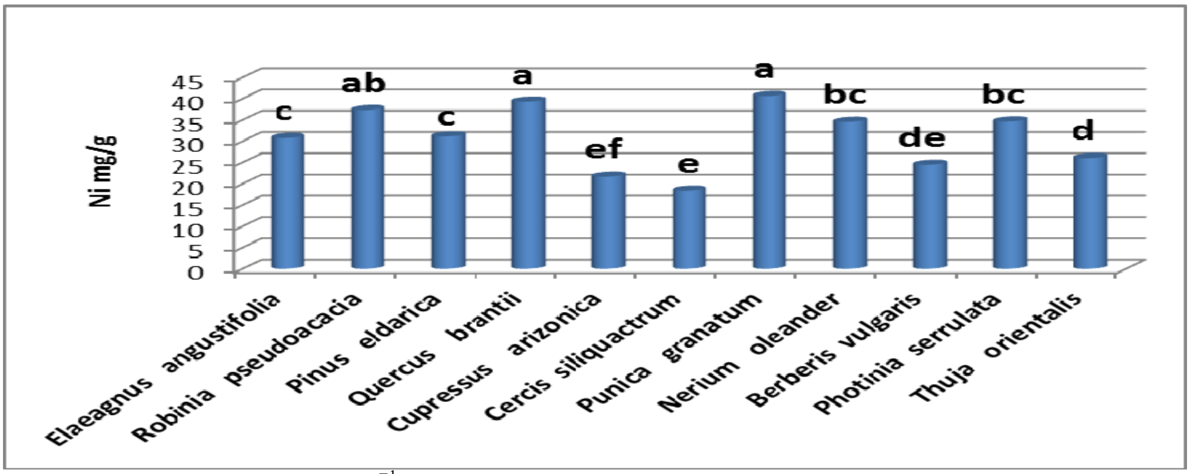

Fig. 3 Concentrations of Ni (mg kg^{-1} DW) in dry tissues of leaves

According to earlier studies natural, normal and toxic limits of Pb for plants are 3, 10, and 30-300 mg/kg D.W., respectively (Allen et al., 1984; Kloke et al., 1984; Kabata-Pendias and Piotrowska, 1984). Leaf Pb concentration in plant species ranged from 7.59 to 28.52 mg kg−1 DW. Pb concentrations in plant species except for Robinia pseudoacacia were higher than normal limit of Pb.

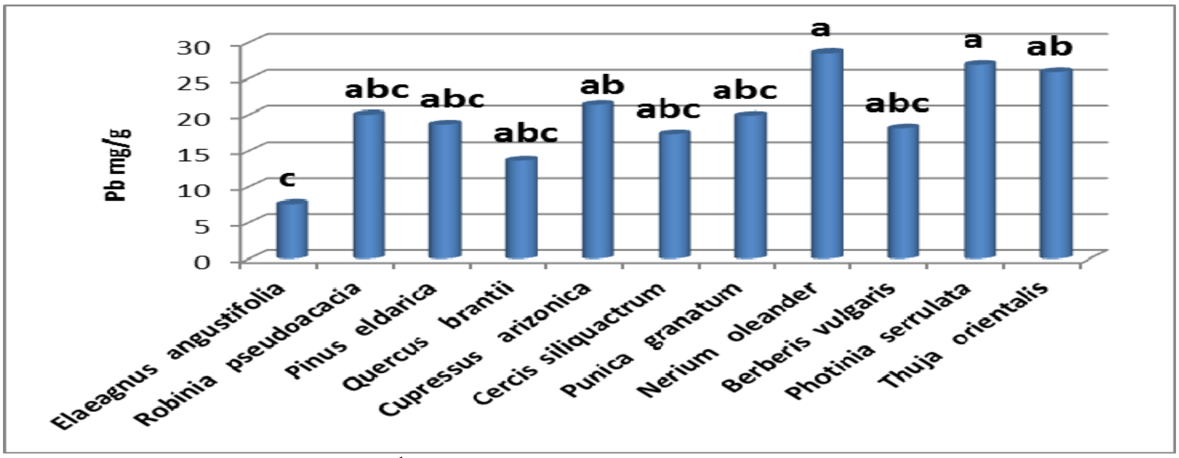

Fig. 4 Concentrations of Pb (mg kg^{-1} DW) in dry tissues of leaves

Turkdogan et al. (2002) also reported high concentrations of Cu, Co, Pb, and Cd in crops growing in the nearby industrial areas. Plant species with higher accumulating of heavy metal recommended for planting in industrial areas.

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Distribution and fractionation of zinc in a calcareous Sandy loam soil by sodium acetate receiving seven treatment

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Abstract

The objective of this study is investigate the effect of three organic amendment usage on soil chemical properties and uptake of heavy metals by soil. The experiment was carried out in a completely randomized block design with seven treatments including : no fertilizer as a control (C), sewage sludge (SW), cow manure (CM), municipal solid waste compost (MC), mixture of sewage sludge and cow manure (50% SW + 50% CM), mixture of sewage sludge and municipal solid waste compost(50% SW + 50% MC) and mixture of cow manure and municipal solid waste compost (50% CM + 50% MC) at three levels of 0, 2.5 and 5 kg/shrub and three replicates in calcareous sandy loam soil at the botanical garden of Mobarekeh steel company. After 2 years, soil samples were collected from 30 cm depth and leaves assemblage. Soil chemical properties such as Soluble/ exchangeable, organic-bound, inorganic precipitates and residual Zn in the soil were sequentially extracted and analyzed by sodium acetate. Results showed The addition of organic amendments declined significantly the concentrations of soluble/ exchangeable Zn, but increased the amounts of these metal in organic-bound and inorganic precipitate forms in the soil.The conversion of soluble/exchangeable Zn to other insoluble forms after the application of organic amendments.

Key words: Fractionation, heavy metals, Organic amendments, soil chemical properties,soil

Introduction

Land application of organic amendments like sewage sludge, municipal solid waste compost and animal manure is an excellent way of recycling both the nutrients and the organic matter contained in them. Apart from the agricultural use, this practice is becoming one of the most promising ways for the reclamation of soils with low organic matter content. However, the potential health risks associated with the presence of pathogens, heavy metals and organic pollutants are well known, as well asthe short and long term effects that these contaminants have on soil, from the agronomic point of view (Sastre et al., 1996; Albiach et al., 2000; and Vasseur et al., 2000). influence of organic substances on the availability of the heavy metals depends on the nature of these metals, soil types, and the organic matter properties, particularly the degree of humification (Walker et al., 2004).Organic matter has a vital role in controlling the mobility of heavy metals in soils. It may decrease the available concentrations of heavy metals in soils by precipitation, adsorption, or complexion processes (Bernal et al., 2007)remediating effects of different organic wastes on heavy metal-polluted soils over the past years. For example, the addition of mushroom compost to a clay loam soil in Ankara, Turkey decreased the concentration of available Zn from 2.20 to 1.90 mg kg^{-1} and Cd from 0.057 to 0.005 mg kg^{-1} (Karaca,2004). Incorporation of pig manure to a paddy soil in China reduced the concentrations of available Zn and Cd by 76.1% and 25.7%, respectively (Li et al., 2008). In a calcareous soil in Murcia, Spain, the amount of available Zn was slightly reduced from 11.3 to 10.4 mg kg^{-1} after the addition of cattle manure (Walker et al., 2003). Application of chicken manure compost in a Ferralsol, China decreased the concentration of soluble/exchangeable Zn by 71.8–95.7%, but increased the values of inorganic precipitated Cd by 0.6–1.5 times and organic-bound Zn by 0.9–7.8 times (Liu et al.,2009). Thus, the aim of this experiment is to study the effects of Residual application of three sources of organic matter: sewage sludge, municipal solid waste compost and cow manure applied to a calcareous soil on pH, electrical conductivity (EC), percentage of soil organic matter (OM%)‹total concentration and DTPA–extractable Zinc and fractionation of Zn in a calcareous sandy loam soil by sodium acetate receiving seven treatments.

Table 1. Some chemical and physical properties of soil

Texture	N (%)	AP (mg.kg^{-1})	AK (mg.kg^{-1})	OM (%)	$CaCO_3$ (%)	CEC (Cmol/kg)	EC (dS/m)	pH (1:2)	Depth (cm)
Sandy loam	0.08	79.9	459	0.7	60	9.26	1.01	8.06	0-30

Materials and Methods

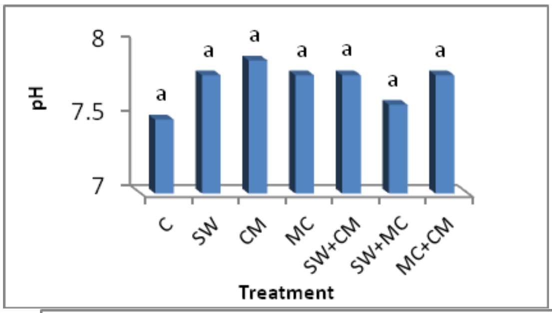
The experiment was conducted at the botanical garden of Isfahan Mobarekeh steel company of Iran. The soil of the experimental plots was Typic Haplocalcids (USDA, 2006). This experiment was carried out in a completely randomized block design with seven treatments including: no fertilizer as a control (C), sewage sludge (SW), cow manure (CM), municipal solid waste compost (MC), mixture of sewage sludge and cow manure (50% SW + 50% CM), mixture of sewage sludge and municipal solid waste compost (50% SW + 50% MC) and mixture of cow manure and municipal solid waste compost (50% CM + 50% MC) and three replicates in calcareous sandy loam soil. Starting in January 2009, several rates (0, 2.5 and 5 kg/shrub) for each treatment of biosolids were applied in state of localized fertilization in distance of 50 cm from each shrub to plots of 24 × 38 m size and then done in ten irrigation periods.

Table 2. Some chemical properties of organic biosolids

Property	Unit	Sewage sludge	Municipal solid waste compost	Cow manure
pH	-	7.0	8.1	7.9
EC	ds/m	6.0	10.2	12.3
CEC	Cmol/kg	40.2	44.3	21.5
OM	%	48.7	42. 5	38.5
Zn	mg/kg	1460	588	66.3

Results and Discussion

Effect of biosolids on some chemical soil properties:
Application of the Residual biosolids decreased the EC of soil significantly (P=0.001) compared to control soil (Figure1). Among treatments only CM had not significant effects on soil pH(Figure 2). Maximum OM% of soil was attributed to CM treatment (Figure 3).



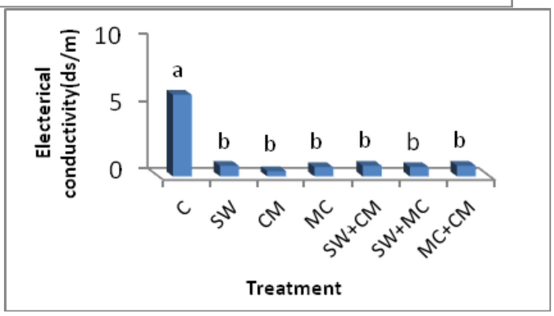


Figure 1. Effect of Residual biosolids application on soil electrical conductivity

Figure 2. Effect of Residual biosolids application on soil pH

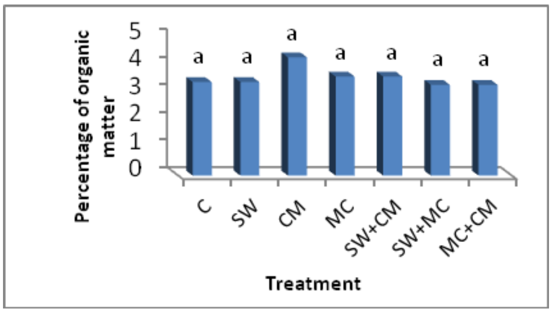


Figure 3. Effect of Residual biosolids application on percentage of organic matter.

Effect of Residual biosolids on Zinc concentrations in soil Total concentrations:

Residual Biosolids application significantly increased the total concentrations of metals in soil (Figure 4).

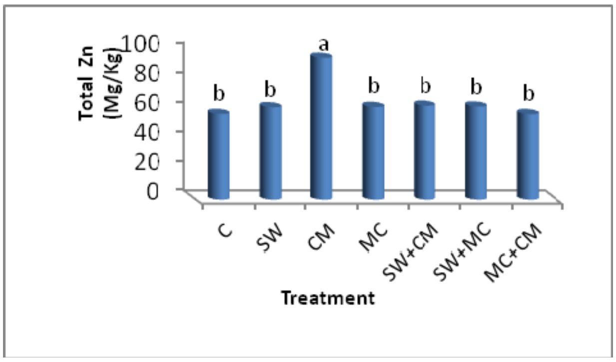


Figure 4. total Zn concentrations in soil

DTPA-extractable concentrations:

Residual Biosolids application not significantly increased the DTPA extractable soil Zn and maximum rates of them showed in municipal solid waste compost treatments, respectively (Figure 5).

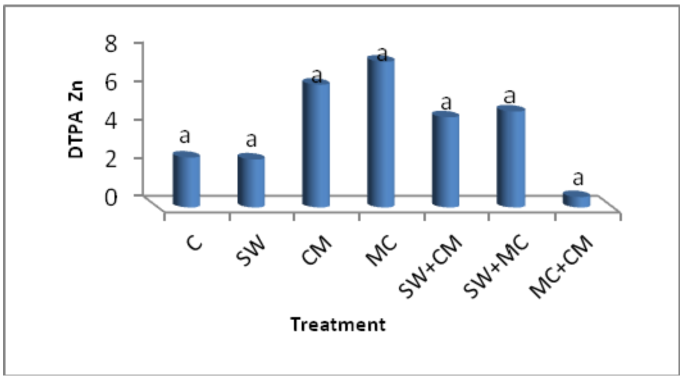


Figure 5. DTPA- extractable Zn concentrations in soil

Fractionation of zinc by sodium acetate with seven treatments:

As shown in Figure 6, the application of seven treatment in the soil significantly reduced the concentrations of Zinc. results showed that in the treated soil, a major proportion of Zn, was associated with ‘carbonate’ Zn fractions included non-labile forms of Zn. Maximum concentration of zinc in soil was attributed to no fertilizer as a Control(C)and mixture of municipal solid waste compost and cow manure treatments(MC+CM).

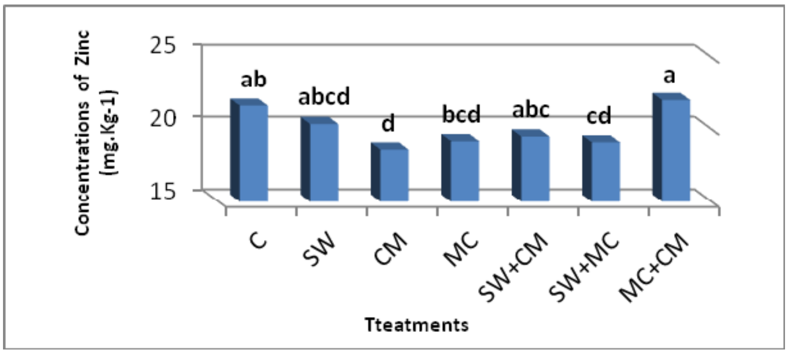


Figure 6. Concentration of Zn influenced by different Treatment

Cow manure application increased soil pH.This mainly was because of the high buffering capacity of this calcareous soil. The result was inconsistent with results of previous studies (Karami et al.,2009; Harding et al.,1984).Increased soil pH is regarded as a major advantage when MSW compost is used Mkhabela and Warman(2005).For example, increases in soil pH from5.8 to 6.7 and 6.1 to 6.5 (Zhang et al., 2006). The increase in the pH of soil may be due to the mineralization of carbon (Mkhabela and Warman, 2005).The EC of the soil increased significantly with Residual biosolids application and maximum value of EC was attributed to control treatment because remarkable salts and frequent application in agriculture soil causes the accumulation of salt in soil (Eghbal et al., 2004).The increase in organic matter after Residual biosolids application could be explained by the large amount of organic matter Pais and Benton Jones(1997). organic amendments application significantly increased the total concentrations of metals in soil (Figure 4), the total Zn concentrations were below the respective limits of maximum acceptable concentrations (MAC) in agricultural soils for these metals in countries such as Germany, Canada and Poland Pais and Benton Jones (1997).However, the concentrations of total Zn exceeded the MAC for Zn in England (150 mg/kg) in sewage sludge treatment. The addition of Residual biosolids significantly increased the DTPA extractable soil Zn compared to control due to higher metal contents (Figure 5). Some authors also reported similar findings (Jordao et al.,2003; Karami et al., 2009; and McGrath et al., 2000).One aspect which should be taken into account is that the addition of sludge not only results in an increase in the total concentrations of metals,but also in the quantity of organic matter, which may have a direct effect on their solubility and bioavailability (McBride et al.,1997).The highest binding of Zn with soil organic matter fractions were recorded in mixture of municipal solid waste compost and cow manure treatments(Tsai et al.,1998).

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Fractionation of lead in contaminated soil by manure and sequential extraction

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Abstract

This study was conducted to evaluate the degree of mobility and fractionation of lead (Pb) after the addition of cow manure (CM) in a sandy loam soil. Treatments were (1) soil application of CM, (2) control soil. The CM application represented a dose of 7.5 Kg per hectare in sandy loam soil at thebotanical garden of Mobarekeh steel company in Esfehan, Iran. After 2 years, soil samples were collected from 30 cm depth and leaves assemblage.An analytical procedure involving sequential chemical extraction has been developed for the partitioning of particulate trace metal such as lead(Pb) into six stage: exchangeable, bound to carbonates, bound to Mn and Fe-Oxides,organic matter and Residual. Sequential extraction results showed that in the treated soil, a major proportion of Pb, was associated with Residual (RES), Some Pb was found in the oxide fraction. However, in mineral soils the 'carbonate' and 'oxide-bound' Pb fractions included non-labile forms of Pb.suggesting that application of this CM to a sandy loam soil at the loading rate used here, may pose a risk in terms of groundwater contamination with Pb.

Keyword: sandy loam soil, Heavy metal, Metal mobility, cow manure, sequential

Introduction

Pollution of agricultural soils with heavy metals such as lead (Pb) is still one of the most serious concerns for the functionality of ecosystems in the world. contamination with Pb is mainly caused by mining and smelting Low activities (Cang et al.,2004; Quartacci et al., 2006) and application of industrial effluents as water source for irrigation of crop plants (Rattan et al.,2005; Abbas et al.,2007). Accumulation of Pb in the soils has toxic effects on microorganisms, plants, and ultimately on animals and human health via the food chain (Rulkens et al., 1998). Several physical, chemical, phyto, and engineering techniques have been used for the remediation of heavy metal-polluted soils (Mench et al., 1994; Shuman 1999). The chemical stabilization methods were considered to be themost cost-effective ways to immobilize heavy metals in the soils (Chen et al., 2000). Heavy metal contamination by cow manure application has received much attention because of concerns regarding uptake by plants, accumulation in the soil thus constituting a long-term environmental hazard, leaching from the soil and re-entry into the food chain, or leaching from the soil and contamination of groundwater and surface waters (Kaschl et al.,2002;Wong et al.,2007). It also increases soil organic matter and improves soil fertility. Under these conditions, the use of CM as a land reclamation technique may be a means to revert desertification processes Toribio and Romanya (2006). it is recognized that MSS may contain heavy metals, and its application should be evaluated to avoid potential hazardous effects over the soil or the plants. Previous soil experiments showed that Pb losses in the percolate were low relative to total amounts of metal applied Jalali and Arfania (2010). Thus, the aim of the present study was to investigate the distribution Pb in the soil and change of heavy metal in different fractions by sequential extraction in a calcareous sandy loam soil receiving cow manure (Tsai et al., 1998).

Materials and Methods

This study was conducted in botanical garden of Esfahan Mobarakeh steel company in the way of Split Plot design (for soil test)three replication.Theincontrol(withod fertilizer) and cow manure in the levels of 7.5 kg per tree and 5 kg per shrub in tree replicates were state of localized fertilization in distance 50 cm from each plant species. This garden is located approximately 80 km south west of Isfahan city in central Iran and lies between longitudes 51° 25′ 19.5″ and 51° 25′ 25.2″ E and latitudes 32° 15′ 31.15″ and 32° 15′ 37.6″N.The climate of the area is arid and the annual precipitation is 140 mm.The soil of the experimental plots was Typic Haplocalcids (USDA, 2006).

Table 1. Summary of the sequential extraction steps of Pb fractionation(Tsai et al,1998)

Steps	Fraction	Extractant	conditions
1	Soluble plus exchangeable	1 mol.L−1 $C_2H_3O_2NH_4$	Shake 30 min at 20°C
2	carbonate	1 mol.L−1 CH_3COONa	Shake 5 h at 20°C
3	Mn-Oxide	0.1mol.L−1 $H_3NO.HCl$ at 0.1mol.L−1 CH_3COOH	Shake 30 min at 20°C
4	Fe-Oxide	0.04mol.L−1 $H_3NO.HCl$ at CH_3COOH 25%	Shake 6 h at 96°C
5	Organic-bound	0.01mol.L−1 HNO_3 and H_2O_2 30%	Shake 30 min at 85°C
6	Residual	4 mol L−1 HNO3	Centrifuge 45 min with rpm 3000

Results and Discussion

Effect of control and Residual cow manure concentrations in soil Total and DTPA-extractable concentrations:

Control and Residual cow manure Treatments application not significantly increased the total and DTPA-extractable concentrations soil Pb and maximum rates of cow manure treatment showed in total Pb, respectively (Figure 1).

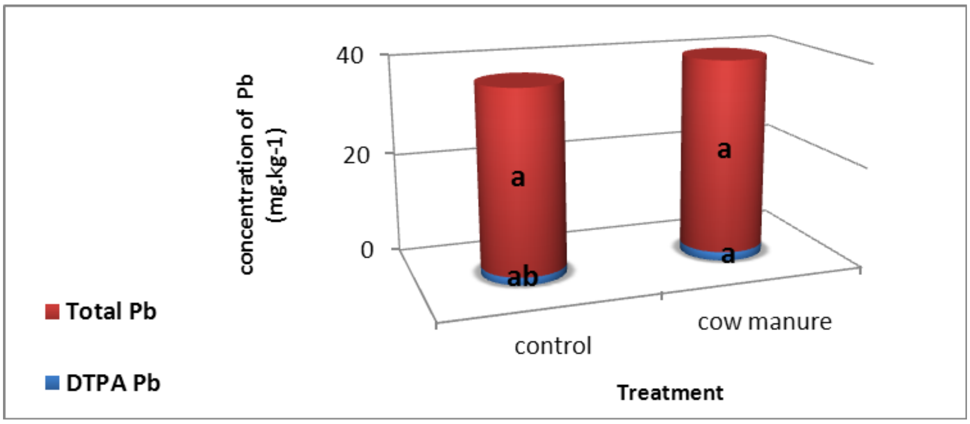

Figure 1. Total and DTPA- extractable Pb concentrations in soil

Table 2. Selected physical and chemical properties of soil and cow manure

Characteristics	soil	cow manure
Textural class	Sandy loam	Sandy loam
pH (1:2)	8.06	7.9
EC (1:2; dS m−1)	1.01	12.3
Organic matter content (%)	0.7	38.5
Extractable Ca^{2+} (meq/l)	4.3	-
Extractable Mg^{2+} (meq/l)	2.71	-
Exchangeable K^{+}(meq/l)	459	0.38
Extractable Na^{+} (meq/l)	2.8	-
Extractable P ($mg.kg^{-1}$)	79.9	0.24
CaCO3 (%)	60	-
CEC ($Cmol.kg^{-1}$)	9.26	21.5
Total N (%)	0.08	1.8
C/N	-	12.4

Effect of control and Residual cow manure on Pb Fractionation in soil:

The soil significantly reduced the concentrations of Fe-Oxide and organic matter Pb as compared to the control. The decrease of soluble/exchangeable Pb was considerable with increasing rates of applied amendments. The amounts of soluble/exchangeable Pb lowered from0.22 mg kg−1 in control(C) to 0.14 in cow manure (CM), respectively. The supply of CM treatment obviously increased the amounts of inorganic precipitate Pb by 0.74-0.92 mg kg−1 respectively. The concentrations of soil residual Pb are illustrated in In general, the addition of CM caused noticeable changes in the amounts of soil residual Pb as compared with C. There are no significant differences noticed with increasing rates of the applied amendments. The highest concentrations of residual Pb were recorded in CM treatment, whereas the lowest values were found in C treatment.

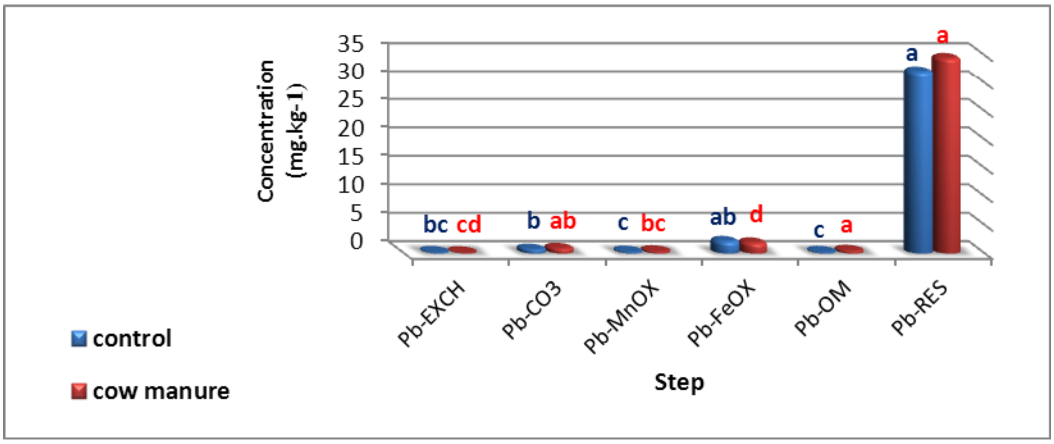

Figure 2. Concentrations of different steps extraction Pb influenced by control and Residual cow manure soil Treatments

The incorporation of cow manure was effective in reducing the solubility of Pb especially at the highest can be used as good applied rates. Also, cow manure and inexpensive substances for the immobilization of Pb in heavy metal-polluted soils. Among different organic materials, cow manure was the most efficient material in contaminated soils. Diminishing the extractability of Pb. In the 0-30 cm soil layers, the proportions of the Pb fractions followed the order RES>>Fe-OX>CO3>Mn-OX >OM>EXCH These findings suggest that Pb were held in a more stable form

(RES) in which the movement of these elements in the soil profile would be negligible and this calcareous soil is able to retain Pb from Residual CM (Tsai et al., 1998).

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Removal of 2,4-D Herbicide from Water by Organo-Modified Iranian Sepiolite

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Abstract
Water pollution by a wide range of organic contaminants such as pesticides is, indeed, a great concern. 2,4-D is one of the this components, which is a herbicide extensively used for control of broadleaf weeds all around the world, due to its low cost and good selectivity. Biological and physicochemical methods have been successfully applied to remove these organic contaminants from water. The physicochemical methods include some based on adsorption by clay minerals. Clay minerals are known to have excellent adsorption properties and therefore they are widely used to remove contaminant from water. However, the negative surface charge of some clays minerals causes them to repel anionic contaminants. Also, these clays are very hydrophilic and consequently show very low adsorption for hydrophobic organic pollutants. By modifying natural clay minerals with quaternary ammonium surfactants, we can improve their sorption capacity for organic compounds. In this study, the removal of 2,4-D that usually exists as anionic forms in aqueous solutions, by natural and modified Iranian sepiolite was investigated. The clay was modified with cationic surfactant N-cetylpyridinium equivalent to the 200% of the sepiolite CEC. Freundlich and Langmuir models were used to obtain sorption parameters. Results showed that both of Langmuir and freundlich isotherms are suitable for modelling the sorption of 2,4-D. The natural sepiolite presented negligible interaction with 2,4-D. Modification of sepiolite, however, enhanced sorption capacity and removal of 2,4-D from aqueous solution. Sorption of 2,4-D is probably due to electrostatic attraction between the anionic adsorbate and the positively charged surfactant molecules.
Keywords: 2,4-D, sepiolite, modification, sorption.

Introduction
2,4-D is a widely used anionic herbicide in agriculture. Since anionic herbicides are very weakly retained by most of the soil components because of their structural negative charge, they remain dissolved in the soil solution and can rapidly move around leading subsequent contamination of surface and ground waters (Ayar, et al., 2008). 2,4-D has been proved to be toxic to human and animals (Garabrant and Philbert, 2002; James and Larry, 2007; Munro et al., 1992). Therefore, its presence in water sources for human and animal consumption is highly objectionable. Consequently, the development of reliable methods for the removal of 2,4-D is of particular worth. Several physical, chemical and biological processes have been evolved so far for the removal of 2,4-D, among them, adsorption by clay minerals has been proven to be an effective and attractive mechanism (Aksu and Kabasakal, 2004). Because of the hydratation of the inorganic cations on the exchange sites, the clay mineral surface is hydrophilic in nature and also due to presence of permanent negative charges on their structural frameworks, natural clays are ineffective sorbents for adsorbing organic and ionic compounds (Prost and Yaron, 2001). Accordingly, in recent years much attention has been paid to the development of modified clays with improved sorption capacities, such as cation-exchanged organo clays by cationic surfactants with a high number of carbon atoms. Some specific alkyl ammonium surfactant cations may impart such favorable surface properties to natural clays in order to enhance their ability for holding anions and organic contaminants. For example, Li et al. (2003) reported improved adsorption of chromate a by HDTMA modified sepiolite and palygorskite as compared to the unmodified clays. Therefore, this study was conducted in order to examine the adsorption of 2,4-D on natural and modified sepiolite and investigate the efficiency of clay surface modification on removal of hydrophobic and anionic contaminants.

Materials and Methods
Sepiolite sample used in this study was obtained from Yazd¸ Iran and used without any purification. The chemical composition of this sepiolite determined from XRF analysis. For XRD analysis, the sepiolite sample was dispersed in the deionized water, dropped on the glass slides and dried in the air. XRD patterns were recorded between 2 and 40° (2θ) at a scanning speed of 2°/min., using a Philips PANalytical X'Pert High Score diffractometer with CuKα radiation.

For preparing Na saturated sepiolite, clay sample was contacted with 1 M NaCl solution for three times. After filtration, the sodium form of clay was washed several times with distilled water to get rid of sodium chloride remains, and the solid residue was dried in the air, ground then sieved by 50µm sieve.

Surface modification of sepiolite with NCP surfactant

N-cetylpyridinium (NCP) bromide was used as a surfactant purchased from Merck company. Modified-sepiolite was prepared by adding quantities of the respective bromide salts equal to twice the cation exchange capacity of the sepiolite in 100 cc of 50% solution water/ethanol and 10 g of the Na-saturated clay, stirring for 24 h. The clay was then washed with deionized water until free of salts and a negative bromide test had been obtained with $AgNO_3$ and it was grounded, sieved through a 50µm sieve, and dried at air and was used for the adsorption studies.

Sorption experiment

Sorption isotherms of 2,4-D (obtained from Merck) by natural and modified sepiolite were obtained using the batch equilibrium technique. Triplicate 200 mg samples were treated with 30 mL solutions of 2,4-D in 0.01 M NaCl in concentrations 100-1200 µM. The suspensions were shaken intermittently at 25±2 ◦C for 24 h in a rotary shaker for reach equilibrium. Subsequently, the suspensions were centrifuged at 3500 rpm for 15 min. The 2,4-D concentration in the filtrates was determined spectrophotometrically by measuring absorbance at λ_{max} of 282 nm.

The amounts of 2,4-D adsorbed were calculated from the concentrations in solutions before and after adsorption using equation below:

$$Qe = \frac{V(Ci - Ce)}{W} \qquad \text{Eq(1)}$$

where *C*i and *C*e are the initial and equilibrium liquid-phase concentrations of 2,4-D in solution (mmol L^{-1}), respectively; *V is* the volume of 2,4-D solution (L), and *W* is the mass of the natural and modified-sepiolite sample used (g). The adsorption data were analyzed to see whether the isotherm obeyed the Langmuir and Freundlich isotherm models.

Langmuir isotherm was applied to quantify the maximum amount and the affinity of 2,4-D sorption on the sorbents. This isotherm has the form of:

$$Qe = \frac{Qmax\ Kl\ Ce}{1 + Kl\ Ce} \qquad \text{Eq. (2)}$$

Where, Q_e is the amount sorbed per unit mass of sorbent and Ce is the equilibrium solution concentration of sorbate. Two parameters, maximum sorption (Q_{max}) and bonding energy coefficient (K_L) were used to describe the sorption of 2-4-D on the sorbents.

Freundlich model is expressed by the following equation:

$$Qe = K_F\ Ce^N \qquad \text{Eq. (3)}$$

where K_F is the Freundlich partition constant and N is a physical factor related to adsorption mode and surface heterogeneity, Q_e and C_e were described before. The Freundlich isotherm has been derived by assuming an exponentially decaying sorption site energy distribution.

Results and Discussion

The chemical composition of sepiolite determined from XRF analysis, the cation exchange capacity and specific surface area of sepiolite are shown in Table 1

Figure 1 shows XRD patterns of this mineral. The presence of intense reflection at 2ɵ=7.4 (12.3˚A) that is related to d_{001} of sepiolite indicated that the sepiolite is the main mineral in the sample.

Table 1: The chemical characteristic of natural sepiolite

Composition (wt%)												CEC (cmol/kg)	Surface area (m^2/gr)
SiO_2	Al_2O_3	Fe_2O_3	CaO	Na_2O	K_2O	MgO	TiO_2	MnO	P_2O_5	SO_3	Loss of ignition		
53.9	0.21	0.01	2.94	0.01	0.01	24.22	0.002	0.001	0.001	0.005	18.21	2.5	102

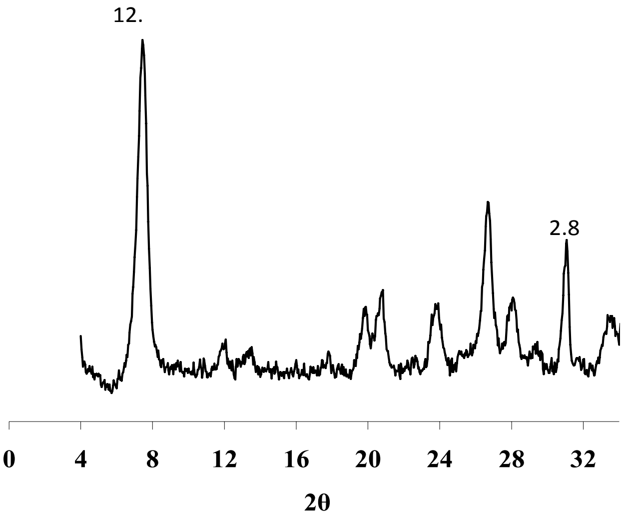

Figure 1. XRD patterns of natural sepiolite

Langmuir and Feundlich equations represented significant fits to the 2,4-D sorption data on modified sepiolite (Fig. 2).

The adsorption constants are obtained by non-linear regression analysis (statistical package GraphPad Prism, version 5.00 for Windows, GraphPad Software, San Diego, California, USA) for each model are listed in Table 2.

Natural sepiolite could not sorb 2-4-D due to its surface hydrophilic characteristic. Xi et al. (2010) investigated 2,4-D sorption on natural palygorskite and reported only negligible amounts of 2,4-D adsorbed by natural palygorskite.

The organic modification of sepiolite facilitated 2,4-D adsorption. Modified sepiolite adsorbed as much as 2.22 mg/gr of 2,4-D. Enhanced adsorption of 2,4-D by organo sepiolite can be related to the hydrophobic interaction between the herbicide and this organoclay. Moreover, modification of sepiolite by NCP^+ probably neutralize the negative charges and even change the clay surface to the positive charge leading to more attraction of 2,4-D anions. Hermosin et al. (2006) reported modification of the montmorillonite sample with HDTMA cations equal to %200 of clay CEC resulted in organoclays with enhanced affinity for the 2,4-D herbicide. Finally, This study concluded that modification of sepiolite with NCP can effectively improve 2,4-D removal from polluted waters.

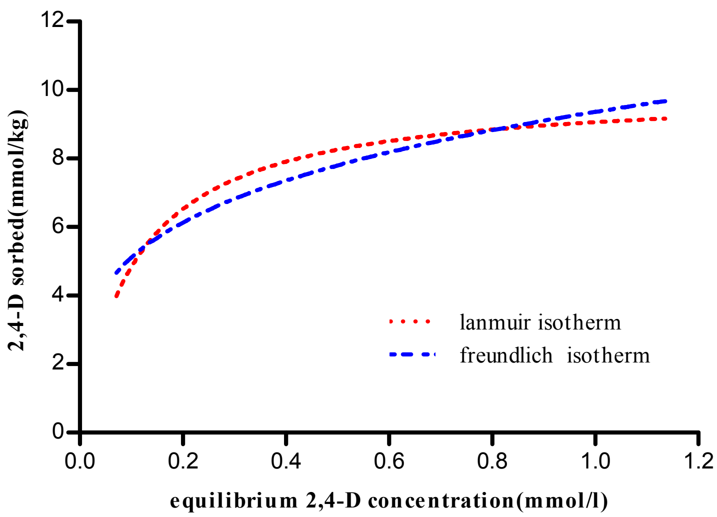

Figure 2. Langmuir and Freundlich isotherms of 2,4-D sorption on modified sepiolite

Table 2: constant parameters of 2,4-D sorption on modified sepiolite

Langmuir			Freundlich		
$Q_{max(mmol/kg)}$	$K_{L(L/mmol)}$	R^2	k_f	n	R^2
10.04	9.27	0.86	9.36	0.26	0.86

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The Zinc Accumulation in Some Links of Food Chain of Aquatic Ecosystems

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Abstract
The aim of the study was to determine the level of zinc accumulation in selected parts of food chain in the carp pond farming. Based on the results, the level of zinc bioaccumulation in the aquatic system was estimated. The study was carried out on two adjacent fish ponds, located in Krakow-Mydlniki. To conduct the analyses the following samples were collected at each pond: water, soil from the bottom of the ponds, benthic organisms (larvae of midge flies), and organs of carp (gills, muscle, gonads, liver) which are involved in the metabolism of the tested element. In all samples, the zinc content was determined by atomic emission spectrometry using JY 238 Ultrace Jobin Yvon Emission analyzer. The preparation of samples was performed according to standard methods defined in the Regulations of the Ministry of Health and the Ministry of the Environment. The results of the research show the good quality of the abiotic components of ecosystems examined on the basis of their zinc content. Water in the ponds was classified as water of first class of quality due to its zinc content, while the soil was classified to A category. The zinc accumulation in fish bodies was as follows: gills> liver> gonads> muscle.
Keywords: zinc, bioaccumulation, food chain, aquaculture

Introduction
Ponds used for fish production fulfil also many other functions. The role they play in the environment depends on their size, location but also on the method of production. Fish ponds are of crucial importance for the natural conditions of their immediate surroundings. Their presence affects modelling water regime in a valley and even in the river catchment. The system of water management in carp ponds positively influences water retention in the catchment where they are situated. Biogens which find their way to a water reservoir with water during pond filling become included in the cycle of biogeochemical changes, which cause that during the production cycle ponds retain not only considerable amounts of nitrogen and phosphorus, the elements responsible for eutrophication, but also trace elements.
Zinc finds its way into aquatic environment mainly from anthropogenic sources. The sources of this element are surface runoffs from agricultural and urbanized areas, but also industrial and municipal sewage. In water this element occurs in soluble or colloidal form and as a component of suspension. Zinc ions have a strong affinity to adsorption on solid particles and to formation of hydrated ions, ionic pairs and complex compounds (Kabata-Pendias and Pendias, 1999). In result of these processes zinc emitted to aquatic environment remains in a soluble form for a short time and accumulates in bottom sediments. Degree of zinc accumulation in bottom sediments depends not only on this element content in water but in the first place on the amount of organic matter and sediment granulation. The more of organic matter is present in aquatic ecosystem, the more intensive is this element binding with bottom sediments. Many authors point to a considerable zinc concentration in bottom sediments, although its amount in water does not indicate anthropogenic enrichment (Liao et al., 2003). A change of aquatic environment conditions may lead to secondary mobilization of this element in the environment, which may result in its incorporation into the food chain in excessive amounts.
Zinc present in water penetrates into fish organisms directly through the skin and gills or with feed to the digestive system. The rate of toxic effect of the metal is connected with transport function of blood, which distributes it all over the organism (Fernandes et al., 2007).

Material and methods
In 2008 research on zinc accumulation was conducted in a fish pond owned by the Experimental Station of the Department of Ichtiobiology and Fisheries, University of Agriculture in Krakow, situated in Mydlniki. The pond is fed by water of Rudawa River. The investigations comprised determining zinc concentrations in water, bottom sediments, benthos and in fish.

Water zinc concentrations were assessed thrice during the vegetation period. Samples were collected at the beginning of the fattening period in May, during the most intensive fish feeding in July and in September, by the end of the fattening period. Water was sampled from six points of a pond. Bottom sediments were collected from the top layer of the pond bottom (0-5 cm) after pond emptying. The pond was divided into 8 zones and laboratory samples were collected (two samples close to the pond inlet box, four samples from the middle part of the pond and two in the vicinity of the pond outlet box). Samples of benthos organisms (larvae of *Diptera Chironomidae*) were collected in the same places. The zinc accumulation in fish organisms were assessed in 25 randomly chosen fish destined for human consumption. The fish sex was determined (by organoleptic method), their age (insight into production documents) and mass of the analyzed carps (gravimetric method). Bottom sediments were dried and sifted through a sieve with 1 mm mesh and ground in a mortar. Carps were killed by decapitation and their organs (gills, muscles, livers and gonads) were prepared. Obtained laboratory samples were wet mineralized in a closed system using microwave energy. The analytical weighted portion was c.a. 0.5 g in conversion to dry mass. Biological material was dissolved in a mixture of nitric acid and hydrogen peroxide, whereas the bottom sediments in *aqua regia*. The water for analyses was condensed ten times. Zinc concentrations in the obtained solutions were assessed by means of atomic emission spectrometry in inductively coupled plasma on JY 238 ULTRACE apparatus (Jobin Yvon Emission) at the wavelength 206.200 nm. The limit of zinc quantification in the applied method was 0.0059 $mg{\cdot}dm^{-3}$. Measurement uncertainty of the applied methods was ±5 %. The limit of quantification of the analyses was 0.058 $\mu g{\cdot}g^{-1}$ f.m. of sediments. The correctness of zinc measurements was checked by a certified reference material CRM 16-050. Moreover, organic carbon content, pH and grain size composition were assessed in the sediments.

Results and discussion

Mean zinc concentration in water from the analyzed pond was 0.206 $mg{\cdot}dm^{-3}$. Its content in water showed the smallest variability of all tested parameters (RSD = 7.78 %) (Tab. 1).

Table 1. Statistic parameters of zinc concentration in studied elements of aquatic ecosystem

Statistic parameters	Element of aquatic ecosystem						
	Water	Sediments	Larvae	Carp organs			
				Gills	Gonads	Muscle	Liver
	$mg{\cdot}dm^{-3}$	$mg{\cdot}kg^{-1}$ d.m.	$mg{\cdot}kg^{-1}$ f.m.	$mg{\cdot}kg^{-1}$ f.m.			
Minimum	0.159	72.87	14.88	79.22	7.833	5.01	43.50
Maximum	0.206	183.2	28.10	231.4	305.3	56.92	167.4
Mean	0.181	115.6	22.87	143.8	87.39	15.48	113.3
SD*	0.016	40.51	4.82	38.6	87.88	10.00	28.03
Median	0.182	96.62	23.64	140.8	69.04	14.07	118.7
RSD** (%)	8.96	35.04	21.06	26.84	100.6	64.56	24.74

*Standard deviation; **Relative standard deviation

In September the content of the analyzed metal in water was statistically significantly lower than its quantity in water in May or July (Fig. 1).

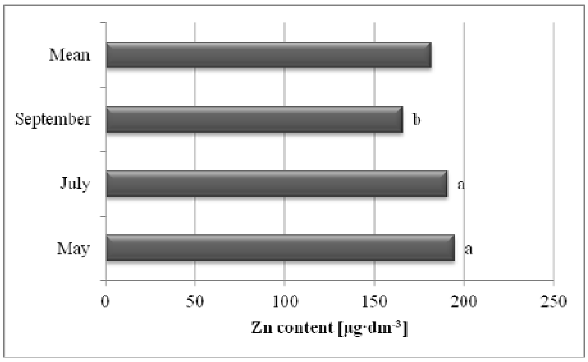

Fig. 1 Concentration of zinc in water of pond

Because of zinc content, the analyzed water was characterized by a very good quality (Dz. U. 2011, 32). Wiśniowska-Kielian and Niemiec (2004) assessed several-fold lower amounts of this element in the agricultural-recreational part of the Dunajec river catchment and only the values registered in the municipal and industrial sewage discharge area were higher than obtained in the presented research. The zinc concentration in unpolluted waters of Hannah Lake maintained at 14 $\mu g \cdot dm^{-3}$, whereas in Whitson Lake was 10 $\mu g \cdot dm^{-3}$ (Eastwood and Couture, 2002). Zinc quantities similar to obtained in presented investigations were noted in reservoirs accumulating runoffs from the national road No. 4 in Krakow neighbourhood (Niemiec and Wiśniowska-Kielian, 2008). Norris et al. (2000) reported this element concentration in the Colorado River on the level of 1090 $\mu g \cdot dm^{-3}$. Such high concentration was caused by pollutants emission from the nearby zinc mine. While comparing zinc quantities assessed in clean and polluted waters one may state that the analyzed waters reveal elevated zinc concentrations.

Average zinc content in bottom sediments from the analyzed ponds maintained at the level of 96.6 $mg \cdot kg^{-1}$ d.m. (Tab. 1). Considering zinc concentrations, the analysed material was classified to the first class quality soils under surface waters (Dz. U. 2011, 257). Zinc content assessed in the analyzed sediments was on the first level of pollution, which indicates anthropogenic enrichment of the studied sediments (Bojakowska and Sokołowska, 1998). Bottom sediments of water reservoirs in which fish farming is conducted are especially susceptible to excessive accumulation of trace elements, which is connected with specific exploitation of such reservoirs. Adhikari et al. (2009) stated zinc content in fish ponds on the level of about 100 mg $Zn \cdot kg^{-1}$, whereas this element content in water was below the detection level. Very strong zinc absorption in bottom sediments is connected with a considerable content of organic matter. Metal accumulation in sediments may lead to increasing their amount in organisms of fish destined for consumption. One of the methods used for pollutant removal from ponds is their dredging (Yuvanatemiya and Boyd, 2006). The authors found over 50 % reduction of zinc quantity in sediments from regularly cleaned fish ponds. According to the research conducted by Balasubramanian et al. (1995) zinc concentration in sediments from farm ponds in India was 23.73 $mg \cdot kg^{-1}$ d.m. On the other hand, Silva-Filho et al. (2006) found that zinc content in polluted sediments from the Buffalo River in Brazil reached 476 $mg \cdot kg^{-1}$ d.m.

Bottom sediments are an integral part of the aquatic environment, place of living, reproduction and growth of aquatic organisms. *Chironomidae* larvae are of crucial importance among all benthos animals. Considering nutritional value, protein content, size and availability they provide the most valuable feed for whitefish, particularly carps. Mean zinc content in *Chironomidae* larvae was 21.5 $mg \cdot kg^{-1}$ f.m., and ranged from 14.9 $mg \cdot kg^{-1}$ f.m. to 28.1 $mg \cdot kg^{-1}$ f.m.

Table 2. Bioacumulation coefficient of zinc

Element of aquatic ecosystem	Larvae	Gills	Gonads	Muscle	Liver
Water	125	790	480	85.1	622
Sediments	0.198	1.24	0.756	0.134	0.980
Larvae		6.29	20.96	3.715	4.956

Zinc bioaccumulation coefficient in the analyzed benthos *versus* content in water and sediment was 125 and 0.198, respectively (Tab. 2). Such low zinc bioaccumulation coefficients may be due to low content of bioavailable zinc forms in the aquatic environment. The area of the Rudawa River catchment, which feeds the analyzed pond, abounds in parent rocks containing considerable amounts of the studied element, which may provide a source of zinc in the analyzed ecosystems. A big amount of calcium carbonate in the bedload influences metal immobilization, which explains a low bioaccumulation coefficient. Goodyear and McNail (1999) assessed much higher zinc concentrations in macroinvertebrates of various aquatic environments. The same authors found statistically significant dependence between zinc amount accumulated in bottom sediments and the level of its accumulation in macroinvertebrate organisms. The value of zinc bioaccumulation coefficient from bottom sediments was about 1. Values of zinc bioaccumulation in invertebrate organisms in relation to this metal content in water fluctuated widely depending on its water concentrations. On the basis of results concerning many invertebrate species Goodyear and McNail (1999) observed a lack of dependence between zinc quantity in water and its bioaccumulation level. Obtained values in authors own studies are not high. Zauke et al. (2003) assessed zinc content in *Gammarus oceanicus* in the Barents Sea on the level of 61 $mg \cdot kg^{-1}$ f.m. Zinc concentrations in *Haliotis* from aquacultures ranged from 46.41 to 111 $mg \cdot kg^{-1}$ f.m. depending on this element amount in water (Liao et al., 2003). Values of zinc bioaccumulation coefficient in *Haliotis* organisms in relation to its water concentrations ranged from 847 to 1320 depending on pollution level. The more of zinc in water the lower bioaccumulation coefficient was registered for *Haliotis* organisms.

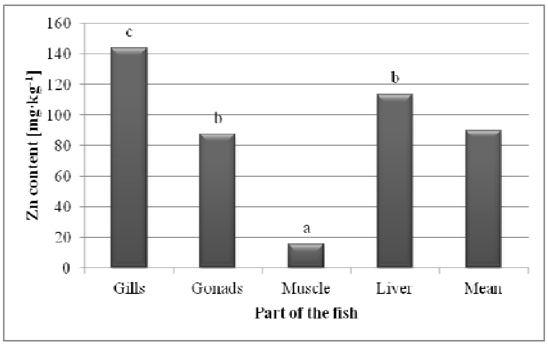

Fig. 2 Concentration of zinc in organs of carp

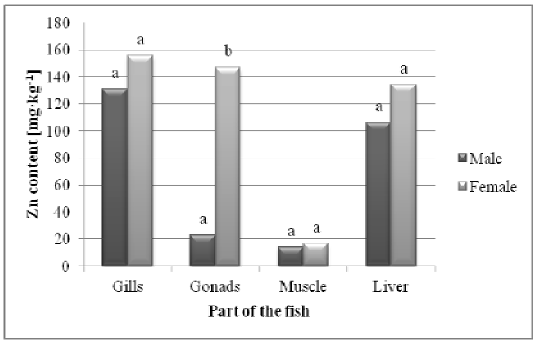

Fig. 3 Concentration of zinc in organs of carp depending on the gender

Average zinc concentration in fish muscles was 15.48 $mg \cdot kg^{-1}$ f.m. (Fig. 2) and fluctuated from 5.01 to 56.92 $mg \cdot kg^{-1}$ f.m., at relatively high variability (RSD = 64.56 %) (Tab. 1). The deference between concentration of this metal in males and females muscles was not found (Fig. 3). Small content of zinc in fish muscles was connected with the specificity of this element metabolism. The organs which finally accumulate zinc in fish organisms are gills, liver, kidneys and gonads (Chi et al., 2007). Brucka-Jastrzębska et al., 2010 determined the natural zinc content in rainbow trout (*Oncorhynchus mykiss*) muscles on the level of about 6.7 $mg \cdot kg^{-1}$ f.m., whereas in *Ctenopharyngodon idella* about 20.1 $mg \cdot kg^{-1}$ f.m. Fish feeding by the bottom are characterized by a higher content of zinc in muscles in comparison with pelagic fish. Chi et al. (2007) report that zinc concentrations in muscles of carps and silver carps (*Hypophtalmichthys molitrix*) from the Jangcy River were 25 mg $mg \cdot kg^{-1}$ f.m. and 21 $mg \cdot kg^{-1}$ f.m., respectively. Despite a considerable concentration of this element in water, no great accumulation was found in muscles. Jones et al. (2000) noted zinc content in muscles of various fish species of about 4.8 $mg \cdot kg^{-1}$ f.m., despite big quantities of this element in the environment. In the same region, zinc content in oysters was 2.5 $g \cdot kg^{-1}$ f.m. Zinc bioaccumulation coefficients in muscles of the studied fish were 0.134 in relation to sediments and 0.677 for benthos. Zinc content in fish muscles is not a good indicator of the environment pollution with this element.

Mean zinc concentration in liver was 113.3 $mg \cdot kg^{-1}$ f.m. (Fig. 2) and fluctuated from 43.50 to 167.4 $mg \cdot kg^{-1}$ f.m., at fairly low RSD amounting 24.74 % (Tab. 1). No statistically significant differences were found between this element content in male or female livers. Liver is the main place of zinc accumulation in fish organisms and is often used as an indicator of the environment pollution with this element. Mormede and Davies (2001) report that average zinc content in livers of fish from the Atlantic Ocean is 62.35 mg·kg^{-1} f.m., which is considered as natural content of this metal in livers of fish from not anthropogenically transformed areas. Schmitt et al. (2007) state mean zinc content in livers of fish from polluted areas as 733 mg·kg^{-1} f.m., whereas the amount of this element in these organs of fish living beyond the pollution zone was on the level of 74 mg·kg^{-1} f.m. The authors point at statistically significant dependence between the distance from the pollution source and zinc concentrations in fish livers. On the other hand, Heier et al. (2009) indicate a significant dependence between zinc concentration in water and fish livers. The value of bioaccumulation coefficient in these organs was 622 in relation to its water concentration and 0.980 for sediments. In the light of the literature data it is a low value, which evidences a small amount of available zinc forms in the studied ecosystem.

Zinc content in gills of the analyzed fish fluctuated from 79.22 to 231.4 mg·kg^{-1} f.m., at an average value of 143.8 mg·kg^{-1} f.m. (Tab. 1, Fig. 2). Zinc content in gills of fish kept in water with elevated zinc content ranged from 313 to 767 mg·kg^{-1} f.m. (Heier et al., 2009). The same authors did not find any dependencies between zinc concentrations in water and gills. Differences in this element accumulation in the organisms of fish examined by these authors were due to randomness and individual variability. Fernandes et al. (2007) report zinc content in fish gills (*Mugil auratus* and *Mugil saliens*) from Esmoris Bay in Portugal on the level of 114 mg·kg^{-1} f.m. Zinc content in the sediments from this bay was on average 213 mg·kg^{-1} f.m. Amounts of zinc assessed in gill were bigger than in liver and muscles by 1.3 and 4.4-fold, respectively. Similar results were obtained in the presented investigations. Coefficient of zinc bioaccumulation for this organ was 1.24 against its content in bottom sediments and 790 in comparison to water. Coefficient of zinc bioaccumulation in gills of analyzed fish is much higher than noted in variously polluted environments presented in literature.

Zinc content in gonads to a considerable extent depended on fish sex (Fig. 3) and fluctuated from 7.833 to 305.3 mg·kg^{-1} f.m., showing the largest variability (RSD = 100.6 %) (Tab. 1). Mean for all samples was 87.39 mg·kg^{-1} f.m. (Tab. 1). Mean content of this element in ovaries was 87.39 mg·kg^{-1} f.m., whereas in testicles 18.64 mg·kg^{-1} f.m. Chi et al. (2007) report over thrice higher zinc concentrations in ovaries in comparison with carp testicles. Results presented by Chi et al. (2007) indicate that ovaries are the organs which accumulate zinc to the greatest extent. The same authors observed a significant dependence between zinc content in water and gonads. Coefficient of zinc accumulation in gonads of the studied fish was 480 in relation to water and 0.756 in comparison to bottom sediments (Tab. 2). Zinc bioaccumulation coefficient in gonads was lower than the literature data from other regions of the world.

Conclusions

1. Zinc content in water from the ponds and bottom sediments indicated enrichment with this element.
2. Zinc concentrations in individual fish organs from the highest values looked as follows: gills>ovaries>liver>testicles>muscles.
3. Zinc concentrations in muscles of all studied carps did not exceed critical values for fish muscles designed for consumption.
4. Lower bioaccumulation coefficients than presented by other authors were stated for muscles, gonads and liver but much higher for gills. Bioaccumulation coefficient ranged from 0.197 to 622.
5. Zinc bioaccumulation coefficient from bottom sediments in *Diptera* larvae revealed very low values, much lower than registered in other ecosystems.
6. Despite big contents of zinc in abiotic elements of the analyzed ecosystem, it was found that relatively small amounts of this element entered the biocycle. Considerable amounts of calcium carbonate in bedload limit metal availability to living organisms.

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Accumulation of Mercury in Some Links of Food Chain of Aquatic Ecosystem

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Abstract
The aim of this study was to determine an accumulation of mercury in selected elements of the fishponds ecosystem. On the basis of the results, the rate of mercury bioaccumulation in the aquatic system was estimated next the degree of mercury contamination of ponds was determined. The study determined the mercury content in water, soil from the bottom of the pond, benthic organisms and the organs of carp (gills, muscle, gonads and liver) which are involved in the metabolism of the tested element. Ponds located in Krakow-Mydlniki were analyzed. The 254th AMA analyzer was used for atomic absorption spectrometry as the method of analysis. Based on the results of water testing it was assessed that ponds were not contaminated and the average mercury content in the water did not exceed standards of water quality class. The level of metal accumulation in the soil from the bottom of the tank was classified as a soil of A Group (the classification for land under water reservoirs). The highest average mercury content in fish was found in muscle, and the lowest one in the gills. The level of mercury in the muscle does not exceed the permissible standards for the element content in fish intended for human consumption. Influence of gender on the level of metal accumulation was found. Females accumulate greater amounts of mercury than males.
Keywords: mercury, bioaccumulation, food chain, aquaculture

Introduction

Aquatic environment plays a crucial role in mercury biogeochemical cycle. Mercury enters aquatic environment from the atmosphere in result of dry or wet deposition. It has been assumed that all over the world its deposition is relatively even, except the areas with elevated emission of this element, for instance volcanic regions and areas of large cities (Dórea et al. 2004). Despite the fact that a major part of this element is accumulated in the geosphere, mercury penetrating into water reservoirs poses the gravest problem. In water mercury undergoes transformations, in result of which it becomes better available to living organisms; therefore its considerable amounts become incorporated into the biocycle (Allan 1999). This element does not perform any biological function. It is characterized by a high toxicity. In a water reservoir mercury circles in result of its compounds transformation and despite its being intensively bound by bottom sediments, it poses a grave hazard to living organisms, particularly fish (Kabata-Pendias and Pendias 1999, Liang et al. 2011). A bottom of carp pond forms on parent soil and its top layer is a bottom deposit. Bottom deposits are the place of accumulation of biogenic elements and heavy metals originating from point source and area pollution. Mineralization process occurring during winter pond emptying and subsequent application of agrotechnical measures (ploughing and fertilization), leads to mobilization of the elements which after pond filling with water in spring become available to living organisms. Therefore, immobilization of toxic trace elements in pond bottom sediments is not durable, so the quality of individual elements of the pond ecosystem should be under control to reduce the accumulation of xenobiotics in the organisms of fish consumed by people (Pilarczyk 2002).
The paper aimed to determine the level of mercury accumulation in some links of food chain of aquatic ecosystem under conditions of extensive fish farming in carp ponds and calculation of bioaccumulation coefficients for this element.

Material and methods

Research on mercury accumulation was conducted in 2008 in two adjoining fish ponds at the Experimental Station of the Department of Ichtiobiology and Fisheries, University of Agriculture in Krakow, situated in Mydlniki. The investigations comprised mercury concentration assessment in water, bottom sediments and benthos organisms and in fish.
Mercury concentrations in water were assessed thrice during the vegetation period. The samples were collected at the beginning of the season in May, during the period of the most intensive fish feeding (in July) and in September, by the end of fish keeping. Water was sampled from six points

in the pond. Bottom deposits were sampled from the top layer of the bottom (0-5 cm) after the pond emptying. The pond was divided into 8 zones from which laboratory samples were taken (two samples in the vicinity of inlet box, 4 samples from the middle part of the pond and two close to the outlet box). Samples of benthos organisms (*Diptera Chironomidae* larvae) were collected in the same places. The level of mercury accumulation in fish organisms was assessed in 25 randomly chosen fish destined for human consumption. The fish sex was determined (by organoleptic method), their age (insight into production documents) and mass of the analyzed carps (gravimetric method). Bottom sediments were dried and sifted through a sieve with 1 mm mesh, and ground in a mortar. Carps were killed by decapitation and their organs (gills, muscles, livers and gonads) were prepared, whereas water and benthos organisms were analysed without previous preparation. Mercury concentrations were determined in fresh material. The weighted portion for analyses ranged between 40-80mg.
Mercury analysis in the collected samples was conducted by means of atomic absorption spectrometry on AMA254 apparatus. The analysis comprised three stages (Tab. 1):

- sample drying in oxygen stream,
- sample incineration and release of mercury vapour which is fixed by amalgamator,
- mercury release from the amalgamator and absorbance measurement in two measuring cuvets at the wavelength of 254 nm.

Tab. 1 Parameters of measurement of mercury (s)

Type of sample	Drying	Ashing	Reading
Water	70	120	40
Sediments	30	130	45
Fish and benthic	100	160	60

Measurement uncertainty of the applied methods was ±7%. The limit of detection of the measurements was 0.1 $ng \cdot g^{-1}$. The correctness of mercury measurements was verified by means of certified reference material CRM 16-050. Organic carbon content, pH and grain size composition were also determined in the sediments.
The ponds are situated in Mydlniki, close to the southern side of Balicka Street and fed by the Rudawa River waters. Extensive carp farming is conducted in them including additional feeding with triticale grain. The ponds are not fertilized. Because of close vicinity, the same source and similar chemical composition of the bottom sediments and water, as well as geological structure of the substratum and catchment, they were treated as replications in the investigations on one ecosystem.

Results and discussion
The forms in which mercury occurs in water depend on the oxidation-reduction conditions in it. In strongly oxidative environment $HgCl^{2-}{}_4$ and $HgOH^+$ prevail, in reductive prevalent are CH_3HgS^- and HgS^-, whereas under transitory conditions the biggest quantities of CH_3HgCl and CH_3Hg^{2+} are assessed. Mercury compounds are absorbed by mineral and organic colloids present in water. Colloids fall to the reservoir bottom (in acid waters) or are transported as a suspension (in alkaline waters). Under the influence of microorganisms a transformation of mercury compounds happens and its vaporization from water reservoir surface. Despite this process, increased concentrations of this element are registered in bottom sediments (Gammons et al. 2006). Mercury present in the reservoir is strongly toxic for aquatic organisms. It easily accumulates in living organisms on all levels of food chain unfavourably affecting the course of their life processes.
Mean mercury concentration in waters of the analysed ponds was 0.486 $\mu g \cdot dm^{-3}$. A statistically significant difference was found between this metal content in water collected in May and in September (Fig. 1).

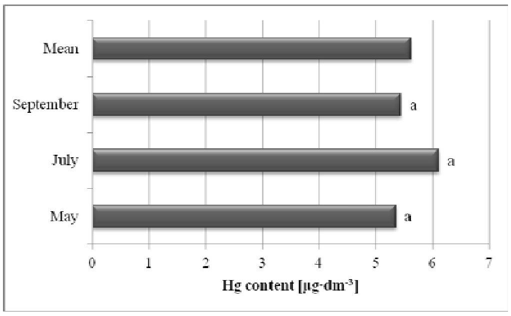

Fig. 1 Concentration of mercury in water from pond

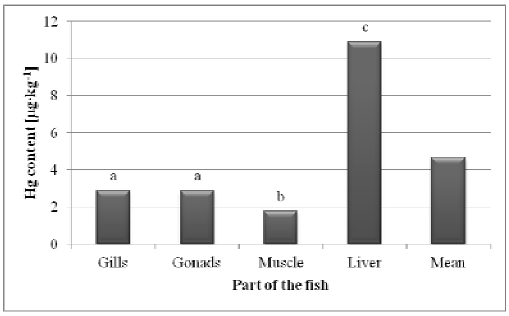

Fig. 2 Concentration of mercury in organs of carps

During an extensive carp keeping period water in the ponds id exchanged in a limited way, which is specific for this kind of production. Increasing mercury amounts in the pond waters in Mydlniki during the research period might have been due to the effect of pollutants from the area of Krakow. Because of their mercury concentration, the analyzed waters collected in May and July were classified to the first class of purity, whereas exceeded critical value for waters of the first class of purity, i.e. 0.5 $\mu g \cdot dm^{-3}$ (Dz. U. 2004, 32), was registered in water from the last sampling. Gammons et al. (2006) assessed mercury content in Titicaca lake water and in rivers flowing into the lake on the level between 0.62 and 244 $\mu g \cdot dm^{-3}$. This metal presence in that region is caused by anthropopressure connected with gold excavation in the neighbouring mines. On the other hand, Southworth et al. (2002) stated an average mercury concentration 1.04 $\mu g \cdot dm^{-3}$ in an industrial pond in Oak Ridge in Tennessee. Much higher content of this metal was observed in the Baltic Sea waters, where it ranged from 5 to 140 $\mu g \cdot dm^{-3}$ depending on the region (Backlund et al. 1992).

Tab. 2. Statistic parameters of data

Statistic parameters	Parameters						
	Water	Sediments	Larvae	Gills	Gonads	Muscle	Liver
	$\mu g \cdot dm^{-3}$	$\mu g \cdot kg^{-1}$ d.m.	$\mu g \cdot kg^{-1}$ f.m.				
Minimum	0.389	79.00	31.12	4.05	3.52	8.33	3.76
Maximum	0.568	95.52	48.50	14.85	23.72	51.28	24.65
Mean	0.474	86.75	37.45	8.24	8.51	23.83	10.10
Standard deviation	0.0636	6.735	6.862	2.82	4.38	12.93	4.34
Median	0.474	86.50	35.12	7.971	7.953	20.89	9.457
Variation coefficient (%)	13.6	7.76	18.32	34.28	51.51	54.26	43.01

Mean content of mercury in the analyzed sediments was 86.7 $\mu g \cdot kg^{-1}$ d.m., while its content in the samples fluctuated between 79.6 and 95.6 $\mu g\ Hg \cdot kg^{-1}$ (Tab. 2). Permissible content of mercury in soil under water reservoirs is 500 $\mu g \cdot kg^{-1}$ d.m. as stated by the Regulations of the Minister of the Natural Environment on the quality standards of soil and earth quality standards (Dz.U. 2002, 165). Differences in mercury content in the analyzed samples were statistically non-significant. Mercury amounts in the studied sediments do not indicate anthropogenic enrichment in this element. The obtained results are lower than registered for sediments from rivers of polluted areas. Działoszyńska-Wawrzkiewicz (2008) reported mercury amount in the sediments from Kłodnica on the level of between 42 and 375 $\mu g \cdot kg^{-1}$ d.m. Weech et al. (2004) investigated mercury level in bottom sediments from Pinich Lake to which sewage from goldmine is discharged. The amounts of mercury in these sediments were 2315 $\mu g \cdot kg^{-1}$ d.m. Mercury concentrations in polluted reservoirs may reach between 460 and 6400 $\mu g \cdot kg^{-1}$. The authors state that in non-polluted lakes, mercury content in sediments should not exceed 49 $\mu g \cdot kg^{-1}$ d.m. The soil from the bottom of analyzed ponds revealed low concentration of mercury, not exceeding the amounts observed in clean water reservoirs.

Mercury content in *Chironomidae* larvae fluctuated between 31.2 and 48.5 $\mu g \cdot kg^{-1}$ f.m. assuming a mean value of 37.45 $\mu g\ Hg \cdot kg^{-1}$ (Tab. 2). Relative standard deviation for all samples was 18.3%. Locarnini and Presley (1996) determined this metal content in *Chironomidae* larvae originating from Keller Bay water reservoir in Texas, polluted with mercury from the power plant, on the level of 101 μg·kg^{-1} f.m. Mercury concentrations in benthos and pelagic organisms in the Patagonia lakes ranged between 14 and 600 μg Hg·kg^{-1} f.m., however this metal content in the benthos organisms ranged from 8-360 μg·kg^{-1} f.m. (Rizzo et al. 2001). Mercury accumulation in benthos organisms is connected with this element content in sediments, but also depends of physicochemical and physical properties of the sediments, on thermal conditions and oxidation-reduction potential in the sediment water. All these environmental parameters affect mercury bioavailability, which to the greatest extent influences this element amount accumulated by living organisms. Mean mercury bioaccumulation coefficient in benthos invertebrates in the analyzed ecosystems was 0.432 (Tab. 3).

Tab. 3 Bioaccumulation coefficient

	Larvae	Gills	Gonads	Muscle	Liver
Water	80.10	22.56	18.19	50.95	21.59
Sediments	0.432	0.122	0.098	0.275	0.116
Larvae		0.282	0.227	0.636	0.269

High value of bioaccumulation coefficient is typical for ecosystems unpolluted with mercury. Value of bioaccumulation coefficient in *Ephemeroptera* larvae in the Irdijca River ecosystems, polluted with this element, fluctuated from 0.079 to 1.366 in conversion to fresh mass (Žižek et al. 2001). High values of the coefficient were characteristic for the sites with very low mercury content. Moreover, the authors demonstrated that on the research site characterised with a bigger amount of organic matter, mercury concentrations in the sediments were higher in comparison with the sites of more mineral substratum. However, mercury accumulated in the substratum was unavailable to living organisms. Mercury content in *Chironomidae* larvae in the lakes of northern Patagonia ranged from 570-2000 μg·kg^{-1} d.m., which in conversion to fresh mass gives the values of about 50-200 μg·kg^{-1}. Value of bioaccumulation coefficient on organic sites was even 10-fold lower in comparison with places with larger amounts of organic matter. Average content of organic matter in the analyzed ponds was low – 3.30 %. In spite of supplying considerable amounts of organic matter to ponds as feed and fertilizers, winter drying and cultivation measures favour its mineralization. Mineralization processes lead to increasing the amount of mercury incorporated to the food chain of aquatic ecosystems.

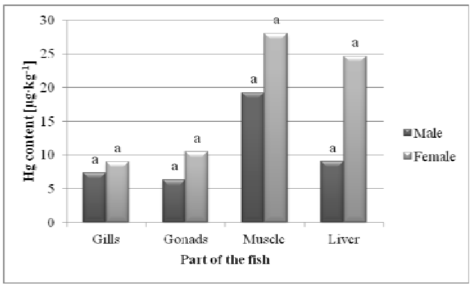

Fig. 3 Concentration of mercury in organs of carps depending on the gender

Mercury present in aquatic environment is easily absorbed by aquatic plant and animal organisms. The metal penetrated to fish organisms through gills, skin and with feed (Tuomola et al. 2008; Leśniewska et al. 2009). Mean content of mercury in the gills of analyzed fish was 8.238 μg·kg^{-1} f.m. and ranged from 4.052 to 14.85 μg·kg^{-1} f.m. A statistically significant content of this element was assessed in female gills (9.031 μg·kg^{-1} f.m.), while its average amount in male gills was 7.38 μg·kg^{-1} f.m (Fig. 3).

Average content of mercury in the analysed fish muscles was 23.83 μg·kg^{-1} f.m. and fluctuated from 8.33 to 51.28 μg·kg^{-1} f.m., at mean standard deviation 12.93. A considerably higher (about 50 %) level of this element accumulation was assessed in female muscles in comparison with the same organs in males (Fig. 3). A big quantity of mercury assessed in muscles is connected with the specificity of this metal cycling in fish organisms. Muscles are the main place of mercury accumulation in fish and their concentrations of this metal have been currently assumed an indicator of mercury pollution of aquatic ecosystems (Stężycka et al. 2005; Leśniewska et al. 2009). Rizo et al. (2011) stated mercury content in muscles of predatory fish from the lakes susceptible to anthropopressure as between 3 and 10 times higher than the results obtained in the presented investigations. Mercury contents in the muscles of investigated fish were comparable with its amounts in carps from unpolluted water of the Pearl River in Hong Kong. Mercury concentrations in carp muscles from this river fluctuated from 17.5 to 84 $\mu g \cdot kg^{-1}$ f.m. (Zhou and Wong 2000). High level of mercury accumulation in fish muscles from Pucka Bay and open Baltic Sea outside the Hel Peninsula were registered by Bełdowska et al. (2007). Among small fish the highest concentrations of mercury were observed in herrings – 95.7 $\mu g \cdot kg^{-1}$ f.m., in perches - 83 $\mu g \cdot kg^{-1}$ f.m. and in common whitefish (*Coregonus lavaretus*) – 80.6 $\mu g \cdot kg^{-1}$ f.m. Coefficient of mercury bioaccumulation in carp muscles in relation to its content in bottom sediments from the unpolluted areas of Hong Kong was 0.2-0.3 (Zhou and Wong 2000). In case of fish presented in the paper the same value was 0.275 (Tab. 3). The value of mercury bioaccumulation coefficient in fish muscles is negatively correlated with the level of environmental pollution. At a considerable accumulation or mercury in abiotic elements of ecosystem, protection mechanisms are activated, which limit this element uptake from the environment (Zhou and Wong 2000, Rizo et al. 2001).

Mean content of mercury in the liver was 10.1 $\mu g \cdot kg^{-1}$ f.m., fluctuating from 3.76 to 16.29 $\mu g \cdot kg^{-1}$ f.m., at standard deviation 4.34 $\mu g \cdot kg^{-1}$ f.m. (Tab. 2). No differences in this element content were assessed between the specimens of different sexes. Much smaller mercury concentrations were registered than in predatory fish from lakes with elevated anthropopressure. In the Patagonia region (Rizo et al. 2001) the average mercury content in fish muscle was about 2.5 times higher in comparison with the liver. Bigger differences in mercury concentrations are observed under conditions of the environment pollution with this element. In the polluted areas Rizo et al. (2001) stated between 4 and 6 times higher mercury content in fish muscles than in livers.

Mercury content in gonads to a considerable degree depended on the fish sex and ranged between 3.52 and 23.72 $\mu g \cdot kg^{-1}$ f.m. Average for all samples was 8.51 $\mu g \cdot kg^{-1}$ f.m. (Fig.1). Mean content of this element in ovaries was 10.97 $\mu g \cdot kg^{-1}$ f.m.,whereas in testicles 6.21 $\mu g \cdot kg^{-1}$ f.m. Gonads are not the organs in which mercury accumulates in big quantities. Mercury content in gonads of some fish from the Baltic Sea contained only 4 $\mu g \cdot kg^{-1}$ f.m.,whereas in the other organs amounts of this metal indicated anthropogenic enrichment (Bełdowska et al. 2007).

Conclusions

Water from ponds and bottom deposits revealed mercury content which does not point to anthropogenic enrichment.

Mercury concentrations in individual fish organs, from the highest were as follows: muscles > liver>gills>gonads.

In the muscles of all analyzed carps mercury concentrations did exceed the critical values for fish muscles destined for human consumption.

High mercury bioaccumulation coefficients, characteristic for clean areas were registered, ranging from 0.097 to 0.432 depending on the kind of organ.

The coefficient of mercury bioaccumulation from bottom sediments in *Diptera* larvae revealed high values, higher than assessed in other ecosystems.

Despite low contents of mercury in the abiotic elements of the analyzed ecosystem, relatively big amounts of this element entered the biocycle.

Fish ponds are the ecosystems more susceptible to excessive mercury accumulation, which is caused by limited binding of this element in bottom sediments.

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Water Quality zoning of Zaringol Stream (Golestan Province, Iran) based on NSFWQI index

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Abstract
Zoning the pollution of a river may be the first or even the most important step in water quality management because it helps us to know how the spatial and temporal pollution changes are. Zaringol stream is one of the Gorgan Rud's branches supplied water source for agricultural uses. In order to resolve its pollution, Biochemical Oxygen Demand (BOD), Dissolved Oxygen (Do), Temperature, pH, Turbidity, Total Solid (TS), total Phosphate, Nitrate and Fecal coliform were measured from 9 point during November 2009 to August 2010. Water quality status was estimated by NSFWQI index. The average of its value is 51.63. NSFWQI index changes in different sampling point shows that maximum (73.3) and minimum value (51.93) are belong to point 1 and 2 respectively. According this result, point 1 has the best water quality condition and point 2 is bad, too. Also, Seasonal index values show that NSFWQI is the minimum in spring. Effluents entering of land uses were located around Zaringol Stream and increasing these activities in spring and summer are reasons of the decline in water quality. Because of important role of this stream in biodiversity conservation it seems must be considered management programs to improve its quality condition and reducing harmful effect of land uses around of it.
Keywords: Water quality zoning, NSFWQI index, Zaringol Steam, Pollution.

Introduction
Water is a vital natural resource that all living organisms depend on to survive, but water quality is being affected by human activity (Khajeh Rahimi et al, 2011). Surface water pollution with chemical, physical and biological contaminants by anthropogenic activities is one of great environmental attention in all over the world (May et al, 2006; Noori et al, 2010; Ouyang et al, 2006). The constant discharges of domestic and industrial wastewater and seasonal surface run-off due to the climate have a strong effect on the river discharge and water quality. However, rivers are the main water sources for domestic, industrial and agricultural irrigation purposes in a region (Yu and Shang, 2003), river water quality is one of important factors directly concerning with health of human and living beings (Kazi et al, 2009). Therefore, regularly assessment and analysis of water quality parameters of rivers help us to taking suitable management decision in using rivers and control its pollution. Thus, it is imperative and important to have reliable information on characteristics of water quality for effective pollution control and water resource management (Mirzaei et al, 2005).
There are many methods to analysis of water quality characters. Water Quality Index (WQI) is one of them which was more simple and common and proposed by Horton (1965) at first. In this method, many measured water quality parameters convert to a dimensionless number described water quality condition as a spatial scale.
NSFWQI is a general index which classified water quality without regard to type of usage (Asadollah Fardi et al, 2000). Because of being simple and availability of required parameters, it is more used for water quality zoning of rivers (Zandbergen and Hall, 1988).
Zaringol stream with 22 kilometer length is one of the Gorgan Rud brunches which located about 57°37 E, 36°52 N. it has important role in water supply of agricultural, aquaculture and domestic usages (Abdoli and Rahmani, 2002). Maximum (17.9 cubic meters per second) and minimum (0.44 cubic meters per second) discharge belong to March and July respectively (Vezarat-niro, 2003).

In recent years, great changes have taken in marginal regions as the forest region has converted to other land use such as agriculture. Despite of the severe effect of human activities, there is no report on water quality assessment or zoning. Also, because there is only one hydrometric station along the stream, so there is not enough data base to evaluating effect of them.
The aim of this study was water quality zoning of Zaringol stream based on NSFWQI index for describing spatial and temporal water quality condition along the Zaringol stream.

Materials and methods
The present study was carried out in the Zaringol stream which is a small stream situated in the eastern Elburz Mountains (Golestan province in northern Iran). Sampling of water was carried out from December 2009 to September 2010, seasonally.
Nine water quality parameters required for calculating NSFWQI consist of biological Oxygen demand (BOD_5), dissolved Oxygen (DO), Nitrate (NO^-_3), Phosphate (PO^-_4), Temperature (ΔT), pH, Turbidity, Fecal coliform and Total Suspended Solid (TSS) were measured from 9 point which showed in Fig 1, by Water checker u-10 and Spectrophotometer.

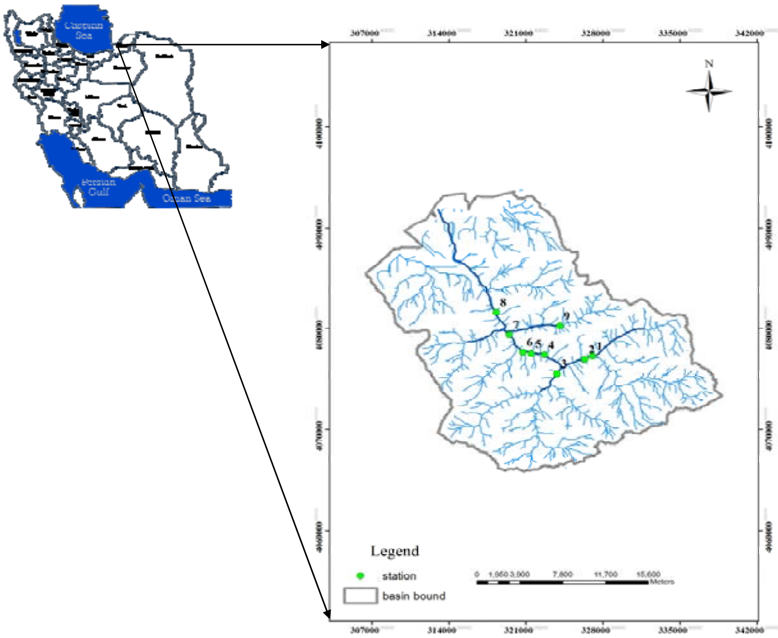

Fig.1: site of water sampling- Zaringol stream

NSFWQI were calculated by follow equation:

$$NSFWQI = \sum W_i Q_i$$

Where, W_i equal to the weight of each water quality parameter (see Table 1):

Table 1. Water quality parameters and their weight used in NSFWQI

parameter	Unit	Weight
DO	Saturate (%)	0.17
Fecal coli form	Colony/100ml	0.16
pH	---	0.11
BOD_5	Ppm	0.11
ΔT	◦C	0.1
NO_3	Ppm	0.1
PO_4	ppm	0.1
Turbidity	NTU	0.08
T.S.S	Ppm	0.07

And Q_i is the value of each water quality parameter in 0-100 scale which got from conversional curve (Fig 2):

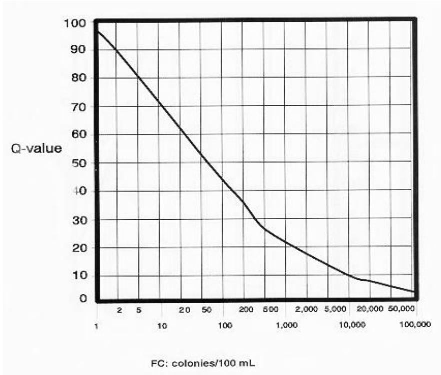

Fig 2. Conversional curve of fecal coli form.

NSFWQI is a reduction index, so if the water pollution increases, NSFWQI decreased. It evaluated for each season and water quality condition assessed according to Table 2.

Table 2. Mean value of NSFQWQI for assessment of water quality condition

NSFWQI	Water quality condition
90-100	Very well
70-90	Good
50-70	Average
25-50	Bad
0-25	Very bad

Results

The average of NSFWQI index was 51.63 and it's ranged between 51.93 to 73.3. In Table 3, the average of NSFWQI index which calculated for each station was shown.

Table 3. The average of NSFWQI index in different station

Station	NSFWQI	Water quality condition
1	73.3	Good
2	51.93	Average
3	57.45	Average
4	58.11	Average
5	53.27	Average
6	54.91	Average
7	55.14	Average
8	57.32	Average
9	59.22	Average

The minimum and maximum value of NSFWQI index calculated in spring and autumn respectively (Table 4).

Table 4. The average of NSFWQI index in different season

Season	NSFWQI	Water quality condition
Spring	54.59	Average
Summer	55.06	Average
Autumn	58.46	Average
Winter	58.25	Average
Annually	51.63	Average

Discussion

Results of NSFWQI index in different stations and seasons show spatial and temporal variation in its values. Maximum value and best water quality condition observed in station 1 which located after spring. Station 2 located after a Trout farm with 15 ton actual capacity. Its NSFWQI shows decrease in water quality because of reception of the Trout farm effluents. After this station, increasing process can observe in NSFWQI values not in quality condition except in station 5. Although self-purification power of stream improved its value, Agricultural plan along the stream prevent water quality trend to better condition. Station 5 located after a Trout farm with 7 ton actual capacity, too. So, decreasing in NSFWQI may refer to entrance of its effluent, too.

Seasonal variation in index value shows minimum value in spring. Because agricultural activity and reproduction period of Trout farm begin in spring and continue in summer, water quality of warm season may affect more than cold season. Also, because of high turbidity in winter NSFWQI is lower than winter's value. It may refer to flood incidence in winter. Generally, water quality condition of Zaringol stream is average.

It seems, using wastewater treatment of Trout Farm, controlling the volume of its entrance and preservation a suitable distance based on self-purification power of stream between fish farms can reduce intensity of water quality decline.

Since, regularly assessment can help us to probe effect of pollutant on water quality and future decisions; NSFWQI is a simple tool to initially water quality ranking can be used.

Acknowledgment

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The effect of Trout farm effluents on the water quality parameters of Zaringol Stream (Golestan, Iran)

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Abstract

Fish processing plants produce a considerable volume of effluents and when they are released in to the river it can affect on the water quality and downstream aquatic systems. The aim of this study was to assess the potential impact of Trout farm effluents on water quality of Zaringol stream (Golestan, Iran). Two Trout farms and 8 stations were selected and Water quality parameters were monitored during November 2009 to August 2010, seasonally. Results show that Trout farms had significantly impact on the average of Turbidity, Phosphate (PO_4), Nitrite (NO_2), Electrical conductivity (EC), Dissolved Oxygen (DO), Biological Oxygen Demand (BOD_5), Chemical Oxygen Demand (COD) and Total Coli forms ($P<0.05$). Also, Turbidity, Sulfate, Biological Oxygen Demand (BOD_5), Total Coli forms values increase and Dissolved Oxygen (DO) decrease in spring significantly ($P<0.05$). Electrical conductivity (EC), Nitrite (NO_2) and Nitrate (NO_3) increase in summer, too. With considering the decline water quality and self-purification power of Zaringol stream, it is necessary to manage the production rate and use methods of wastewater treatment based on environmental standards to avoid future risks.
Keywords: Water Quality, Trout Farm, Effluent, Zaringol Stream.

Introduction

With increasing population growth, the food requirement especially protein demand increased (Sohrabian et al., 2009). In addition, worldwide harvest of aquatic products by fisheries is closed to its maximum sustainable level of productivity and marine fishes have been under pressure of fishing, too (Arjmandi et al., 2007). Therefore, aquaculture is the only means of promoting production to meet the increasing demand for aquatic products, as currently it supply more than one-quarter of fish directly consumed by humans (FAO, 2000; Naylor et al, 2000).
Aquaculture has various impacts on the surrounding environment and ecosystem (Uzbilek Kirkagac et al., 2009). Based on report of Costa Pierce (2002), producing 1 ton fish product 0.5 ton suspended solid. Generally, this effluent release into the water bodies such as river without treatment. So, it makes to disturb balance of ecosystem and pollutes it.
Trout farm effluents consist of three main parts, first: suspended solid matters such as waste feed and fish feces; second: dissolved matters such as nitrogen compound (Urea, ammonium ion) and organic carbon; and third: chemical and pharmaceutical matters such as fungicides (Malachite green) and antibiotics (Sulfonamides) (Naderi Jelodar et al., 2007). All the items have important role in water chemical balance disorder (Selong and Helfrich, 1998).
The environmental impact of aquaculture varies and includes conflicts between the needs of different users of its products, alternation of the hydrological regime, introduction of exotic species to the wild and pollution of water resources (Pulatsu et al., 2004).
It has been reported that dissolved oxygen and pH values together with ammonia-nitrogen, total phosphorous and orthophosphate concentrations were affected by land based salmonid farms (Uzbilek Kirkagac et al., 2009).
As regards to only one percent of total water resource are fresh waters (surface waters, lakes and ponds) is used by humans so, decrease in quality of surface waters is one of the major human concern.
Therefore, the aim of this study was to assess water quality parameter of Zaringol and effect of two Trout farm effluents on it.

Materials and methods

Zaringol stream is located in Golestan province in northern Iran at 54° 43' 40" to 55° 11' 36" E and 36° 43' 30" to 37° 8' 44" N. Two Trout farms and 8 stations were selected and their water quality parameters were monitored during November 2009 to August 2010, seasonally (Fig 1).

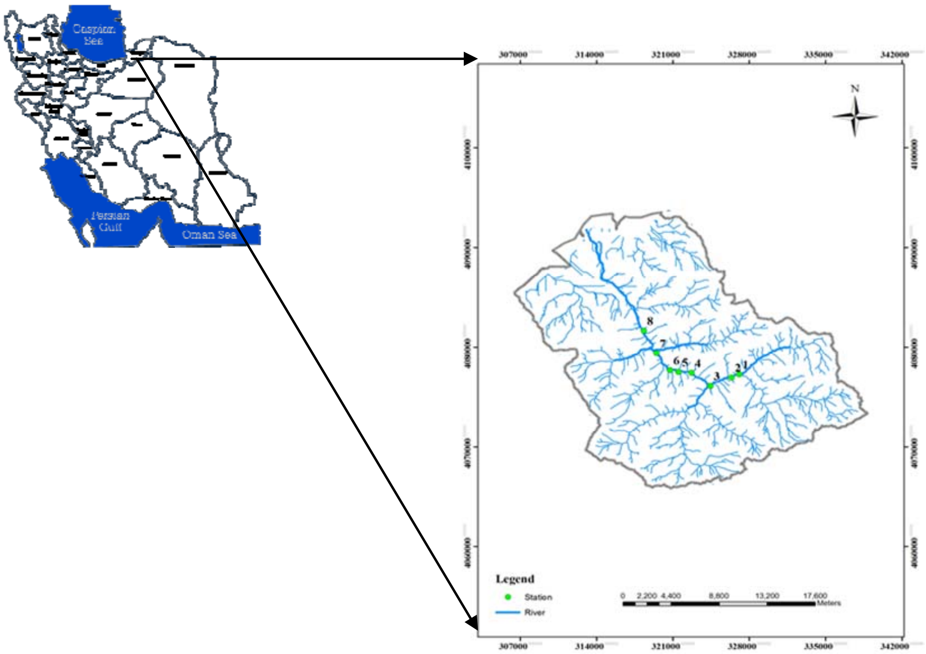

Fig.1: site of water sampling- Zaringol stream

According to Fig 1, after station 1 as a testimonial station, two Trout farms locate at station 2 and 5 and their actual capacities are 15 and 7 ton respectively. Other stations locate along the stream, between and after Trout farms and along the agricultural land use surrounded the stream.

12 number of water quality parameters including Salinity (ppt), Turbidity (NTU), Electrical conductivity (μmoscm^{-1}), Phosphate (ppm), Nitrite (ppm), Nitrate (ppm), pH, Temperature (°C), Dissolved Oxygen (ppm), Biological Oxygen Demand (ppm), Chemical Oxygen Demand (ppm) and Fecal Coli forms (colony100ml^{-1}) were measured by Water checker u-10 and Spectrophotometer.

Data checked for normality distribution with the Kolmogorov-Smirnov test. Spatial and temporal variation of water quality parameter were analyzed using one-way ANOVA and Duncan's post-hoc test, assuming a significant level of $\alpha=0.05$ by SPSS 17 software package.

Results

Data on water quality parameters of different station of Zaringol stream are given Table 1. Salinity, Turbidity, EC, PO_4, NO_2, BOD_5 and Fecal coli form increased after Trout farms (station 2 and 5) which in some parameters showed significantly increased. DO and COD showed significantly decrease, too. Other parameters such as Temperature, pH, NO_3 AND SO_4 have no difference between stations ($P<0.05$).

Table 1: Comparison of water quality parameters between different stations - Zaringol stream

Parameter/station	1	2	3	4	5	6	7	8
Salinity (ppt)	0.035[a]	0.111[b]	0.067[ab]	0.05[ab]	0.06[ab]	0.045[ab]	0.055[ab]	0.035[a]
Log Turbidity (NTU)	1.46[ab]	2.36[b]	1.26[a]	2.33[b]	2.49[b]	2.07[ab]	2.21[b]	2.05[ab]
EC (μmoscm^{-1})	0.29[a]	2.5[b]	1.59[b]	1.29[ab]	1.42[abc]	1.22[ab]	1.41[abc]	1.25[ab]
PO_4 (PPM)	0.36[ab]	0.88[b]	0.47[b]	0.16[a]	0.2[a]	0.12[a]	0.08[a]	0.06[a]
NO_2 (ppm)	0.017[a]	0.08[b]	0.03[ab]	0.02[a]	0.05[b]	0.03[ab]	0.03[ab]	0.01[a]
NO_3 (ppm)	1.37[a]	2.17[a]	1.81[a]	0.62[a]	2[a]	2.07[a]	2.27[a]	1.25[a]
pH	8.75[a]	8.58[a]	8.48[a]	8.76[a]	8.36[a]	8.56[a]	8.66[a]	8.37[a]
SO_4 (ppm)	37[a]	46.25[a]	64.43[a]	54.25[a]	55[a]	57.25[a]	56.75[a]	55[a]
T (°C)	14.05[a]	16.65[a]	13.73[a]	20.82[a]	16.97[a]	18.67[a]	19.3[a]	17.95[a]
DO (ppm)	9.12[b]	8.25[a]	8.59[ab]	9.1[b]	8.16[a]	8.45[ab]	8.7[ab]	8.52[ab]
BOD_5 (ppm)	1.77[a]	2.75[bc]	2.37[ab]	2.22[ab]	2.82[bc]	3.30[c]	3.45[c]	2.65[bc]
COD (ppm)	9.8[c]	8.85[ab]	9.75[c]	9.32[bc]	8.3[a]	8.6[ab]	8.3[a]	8.75[ab]
Fecal coli form (colony100ml^{-1})	1[a]	2.2[bc]	2.1[bc]	1.62[ab]	2.75[c]	2.2[bc]	1.92[bc]	1.55[ab]

Data present as mean.

Similar letter shows no difference between stations.

Comparison water quality parameters between different seasons shows significantly increase in Turbidity, SO_4, temperature, BOD_5 and Fecal coli form in spring and EC, NO_2 and NO_3 in summer. pH decreased in summer too. PO_4 and COD and Salinity did not show significantly difference between seasons (Table 2; $P<0.05$).

Table 2. Comparison of water quality parameters between different seasons - Zaringol stream

Parameter/station	Spring	Summer	Autumn	Winter	Total
Salinity (ppt)	0.038[a]	0.083[a]	0.064[a]	0.403[a]	0.057
Log Turbidity (NTU)	2.24[b]	1.93[ab]	1.6[a]	2.36[b]	2.03
EC (μmoscm^{-1})	1.17[ab]	1.88[b]	1.54[ab]	0.88[a]	1.37
PO_4 (PPM)	0.45[a]	0.42[a]	0.16[a]	0.12[a]	0.29
NO_2 (ppm)	0.046[ab]	0.063[b]	0.013[a]	0.01[a]	0.033
NO_3 (ppm)	1.87[b]	2.23[b]	2.35[b]	0.33[a]	1.7
pH	8.56[b]	8.25[a]	8.72[b]	8.74[b]	8.57
SO_4 (ppm)	88.87[c]	56.62[b]	41.09[ab]	26.37[a]	53.24
T (°C)	24.07[c]	21.03[b]	10.21[a]	13.75[a]	17.27
DO (ppm)	8.2[a]	8.86[b]	8.68[ab]	8.7[ab]	8.61
BOD_5 (ppm)	2.87[b]	2.7[ab]	2.96[b]	2.13[a]	2.67
COD (ppm)	8.76[a]	8.88[a]	8.9[a]	9.28[a]	8.96
Fecal coli form (colony100ml^{-1})	2.52[b]	1.96[b]	1.65[a]	1.55[a]	1.55

Data present as mean.

Similar letter shows no difference between seasons

Discussion

The effect of fish farm effluents on receiving waters depending on local condition such as volume and concentration of substances, flow rate of water and time of effluent discharge (Pillay, 2004).

Results of Dissolved Oxygen concentration reveal a reduction in upstream and downstream of the outlet of Trout farms. It decrease from 9.12 to 8.25 (station 1 to 2) and 9.1 to 8.16 (station 4 to 5). Also it shows seasonally change and minimum value belongs to the summer. With considering that the releasing time of fry is end of May and period of producing is about 5-6 months, so it seems this decline relies to fish farms activities. Limited value reported for DO concentration is 6 ppm (EPA, 1996). So, the lowest value still exceeded the upper limit of DO concentration. Result of BOD_5 changes shows that Biological Oxygen Demand increase after the outlet of Trout farms. It increases in spring, too. Since that, with increasing organic matters, increasing Oxygen required for aerobic decomposition. So, when organic matter increased, BOD_5 value increases. Although, composition of Trout farm effluent has important role in Oxygen consumption, increase of BOD_5 approves effect of their effluent on decrease of DO. USEPA[1] reports BOD_5 range for clear, related polluted and polluted water are 0-2, 3-5 and > 5 ppm, respectively. According to this classification, water is related polluted except in station 1.

Phosphate ion is a water quality parameter had high correlation with geology and usually less than 0.1 ppm (EPA, 1996) in rivers. Mean phosphate concentration shows that its value exceeds the standard value spatially in stations 2, 3, 5 and warm seasons. One of the probably reason is entrance of effluent of Trout farm and agricultural land use into the stream. Although geology and slop is effective.

Costa Pierce (2002) reported production of 1 ton fish produced 510 kg solid matters, 108 kg Nitrogen and 19 kg Phosphor can be found as their compounds in the environment. Our results confirm that both of Nitrite, Nitrate and Turbidity increase in station 2 and 5. Standard value of NO_2 and NO_3 in surface water are 0.51 and <1 ppm (EPA, 1996). So, their mean concentration show that NO_2 is at the normal range but the NO_3 is higher.

Electrical Conductivity represents power of water electricity transmission and amount of dissolved ions, approximately (Allan, 1995). Electrical Conductivity variation depends on effluent entrance, erosion of bed, sides and dissolved ions. Results show it increases at the outlets of fish farms, too.

There are many reports on impact of aquaculture effluents on water quality. Naderi Jelodar et al (2007) studied the effects of Trout farm effluents on water quality parameter of Haraz River and represented DO decreased and Turbidity, BOD_5, TSS and NH_4 increased significantly. Pulatsu et al (2004) assess impact of Rainbow Trout farm effluents on water quality of Karasu Stream (Turkey) and found that DO decreased and Turbidity, NO_2, NO_3, total Phosphorus, TSS and NH_4 increased in downstream. Ghanea Sasan Saraei (2005) studied effect of three Trout farm on water quality of Haraz River and reported DO decreased and pH, EC, BOD_5, NO_3 and NH_4 increased in downstream significantly. Our results accordance with previous studies.

Finally, we can conclude Trout farm effluent effect on some water quality parameter such as DO, BOD_5, Turbidity, EC, NO_2 and PO_4 significantly. Also, its effect increased on seasons were period of production (spring and summer). Capacity of production is another parameter had effect on water quality variation. As we observed in station 2 some parameters (EC, PO_4) show more increase than station 5.

With considering that some parameters are higher than standard value (PO_4, NO_3 and BOD_5), it is necessary to manage the production rate and use methods of wastewater treatment based on environmental standards to avoid future risks.

Acknowledgment

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The effect of bacteria containing ACC-deaminase enzyme on Zea mays chlorophyll content in a lead and cadmium polluted soil

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Abstract

ACC-deaminase enzyme in plant growth promoting bacteria (PGPR) increases plant growth by decreasing ethylene concentration in highly heavy metal polluted soil. The objective of this study was to investigate the effect of seed inoculation by two species of Pseudomonas bacteria (with and without ACC-deaminase enzyme) on Zea mays chlorophyll content in presence of various levels of lead and cadmium in a soil. A pot experiment was carried out in split plot factorial based on complete randomized block design in three replications in a research greenhouse. For this reason, soils were artificially contaminated with lead (500, 1000 and 1500 mg $Pb.kg^{-1}$soil) and cadmium (3, 5 and 10 mg $Cd.kg^{-1}$soil). The results indicated that the existence of Pseudomonas containing ACCD enzyme increased leaf chlorophyll content of Zea mays in high concentration of both metals, especially in the highest level of cadmium. It seems that pseudomonas bacterium containing ACC-deaminase enzyme is a potent plant growth due to its stability and persistence in the presence of heavy metals.

Keywords: ACC-deaminase enzyme, Phytoremediation, Chlorophyll, Lead, Cadmium.

Introduction

Heavy metals are important environmental pollutants present in soils and toxic levels of some of them (cadmium, copper, lead, etc.) could appear in natural and agricultural areas as a result of anthropogenic activity. Heavy metals are implicated in the generation of oxidative stress in plant cells (Groppa et al., 2007). Cadmium (promotes the formation and exudation of proteinogenic and non-protein amino acids in maize roots (Bergmann et al., 1996) and the biosynthesis of polyamines in plants, as well (Smith, 1985; Bergmann, 1996).

Phytoremediation is the use of green plants to remove contaminants from soil and groundwater or to lower contaminant mobility (Madrid et al., 2003). It has become a promising remediation technique with the discovery of hyperaccumulators plants that are able to take up large quantities of metals (Roosens et al., 2003). Achievement of hyperaccumulator status is based on the ability to uptake and retain within the shoot (stem and leaves) (Roosens et al., 2003; Turgut et al., 2005). Plants do not accumulate sufficient biomass (particularly roots) in heavily contaminated soils for effective remediation.

At present, plant-growth-promoting rhizobacteria (PGPR) *Pseudomonas* are quite an attractive research object in view of their potential application for phytoremediation. Reduced root growth in contaminated soils might be owing to stress-induced ethylene, an established phytohormone. ACC is the immediate precursor of ethylene in plants (Abeles et al., 1992). Interestingly some plant growth-promoting rhizobacteria (PGPR) contain the enzyme 1-aminocyclopropane-1-carboxylate (ACC) deaminase, which metabolizes ACC into α-ketobutyric acid and ammonia, and thus regulates the biosynthesis of ethylene in inoculated plant roots (Glick, 2005). The resultant increase in root growth provided by ACC deaminase might therefore enhance the effectiveness of phytoremediation processes in contaminated soil (Huang et al., 2004).

Heavy metal contamination of soil is often associated with iron deficiency in a range of different plant species (Wallace et al., 1992; Ma and Nomoto, 1993). Their low iron content generally inhibits both chloroplast development and chlorophyll biosynthesis (Imsande, 1998). However, microbial iron–siderophore complexes can be taken up by plants as a source of iron (Wallace et al., 1992; Burd et al., 2000), thus facilitating plant growth as observed for mung plants inoculated with siderophore-producing bacteria and grown in the presence of $CdCl_2$ (Tripathi et al., 2005).The study of Ali and *et al. (*2003) on the effects of lead, nickel and copper on species *salix acmophylla Boiss* showed that all the three metals reduced total chlorophyll content in concentration dependent manner. The principal aim of this study was to investigate the effect of two species of

Pseudomonas bacteria (with and without ACC-deaminase enzyme) on Zea mays chlorophyll content in a Pb and Cd contaminated soil.

Materials and methods

- Pot experiment

A pot experiment was carried out in split plot factorial based on complete randomized block design in a research greenhouse with two kinds of heavy metal (Pb and Cd) in three levels: (500, 1000, 1500 mg Pb/kg soil and 3, 5, 10 mg Cd /kg soil , respectively), 3 levels of bacteria (free of bacteria as a control, bactera *Pseudomonas putida* strain 11 with ACC- deaminase enzyme, and *Pseudomonas putida* strain 153 without ACC-deaminase enzyme) with three replicates. Both bacteria were able to produce siderophore and were resistant to Cd and Pb. The soil was sampled from noncontaminated agricultural fields, air dried and passed through a 2 mm (10 mesh) sieve. Chemical and physical properties of studied soil are presented in table 1. To prepare the contaminated soils different concenterations of Pb and Cd were sprayed to the soil. Zea mays seeds were surface-sterilized with a solution of 1.5% (v/v) sodium hypochlorite for 10 min and washed with sterile water. Then they were inoculated with bacteria and were planted in pots containing polluted soil and sand. The pots were placed in a greenhouse with a 25 ^{o}c temperature and were watered with deionized one to maintain the moisture content at approximately 60% water-holding capacity (WHC) of the soil.

At the end of experiment, plant samples were collected, divided into above-aground shoots and roots and washed three times with deionized water. To determine the dry weight, shoots and roots oven-dried separately at 75 ^{o}c and were ground to 0.5 mm for analysis. Then shoots and roots were digested with the HNO_3/HCL method (Harmon and Lajtha, 1999) and the Pb and Cd concentration was meausured with an AAS.

- Determination of chlorophyll content

The chlorophyll content of leaf samples were determined by spectrophotometer (Hiscox and Israelstam, 1979).

Table 1. Chemical and physical charactristics of the soil

Texture	pH	EC	OM	P[a]	K[a]	Total content of elements (mg kg^{-1})		
		(ds m^{-1})	*(%)*			**Fe**	**Pb**	**Cd**
Clay Loam	7.1	1.5	0.8	17	280	185.5	11	0

[a] The datas are given as Phosphorous and Potassium concentration is available content (mg kg^{-1}).

Results and discussion

The highest and lowest chlorophyll content was related to 500 and 1000 mg pb/kg soil respectively (fig. 1). In the existence of *Pseudomonas* containing ACCD-enzyme, chlorophyll content nonsignificantly was increased in comparison with *Pseudomonas* without ACCD-enzyme at the concentration of 500 mg Pb kg^{-1} and 3 and 10 mg Cd kg^{-1} (fig 1. and 2.). In the absence of both bacteria, increases of the content of leaf chlorophyll in zea mays were obtained when the soil was contaminated with Pb, althouge it wasent significant (fig. 1).

Kumar et al. (2008) showed that in presence of heavy metals significant increase in chlorophyll content, was observed when siderophore over producing mutant NBRI K28 SD1 was added to plant as bioinoculant.

Bacterial inoculation (with and without ACCD enzyme) led to increasing leaf chlorophyll content at the lowest level of Cd and Pb. The highest chlorophyll content was abserved in exsistence of *Pseudomonas* produce ACCD – enzyme in 10 mg/kg Cd concentration (fig. 2). Dell Amico et al. (2008) reported that in the absence of cadmium, the inoculation of *B. Napus* seeds with the PGPR did not affect the content of leaf chlorophyll. But, these chlorophyll contents increased significantly when *B. napus* inoculated with *P. fluorescens* ACC9 and *P. tolaasii* ACC23 (were able to produce

IAA, siderophore and ACCD – enzyme) in a cadmium-contaminated soil. *Pseudomonas putida* strain 11 seems to play a dual role in facilitating plant growth in the presence of cadmium and lead. On the other hand the bacteria enables the plant to acquire sufficient iron in the presence of heavy metal levels that might otherwise make its acquisition difficult (Burd et al., 2000).

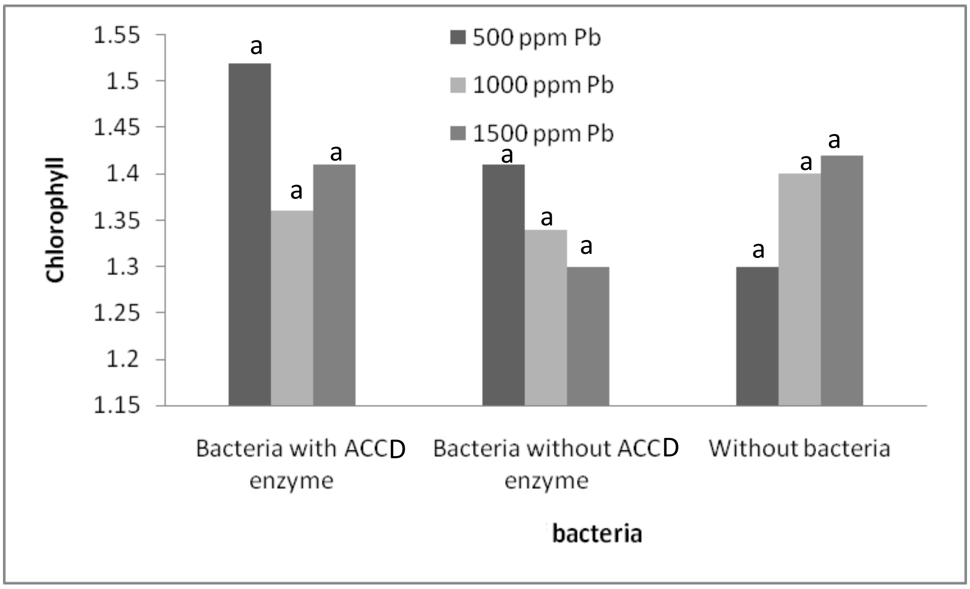

Figure 1. The effect different strains of bacteria on chlorophyll content of Zea mays, at different concentration of Pb (mg kg^{-1})

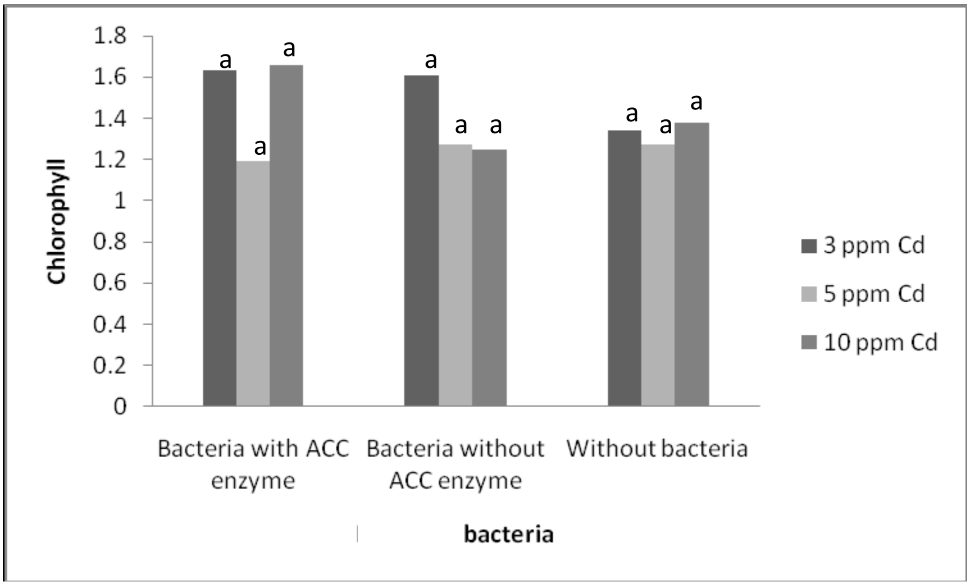

Figure 2. The effect different strains of bacteria on chlorophyll content of Zea mays, at different concentration of Pb (mg kg^{-1})

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Influence of Heavy Metals Pollution on Quality of Coastal Sediments near Modrac Lake

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Abstract
Influence of heavy metals polution on quality of coastal sediments near Modrac Lake has been investigated in this study. The investigation has been performed at two locations from the right side of the Spreca River, immedietly in front of its mouth in Modrac Lake at the water level of 197.2 meters above the sea level. Samples were taken at the distance 50 m from Spreca river and 50 m from Modrac Lake (L1) and 100 m from Spreca River and 100 m from Modrac Lake (L2). The obtained results showed that concentration of Ni was the highest at the both locations (206 and 292 $mgkg^{-1}$ respectively) as well as concentration of Cr (67.6 and 129.6 $mgkg^{-1}$ respectively). The obtained concentrations of Cd, Hg, Pb, Mo, As and Co were below of $0.00001 mgkg^{-1}$. The increased concentration of Ni in the sediment came from the coal mining, and it is of antropogenic origin.
Keywords: sediment, heavy metals, polution, exploatation of coal

Introduction

Harmful substances that reach the aquatic environment are subject to dilution, and various chemical changes. Some matters are broken down relatively quickly, while those that are most resistant accumulate in the sediment and may bio-accumulate in aquatic organisms.

Waste matter can cause a various changes in the water such as reducing transparency, color changes, changes in the composition of benthic communities, the reduction of biomass of valuable species of economic value.

Different mass portions of heavy metals occur in naturally contaminated sediments (Prohić et al., 1989).

The degree of contamination is determined by mass portion of heavy metals in sediment. The higher mass portion of heavy metals in the surface layer of sediments indicates recent contamination (Bogner et al., 1998).

The binding capacity of the sediment is determined by its granulometric composition and organic matter content (Ujević et al., 2000; Ujević et al., 1998).

Sediment with high organic content has a high affinity to acumulate many organic and anorganic contaminants.

The content of organic matter is usually related to granulometric composition of the sediment (Ujević et al., 2000).

Heavy metal concentrations of coastal sediment samples near Lake Modrac have not been studied previously and this study highlight the first data on the heavy metal contamination degree from this area.

Materials and Methods

Study area

Modrac Lake is located in Bosnia and Herzegovina, in Tuzla Canton, near the town of Tuzla. Modrac Lake is the biggest artificial lake in the region, it was created in 1964 by building a dam on the Spreča River. The 11 km long and 1.6 km wide lake covers about 17 km^2 area. Modrac Lake was originally created for industrial purposes but nowadays, the lake is used for drinking water supply and for supplying drinking water to the city of Tuzla.

The basin of the lake covers 1189 km^2, of which the Spreča River basin belongs 832 km^2, accounting for 70% of the lake basin area, Turija River basin belongs 240 km^2, accounting for 20%

of the lake basin area and the immediate basin of the lake 117 km^2, accounting for 10% of the the lake basin area.
Many outlets of industrial and urban wastewater discharge their untreated efluents into the lake directly or through the rivers Spreča and Turija. The lake also receives water from agricultural and urban runoff. Such discharges usually contain heavy metals and organic matter. During 45 years usable volume of Lake Modrac is reduced about 15 x 10^6 m^3 due to sediment entering, large amounts of organic pollution, nutrients and suspended material, through the tributaries and eutrophication phenomena, especially in the shallower parts of the reservoir. According to estimate, Lake Modrac receives yearly pollution load of about 700,000.00 population equivalent.

Sample collection
All sediment samples were collected on 21st of July, 2011 at two localities from the right side of the Spreča River, immedietly in front of its mouth in Lake Modrac at the water level of 197.2 m above sea level.
Samples were taken at the distance 50 m from Spreča River and 50 meters from Lake Modrac (L1) and 100 m from Spreča River and 100 m from Lake Modrac (L2).
At both localities (L1 and L2) the thickness of sediment is determined, by piercing the sediment with steel rod to the hard nature of the soil. Determined sediment thickness ranges from 824 mm for L1 locality, and 757 mm for the locality L2.
Sampling was carried out using an Ekman grab on the surface layer of sediment, about 1.5 kg from each locations. (Figure 1).

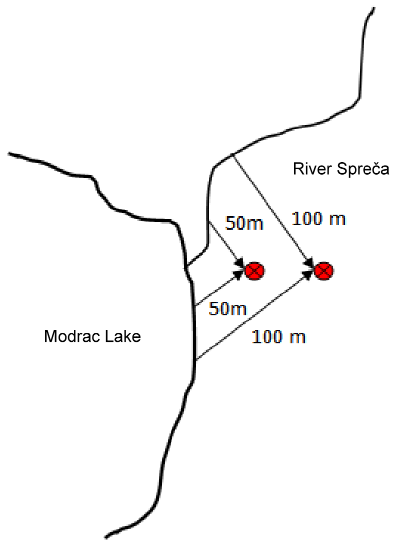


Figure 1. *Sample collection sites*

The samples were placed in plastic bags, frozen at -18 °C and kept at this temperature until analyzed in laboratory.

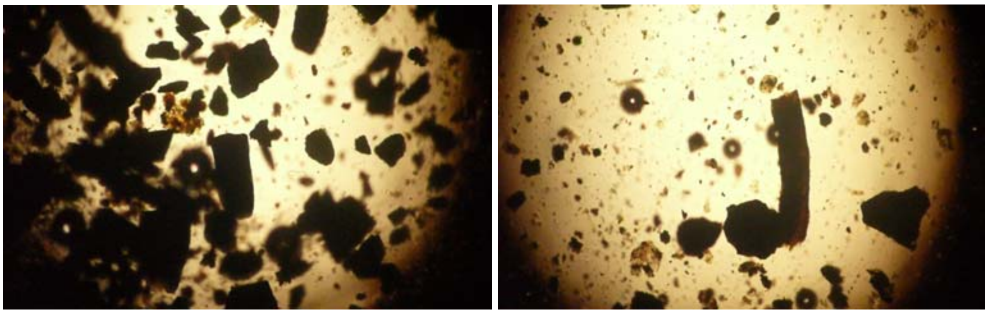
Methods

Sediment samples were dried in the laboratory at room temperature in a shallow plastic tubs and sieved. Laboratory determined the mineralogical and granulometric composition of sediment and the concentration of Ni, Cr, Cu, Zn and Mn. Granulometric composition was determined by sieving on woven wire mesh sieves with diameter d = 2,000 mm, d = 1,000 mm, d = 0,200 mm and 0,063 mm.
To determine concentrations of heavy metals the basic solution of the sediment samples was prepared by the destruction of dry sample digestion in aqua regia according to ISO 11464:1994. Nickel, Cr, Cu, Zn and Mn were determined by Inductively Coupled Plasma-Optical Emission

Spectrometer, ICP-OES OPTIMA 2100 DV with the standard of 100 $mgdm^{-3}$ for metals. Data presented are avarage of three replications.
Mineralogical composition of sediments was determined by petrographic examination of transparent preparations by Polarization Microscope Leica DM 2500 with magnification of 200 times. Parallel and crossed nicols for medium samples from both sites (L1 and L2) were prepared.

Results and discusion

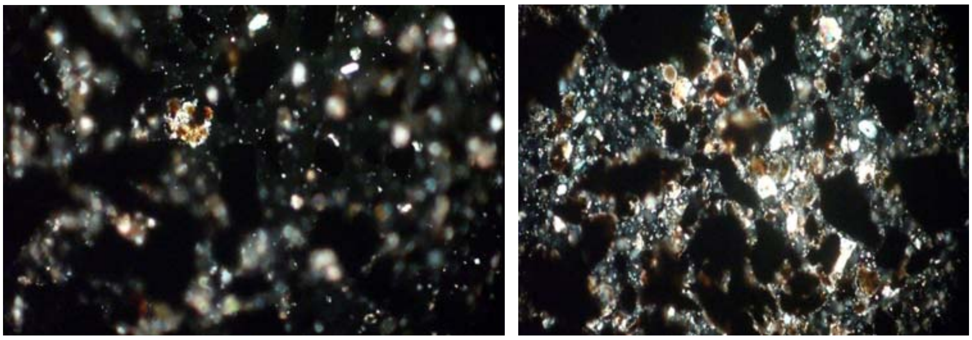
Mineralogical analysis shows that the composition of the sediment from Modrac Lake is a mixture of silty material (clay and force) and coal dust particles.
Particles of coal dust belong to the so-called opaque minerals (fr. opaque - black, opaque), and are seen as black angular structure in a parallel nicol (Figure 2).



L1 *L2*

Figure 2. *Parallel nicol*

Siliceous material is formed in a natural way of decomposition and is visible in crossed nicol of samples (Figure 3). It is assumed that the siliceous material from the samples is from the composition of direct slope of Banovići and Đurđevik basin so called "Bosnian Serpentinites Zone" which is built from basic and the ultra basic rocks.



L1 *L2*

Figure 3. *Cross nicol*

From the mineralogical analyses we can conclude that the composition of sample is powdered material that in the addition to coal dust particles has a share of marl - clay and silicate materials originating from the "Bosnian Serpentinites Zone".
Granulometric composition of sediment at both localities is given in Table 1. The samples represented the highest portion of fractions from 0.063 to 0.2 mm, from which it can be concluded that the sample belongs to the sandy powder materials, which are represented in the largest quantity of sand sized particles (about 70%) and particle size of silt and clay (about 30 %).

Table 1. Granulometric composition of sediment

Diameter of grain d/mm	Class fraction (%)	
	L1	*L2*
> 2,000	0,16	0,04
1,000-2,000	0,58	0,44
0,200-1,000	22,82	21,22
0,063-0,200	51,26	53,54
< 0,063	25,18	24,76

Minimum and maximum detected concentrations of metallic elements on both localities are given in Table 2.

Table 2. Minimum and maximum detected concentrations of tested metallic elements on both localities

Metals ($mgkg^{-1}$)	***L1***			***L2***		
	min	***max***	***average***	***min***	***max***	***average***
Ni	141,4	319,8	206,0	238,1	359,9	292,0
Cr	41,0	87,6	67,6	106,7	146,7	129,6
Cu	29,6	50,3	42,8	41,5	60,7	42,5
Zn	29,7	49,8	38,89	51,4	124,5	84,5
Mn	325,3	555,3	432,0	687,7	1502,5	1072,3

Graphical display and evaluation of sediment quality in relation to the content of each metal are given below.

Sediment quality was assessed by comparison with the Canadian guidelines, national legislation according to limit values depending on the soil texture, the legislation of the Republic of Serbia, as well as using the Dutch methodology (Table 3).

Table 3. Target value and intervention value according to Dutch and Canadian law and MDK values of toxic matter in soil according of law of Federation of Bosnia&Herzegovina and Republic of Serbia

Metals ($mgkg^{-1}$)	***Dutch methodology***		***Canadian guidelines***		***Federation of Bosnia&Herzegovina***	***Republic of Serbia***
	Target value	Intervention value	ISQG	PEL	MDK of toxic matter in soil	MDK of toxic matter in soil
Ni	35	210	*	*	30	50
Cr	100	380	37,3	90	50	100
Cu	36	190	35,7	197	*	100
Zn	140	720	123	314	100	300
Mn	*	*	*	*	*	*

ISQG - interım freshwater sedıment qulıty guıdelınes; PEL - probable effect levels; * - not defined;
MDK - maximum allowed value

The results of analyses of metals in coastal sediments near Modrac lake on both localities (L1 and L2) are present on Figure 4.

The nickel content in the sediment ranged from 206 $mgkg^{-1}$ to 292 $mgkg^{-1}$. Limits are not defined by Canadian law. When applying the Dutch methodology all of the samples are classified as polluted sediments and are classified into four classes, which indicates that it is extremely contaminated sediment and needs to be remediated, and according to national legislation, the content of nickel was exceeded allowable values (Table 3).

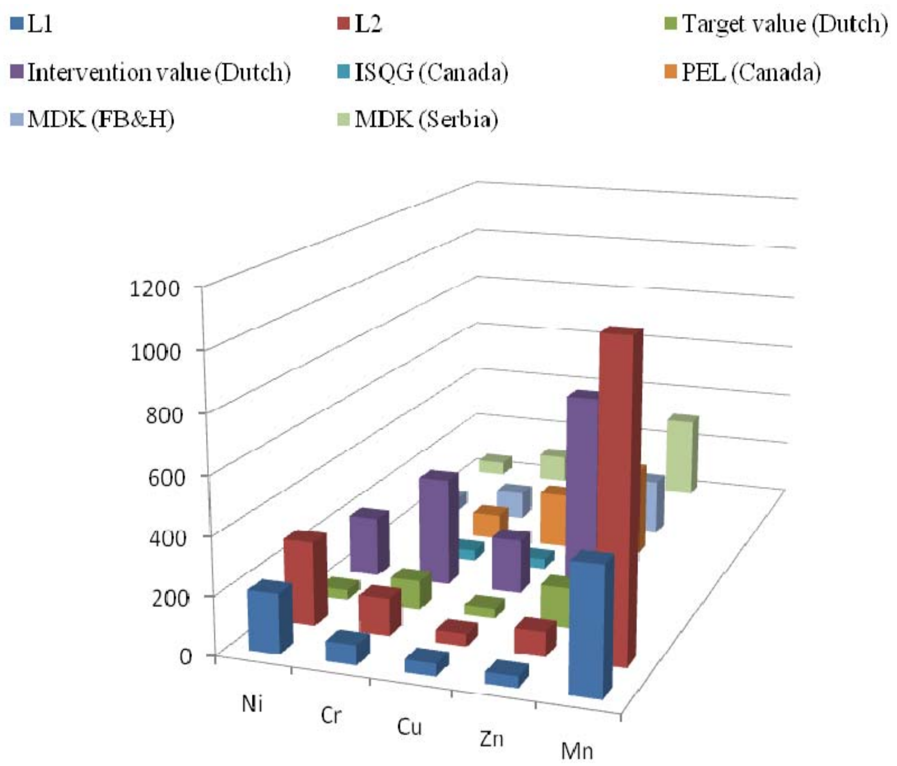


Figure 4. Concentration of metals on localities L1 and L2

The high nickel content in the sediment probably comes from Banovići Coal Mine which directly discharged its untreated wastewater into the Spreča River, and further in Modrac Lake.

The content corresponds to the concentration of nickel in coal from Banovići Coal Mine (291 $mgkg^{-1}$) (data source: "Reintegration of Coal Ash Disposal Sites and Mitigation of Pollution in the West Balkan Area" (Project No. 509173; www.rhizo.at/RECOAL) funded by the 6th Framework Programme of the European Union).

The content of chromium in the sediment ranged from 67.6 $mgkg^{-1}$ to 129.6 $mgkg^{-1}$. In the samples from locality L1 chromium content was above the Canadian theoretically possible effect value, while on locality L2 chromium content was above the Canadian empirically possible effect value, which indicates the potential ecotoxic effect. Also, the concentration of chromium exceeded the MDK (maximum allowed value) value of heavy metals in the soil under the legislation of Federation of Bosnia&Herzegovina and Republic of Serbia.

Comparing with current recommendations and quality standards for sediment, the concentration of copper was above the Canadian theoretically possible effect value and above Dutch reference values. These results indicate possible negative effects. If we compare results across localities it can not be seen significant differences in the content of copper in sediment.

The content of zinc in the analyzed sediment samples ranged from 38.89 $mgkg^{-1}$ to 84.5 $mgkg^{-1}$. Comparing with current recommendations and standards, it can be seen that the sediment quality in terms of zinc content on both localities is satisfying.

The content of manganese in the analyzed sediment samples ranged from 432 $mgkg^{-1}$ to 1072.3 $mgkg^{-1}$. Limit values are not defined by any of quality standards. The occurrence of manganese in samples of sediment is found in the fact that coal from Banovići Coal Mine in its composition contains 225 $mgkg^{-1}$ Mn (data source: "Reintegration of Coal Ash Disposal Sites and Mitigation of Pollution in the West Balkan Area" (Project No. 509173; www.rhizo.at/RECOAL) funded by the 6th Framework Programme of the European Union).

In all of the samples from both locality, concentration of cadmium, mercury, lead, molybdenum, arsenic and cobalt were under 0.00001 $mgkg^{-1}$.

Conclusions

From the results of analyses of quality of coastal sediments near Lake Modrac we derive the following conclusions:

- Mineralogical analysis shows that the composition of the sediment is a mixture of silty material (clay and force) and coal dust particles.
- The samples represented the highest proportion of fractions from 0,063 to 0,200 mm.
- The high nickel content in the sediment, and the presence of manganese most likely comes from Banovići Coal Mine wastewater.
- It is recommended to continually monitor coastal sediments pollution degree near Lake Modrac and examine their impact on other parts of the ecosystem (primarily on aquatic organisms).

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Growth and Physiological Responses of Rice Cultivars to Cadmium Toxicity

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Abstract
Contamination of soils to Cadmium (Cd) is one of the most important agricultural and environmental issues in world. Screening of rice cultivars is a useful method for rice production in soils with high Cd levels. To investigate the effect of Cd toxicity on growth, Cd concentration, and physiological responses of seven rice cultivars, a pot experiment was conducted in a completely randomized design with three Cd levels (0, 45, and 90 mg kg^{-1} soil) and seven rice cultivars (Ghasrodashti, Khazar, Anbarboo, Dasht, Hasani, Taroom, and Kadoos). There are considerable variations among rice cultivars in shoot Cd concentrations in the range of 0.25 to 16.55 mg kg^{-1}, with mean of 6.3 mg kg^{-1}. Dry weight of roots and shoots and number of main and lateral tillers decreased significantly when Cd level in soil was increased to 45 mg kg^{-1}. However no significant differences were observed between 45 and 90 mg kg^{-1} Cd levels. Application of 90 mg Cd kg^{-1} soil decreased shoot height, number of lateral, main, and total tillers, shoot dry weight, and mean relative growth rate (RGR) of seven rice cultivars by 35, 13, 88, 40, 65.5, and 65 % as compared to those of control, respectively. Shoot dry weight, and height of plants showed significant and negative correlation with Cd concentration in shoot. There were significant differences among cultivars to Cd toxicity. Our tentative conclusion is that Hasani cultivar was superior cultivar compared to other studied cultivars due to its greater performance, whereas Khazar cultivar was the most sensitive cultivar.
Keyword: Cadmium; Rice cultivars; Root and shoot growth parameters

Introduction
Cadmium contamination in soil is now an important agricultural and environmental issue throughout the world (Obata and Umebayashi 1997). Cadmium can be effectively absorbed by plant roots and translocated very easily to shoots (Kabata-Pendias and Pendias 2001). Plant metabolisms, such as photosynthesis and nutrient absorption have been affected by Cd (Wu et al., 2003). As a result, plant growth is inhibited by Cd application to growth medium (Wu et al., 2007). Limited information is available on the effect of Cd on growth of different rice cultivars. Therefore, the objectives of this experiment were to study 1) the effect of Cd toxicity on some growth and some physiological responses of seven rice cultivars, 2) influence of Cd toxicity on the concentration of Cd in seven rice cultivars.

Materials and Methods
Bulk soil sample was collected from surface horizon (0-30 cm) of a Typic Xerorthents in Bajgah Agricultural Station of Shiraz University, Iran. The soil sample was air dried, passed through a 2-mm sieve, and used for laboratory analyses. Selected properties of soil were determined to be as follows: saturation extracts electrical conductivity (ECe) of 0.79 dS m^{-1}; saturation paste pH of 7.55; organic matter (OM) of 0.5%; DTPA-extractable Cd of 0.37 mg kg^{-1}. To investigate the effect of Cd toxicity on growth, Cd concentration, and physiological responses of seven rice cultivars, a pot experiment was conducted in a completely randomized design with three replications. Treatments consisted of three soil-applied Cd levels for contamination of soil (0, 45, and 90 mg kg^{-1} soil as $CdSO_4$) and seven rice cultivars (Ghasrodashti, Khazar, Anbarboo, Dasht, Hasani, Taroom, and Kadoos). Rice cultivars were sown in each pot (63 pots, each containing the equivalent of 4 kg dried soil). Pots were maintained under flooded conditions (with 3 cm of water standing above soil surface) during the vegetative stage. Eight weeks after emergence shoot height, and number of lateral, main, and total tillers of the plants were measured. Then, whole rice plants were harvested and washed. Roots and shoots were oven-dried at 70°C to constant weight. Oven-dried samples were ground using a stainless steel grinder. The concentration of Cd was measured by atomic absorption spectrophotometer. Statistical analysis was performed using SPSS and Excel statistical software packages.

Results and Discussion

Rice height decreased significantly by 23 and 35 % at 45 and 90 mg Cd kg^{-1} soil, respectively, due to the effect of Cd toxicity on rice growth (Table 1). Similarly Tiryakioglua et al. (2006) found that application of Cd decreased shoot height of two barley genotypes. Huang et al. (2007) reported that Cd toxicity led to a significant decrease in plant height of four barley genotypes. Reduction in plant height might be mainly due to the reduction of root growth and lesser transport of nutrients to the above parts of the plant, and consequently disturbing cellular metabolism of shoots (Shanker et al., 2005).

Significant differences were observed in the term of reduction of height among rice cultivars (Table 1). Ghasrodashti and Hasani cultivar had the highest height, whereas Khazar cultivar had the lowest height among other cultivars (Table 1).

Average of number of lateral and total tillers in rice cultivars decreased significantly with increasing Cd levels. However, they were not significantly different between 45 and 90 mg Cd kg^{-1} treatments. In comparison with control, addition of 90 mg Cd kg^{-1} soil decreased mean number of lateral and total tillers by 88 and 40 %, respectively (Table 1). The number of main tillers in rice cultivars decreased with increasing Cd level. Application of 90 mg Cd kg^{-1} soil significantly decreased mean number of main tillers by 13 %, as compared to that of control (Table 1).

The difference in number of tillers was observed among rice cultivars (Table 1). Kadoos and Taroom cultivar had the highest and lowest mean total tiller, respectively.

Table 1. Effect of Cd levels on height and number of main, lateral, and total tillers of seven rice cultivars

	Height (cm)	Main tiller	Lateral tiller	Total tiller
Cd levels (mg kg^{-1})	each figure is average of 7 rice cultivars			
0	96 a*	30 a	17 a	47a
45	74 b	28 ab	4 b	33b
90	62 c	26 b	2 b	28b
Cultivar	each figure is average of 3 Cd levels			
Ghasrodashti	107 a	30 a	8 b	38 b
Khazar	57 c	29 ab	1 c	30 bc
Anbarboo	79 b	25 b	7 ab	32 bc
Dasht	69 bc	27 ab	10 b	37 bc
Hasani	97 a	30 ab	4 ab	34 bc
Taroom	66 bc	28 ab	0 c	29 c
Kadoos	65 c	27 ab	23 a	50 a

* Means in each column followed by the same letters are not significantly different ($p \leq 0.05$) by Duncan's Multiple Range Test

Results showed that application of 45 and 90 mg Cd kg^{-1} soil decreased mean dry weight of all cultivars by 49.9 and 65.6 % for shoots and 37 and 45.7 % for roots, respectively, as compared to control (Table 1). Thus, shoots dry weight was more affected by Cd levels than that of roots. There were no significant differences on average of shoots or roots dry weights between 45 and 90 mg Cd kg^{-1} treatments. Reduction in root growth due to trace elements toxicity could be due to the inhibition of root cell division, root elongation, or to the extension of cell cycle in the roots (Barcelo et al., 1986). Our results are in close agreement with findings of Liu et al. (2010) who stated that application of 50 mg Cd kg^{-1} soil decreased root and shoot weights of two rice cultivars. Hu et al. (2009) showed that Cd stress (50 μM) decreased dry weight of rice seedling by 36.7% as compared to control.

Rice cultivars showed significant differences in terms of decrease in shoots and roots dry weights as affected by Cd application (Table 2). Mean shoot dry weight of Hasani cultivars was 7.48 g pot^{-1} and it was significantly highest among other studied cultivars. Application of 90 mg Cd kg^{-1} soil decreased shoot dry weight by 46, 59, 77, 80, 47, 72, and 72 % in Ghasrodashti, Khazar, Anbarboo, Dasht, Hasani, Taroom, and Kadoos cultivar as compared to that of control, respectively (Table 2). Hasani and Ghasrodashti had the lowest decrease in shoot growth, indicating the highest tolerance to Cd toxicity. According to Adhikari et al. (2006), rice yield was decreased by application of Cd to nutrient solution; however, yield response varied among the cultivars.

Results showed that addition of 45 and 90 mg Cd kg^{-1} soil significantly decreased RGR in all seven cultivars by 50 and 64.7%, respectively, as compared to that of control (Table 2). Abo-Kassem et al. (1995) also observed that RGR of wheat plants decreased by 20 % with application of 10 µM Cd. Vasillev and Yordanov (1997) reported that RGR of wheat and sugar beet plants decreased with application of Cd to soil. The order of RGR was Hasani> Ghasrodashti> Kadoos> Taroom> Khazar> Anbarboo> Dasht cultivar respectively. However, no statistically significant differences were observed among Hasani, Ghasrodashti, Kadoos, Taroom, Khazar, Anbarboo cultivars (Table 2).

According to our results shoot Cd concentration in the seven cultivars significantly increased with increasing Cd levels (Table 2). Our results were in close agreement with findings of others. Qin et al. (2009) noted that shoot and root Cd concentration in two rice genotypes increased with addition of 0.1 to 5 µM Cd under hydroponic culture. There are considerable variations among seven rice cultivars in shoot Cd concentrations in the range of 0.25 to 16.55 mg kg^{-1}, with mean of 6.3 mg kg^{-1}.

Shoot dry weight and height of rice plants showed significant and negative correlation with Cd concentration in shoots ($r = -0.426^{**}$ and $r = -0.336^{**}$, respectively).

Table 2. Effect of Cd levels on shoot and root dry weight, RGR, and shoot Cd concentration of seven rice cultivars

	Shoot dry weight (g pot^{-1})	Root dry weight (g pot^{-1})	Shoot RGR (%)	Shoot Cd concentration (mg kg^{-1})
Cd levels (mg kg^{-1})	each figure is average of 7 rice cultivars			
0	8.13 a*	0.81 a	79.1 a	1.59c
45	4.07 b	0.51 b	40.5 b	7.42b
90	2.80 b	0.44 b	27.9 c	9.87a
Cultivar	each figure is average of 3 Cd levels			
Ghasrodashti	4.86 bc	0.49 bc	54.6 a	4.13 d
Khazar	3.17 c	0.18 c	51.7 ab	4.47 d
Anbarboo	5.16 bc	0.51 bc	40.3 ab	7.88 ab
Dasht	4.82 bc	0.54 bc	34.7 c	5.94 cd
Hasani	7.48 a	1.27 a	56.8 a	9.58 a
Taroom	3.70 c	0.31 c	52.3 ab	4.56 d
Kadoos	5.83 ab	0.78 b	53.6 a	7.50 bc

*Means in each column followed by the same letters are not significantly different ($p \leq 0.05$) by Duncan's Multiple Range Test

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Adsorption behaviors of nano-sized TiO_2 in removing of Cd^{2+} from contaminated water

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Abstract
Cadmium is a toxic heavy metal of significant environmental and occupational concern and has been classified as a human carcinogen and teratogen impacting lungs, kidneys, liver and reproductive organs. The World Health Organization (WHO) has set a maximum guideline concentration of 0.003 mg L^{-1} for Cd in drinking water. Given pervasive cadmium contamination and the low drinking water guideline, there is considerable interest in the development of techniques to remove cadmium from contaminated water. In this research, nano-sized TiO_2 was used to investigate the removal of Cd^{2+} from aqueous solution in the various initial concentration of Cd^{2+} (0-80 mg L^{-1}) in three value of pH (6, 7, 8) which ionic strength fixed in 0.01 M by adding of $Ca(NO_3)_2$. Results showed that the Cd^{2+} could be efficiently removed by nano-TiO_2 in the pH range of 6-8 (natural water pH) from aqueous solutions and adsorption of Cd^{2+} on the surface of the adsorbent was increased with increasing solution pH. The experimental data were fitted to the Langmuir, Temkin, and Freundlich models; as Langmuir and Freundlich models better fitted (R^2=0.92-0.98) than Temkin model (R^2=0.74). Finally, it is interesting to notice that TiO_2 nanoparticles could be used to adsorb Cd^{2+} from contaminated/waste water in order to achieve environmental cleanliness.
Keywords: Cadmium, contaminated water, waste water, nanopaticles, titanium oxide

Introduction

Cadmium (Cd) is one of the heavy metals with a greatest potential hazard to humans and the environment. Cadmium poses a serious threat to human health as it accumulates on the environment throughout the food chain. Besides, the industrial uses of cadmium are widespread and increasing in electroplating, paint pigments, plastics, alloy preparation, mining and silver–cadmium batteries (Hutton, 1983). This heavy metal has resulted in serious contamination of both soil and water. Cadmium has been classified as a human carcinogen and teratogen impacting lungs, kidneys, liver and reproductive organs (Waalkes, 2000). The World Health Organization (WHO) has set a maximum guideline concentration of 0.003 mg L^{-1} for Cd in drinking water (WHO, 2008). Given pervasive cadmium contamination and the low drinking water guideline, there is considerable interest in the development of techniques to remove cadmium from contaminated water.

Physicochemical methods such as precipitation, ion exchange and ultra-filtration have been mainly used for the removal of soluble cadmium; however, they are costly (Kashiwa, 2000). Adsorption has been found to be superior to other techniques for water reuse in terms of initial cost, simplicity of design, easy of operation and insensitivity to toxic substances (Mohan and Singh 2002). A variety of adsorbents, including clays, zeolites, dried plant parts, agricultural waste biomass, biopolymers, metal oxides, microorganisms, sewage sludge, fly ash and activated carbon have been used for cadmium removal (Visa etal, 2010).

Research is underway to use advance nanotechnology in water purification for safe drinking. Nanotechnology, the deliberate manipulation of matter at size scales of less than 100 nm, holds the promise of creating new materials and devices which take advantage of unique phenomena realized at those length scales, because of their high reactivity due to the large surface to volume ratio (Wang et al, 2010). Nanoparticles are expected to play a crucial role in water purification (Singh et al, 1998). Nano particles have two key properties that make them particularly attractive as sorbent. On a mass basis, they have much larger surface area than bulk particles and they can also functionalize with various chemical groups to increase their affinity towards target compounds (Dhermendra, 2008).

Titanium dioxide, especially as nanoparticulate anatase, is also an interesting antibacterial, with notable photocatalytic behavior. In addition to having high specific surface areas, nano-TiO_2 also has unique adsorption properties due to different distributions of reactive surface sites and disordered surface regions. The effect of pH on the adsorption of cadmium to titanium dioxide nanoparticles is a matter laboratory-scale experiments. The objective of this research is to investigate the efficiency of nano-sized TiO_2 in removing of cadmium ions from aqueous solution and adsorption isotherm models of cadmium removal at varying pH.

Materials and Methods:
A batch experiment was carried out by contacting a constant absorbent, 0.1 g of TiO_2 with a range of different concentration of the cadmium ion solutions (0-80 mg/L), in 3 ranges of pH (6, 7 and 8). Then, the cadmium ion solutions were agitated in a series flasks with equal volumes of solution (5 ml) for a period of 24 h at 25 ^{0}C. After shaking, the samples were centrifuged at 10,000 rpm for 30min. The final concentration of the cadmium in solution was determined using atomic absorption spectroscopy (AA-6300 Shimadzu atomic absorption spectrometry) and the amount of cadmium adsorbed on the anatase was calculated as follows:

$$q = (C_o - C_e) * V/m$$

Where, C_0 and C_e are the concentrations of the initial and the final metal ions (mg/L), respectively; V is the volume of the solution (L); and m is the anatase weight (g) in dry form. The amount of removing efficiency (%RE) was determined as follows:

$$\%RE = ((C_0 - C_e)/C_0)*100$$

Where, C_0 and C_e are the concentrations of the initial and the final metal ions (mg/L), respectively.

The isotherm data on Cd adsorption were fitted to three two-parameter equations (Freundlich, Langmuir and Temkin). The Langmuir model (L type, based on monolayer adsorption of solute) and the Freundlich model which are the most widely accepted and used in literatures to describe the relationship between equilibrium metal uptake (q_e) and final concentrations (C_e) at equilibrium.
The Langmuir equation can be represented as follows:

$$q_e = (K_L C_e q_m)/(1+K_L C_e)$$

Where K_L (L/mg) is the equilibrium adsorption constant which is related to the affinity of the binding sites and q_m (mg/g) is the maximum amount of metal ion sorbed per unit mass of sorbent when all binding sites are occupied.
The Freundlich equation is given:

$$q_e = K_F C_e^{1/n}$$

Where K_F and n are the Freundlich constants and are related to the adsorption capacity of the sorbent and the adsorption intensity, respectively.

The Temkin isotherm model is given by the equation:

$$q_T = A + K_T \mathrm{Ln} C_e$$

Where K_T is the Temkin constant related to the heat of sorption (J mol^{-1}) and A is the Temkin isotherm constant (L. g^{-1}) (Elebi, 2007).

In this study, the isotherm data were fitted to the 3 models by non-linear regression using the method of least squares by SOLVER program and statistical analyses were preformed with SAS.

Results and discussion
Effect of pH and initial cadmium concentration
The effect of pH on the Cd adsorption on theTiO_2 nano particles between pH 6 and 8 presented in Fig. 1. It can be found that the Cd adsorption tends to increase significantly with the increase of pH from 6 to 8. By increasing pH from 6 to 8, average values of Cd adsorbed by nano-TiO_2 increased from 538 to 780 mg/g respectively. Also the results showed that removing efficiency of Cd by nano TiO_2 increased significantly from %67 at pH 6 to %83 at pH 8. A similar trend was also observed for Cd adsorption on boehmite (AlOOH) (Guanghui et al, 2008). The pH of the aqueous solution is an important variable that influences the adsorption of anions and cations at the solid–liquid interfaces (Waranusantigul et al, 2003). The pH_{ZPC} of nano-TiO_2 is 6.8, so increasing pH causes the TiO_2 surface to carry more negative charges and thus would more significantly adsorb the positively charged species in solution. Therefore, the higher adsorption of Cd at pH 8 resulted from

a electrostatic sorption between the more positively charged Cd^{2+} species and negatively charged surface sites, When pH is less than pH_{PZC} results in creation of the positive surface charge thus would more significantly repulse the positively charged species in solution. Therefore, the lower adsorption of Cd at pH 6 resulted from an increased repulsion between the more positively charged Cd^{+2} species and positively charged surface sites (Suttiponparnit et al, 2011). The effect of pH on Cd adsorption by nano TiO_2 can be modeled as follows:

$$Ti\text{-}OH_2^+ \overset{(2)}{\longleftrightarrow} Ti\text{-}OH \overset{(1)}{\longleftrightarrow} Ti\text{-}O^- + H^+$$

Sorption of the Cd^{2+} increased with increasing the initial metal concentration tending to saturation at higher metal concentrations (Fig. 1). As initial Cd^{2+} concentration increased from 0 to 80 mg/L, the adsorption capacity of nano-TiO_2 increased significantly from 0 to 1566, 1925, 2391 mg/g at pH 6, 7 and 8, respectively. A higher initial concentration provides an important driving force to overcome all mass transfer resistances of the pollutant between the aqueous and solid phases, thus increases the uptake (Aksu and Tezer, 2005). A similar trend was also observed for Ni adsorption on nano-crystalline calcium hydroxyapatite (Mobasherpour et al, 2010).

The removing efficiency (RE) of Cd^{2+} by nano TiO_2 decreased by increasing of cadmium initial concentration due to the inhibition of available active sites on the surface of TiO_2 nano material. By increasing Cd^{2+} initial concentration from 2.5 to 80 mg/L, removing efficiency decreased significantly from % 92 to % 39 at pH 6, % 94 to % 48 at pH 7 and % 95 to % 71 at pH 8. These results were an agreement with reports by other authors (Mataka, 2010 & ZHU Zhi-liang, 2007).

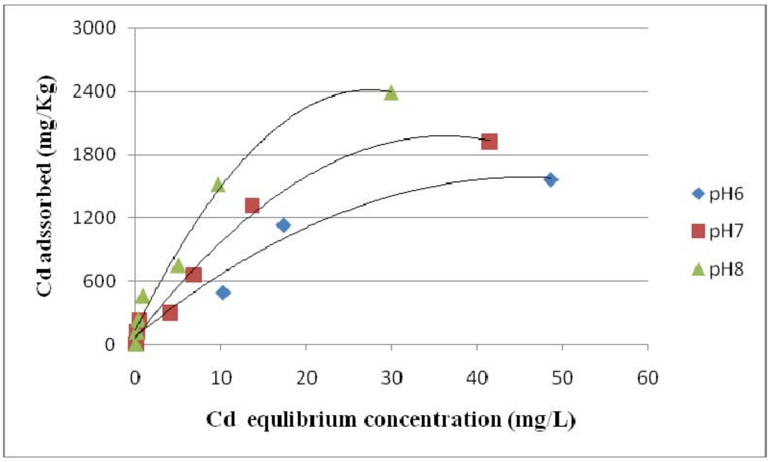

Fig. 1: Effect of pH on Cd(II) adsorption at 25^{0C} on TiO_2 at 3 pH

Adsorption isotherms modeling

Freundlich, Langmuir, and Temkin isotherms were tested for their ability to correlate with the experimental results by comparing theoretical plots of each isotherm with the experimental data for the sorption of Cd^{2+} ions by nano-TiO_2 particles at different pH in Fig. 2. The quality of the fit assessed using the correlation coefficient (R^2). Both Langmuir and Freundlich adsorption models showed a better fit than Temkin model. Table 1 shows isotherm parameters obtained by using non-linear method.

Table 1: Estimated isotherm parameters for Cd adsorption on TiO_2 nano particles

pH	Langmuir			Freundlich			Temkin		
	q_m (mg/Kg)	$K_{L(L/mg)}$	R^2	n	K_f	R^2	K_T	A	R^2
6	2492 c	0.03 b	0.95	0.56 a	183 c	0.95	210 ab	461 c	0.84
7	2981 b	0.04 b	0.97	0.58 a	231 b	0.95	233 a	559 b	0.77
8	3765 a	0.06 a	0.94	0.66 a	320 a	0.98	236 a	643 a	0.86

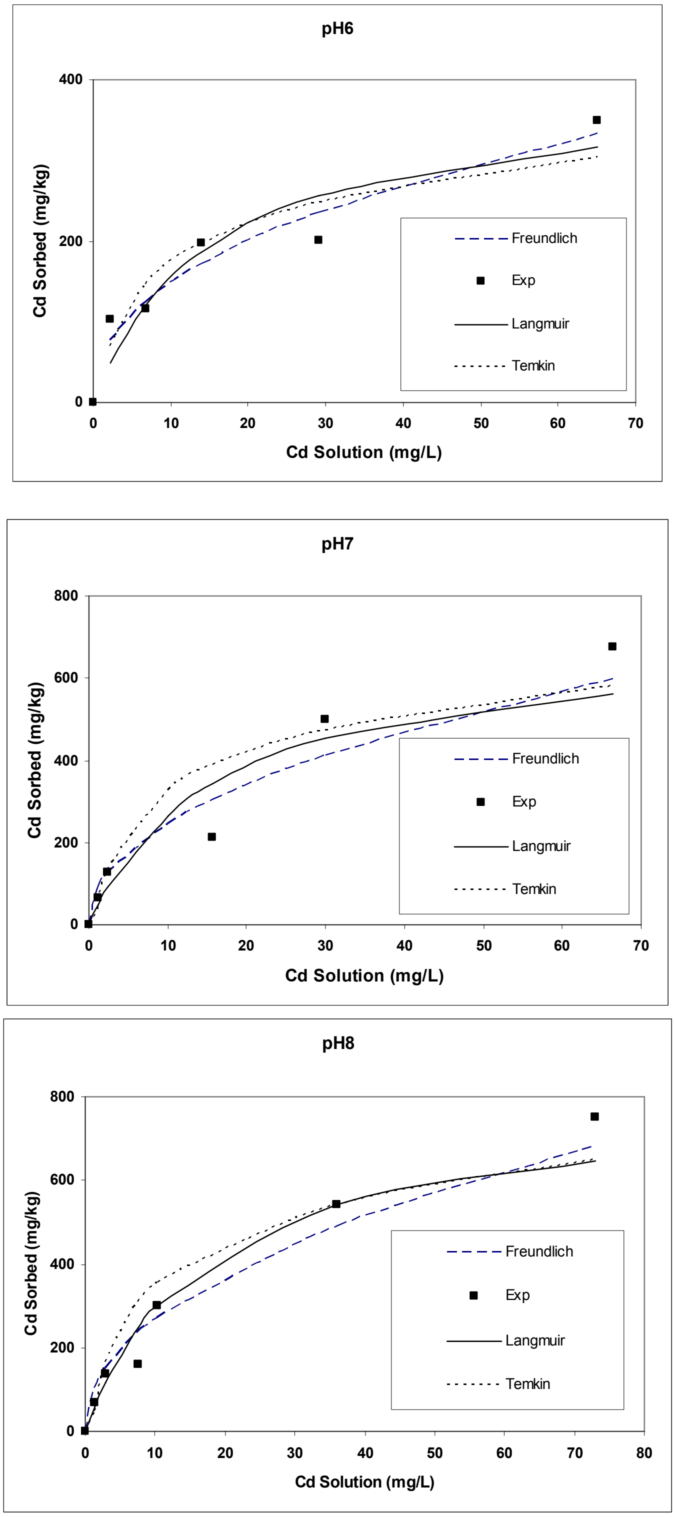

Fig 2. Comparison of experimental and estimated data by different isotherms at 3 ranges of pH

It is demonstrated that the nano TiO_2 are an effective adsorbent for Cd^{2+} removal from contaminated water and adsorption process is pH dependant. By increasing pH of solution removing efficiency significantly increase but it was decreased with increasing initial concentration of cadmium.

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Sorption of copper (II) by nano-sized γ-Al_2O_3

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Abstract

Copper (Cu) is important metal having many applications in industries and its pollution cause a risk to the environment as well as threaten human health through the food chain. In order to eliminate/reduce Cu^{2+} from aqueous solutions, nano-sized aluminum oxide (20 nm) was investigated for adsorption of Cu^{2+} in various pH. Each flask contained 0.05 g of adsorbent and 5 ml of Cu^{2+} solution series (0 - 80 mg L^{-1}), and ionic strength was fixed with 0.01 M $CaCl_2$. Results showed that nano-alumina have high efficiency in removing of Cu^{2+} from aqueous solution and removal efficiency (RE) of copper increased with increasing pH and initial concentration of Cu^{2+}. Experimental data were analyzed with Langmuir and Freundlich models, both models are better fitted at three level of pH (R^2 > 0.95- 0.99). These finding indicate that nano-sized aluminum oxides are effective materials for Cu^{2+} removal and can be applied in the removal of Cu^{2+} from contaminated/waste water.

Keywords: Copper, contaminated water, waste water, nano particles, metal oxide

Introduction

Copper is an important metal having many applications in industries such as electroplating, transportation equipment, construction, military industry, and electrical industrial (Bailey et al. 1999). High levels of Cu ions in the environment are harmful for many life forms. Industrial and mining wastewaters are important sources of pollution of heavy metals (Quek et al., 1998). The pollution of heavy metals may cause a risk to the environment as well as threaten human health through the food chain (Chamarthy et al. 2001). Drinking water that contains higher than normal levels of Cu may cause vomiting, diarrhea, stomach cramp and nausea. As an effort to reduce the heavy metal levels in waste water, drinking water and water used for agriculture to the maximum permissible concentration. The suggested safe level of Cu in drinking water for humans varies depending on the sources, but tends to be pegged at 1.5 to 2.0 mg/L. Therefore, it is very important to determine an effective way to remediate heavy metal contaminations.

Numerous treatment technologies to eliminate or reduce environmental problems have been developed, such as biodegradation, adsorption, and photocatalysis (Huang et al., 2002). The development of nanotechnology at the end of the 20th century has widened the variety of adsorbents. The most common absorbents are porous materials, such as activated carbon, quartzsand, activated alumina, and zeolite. The main advantages of using nanoparticles as adsorbents are: (a) they can be easily synthesized at a lower cost, (b) the amount of nanoparticles used is much less, and (c) their adsorption capacity is large due to their large surface area. Aluminum oxides can effectively adsorb heavy metals (Matagi et al., 1998; Scholz, 2003; Murray et al., 2005). Iron oxides, such as goethite (a-FeOOH) and hematite (a-Fe2O3), can effectively adsorb heavy metals (Swift, 1991). There have been a number of studies about the goethite or hematite adsorption systems, and these studies are related to the adsorption modeling of the surface reactions (Weerasooriya, 2004). Ionic strength and pH influence cation adsorption due to the variation of the electric charge in soils rich, in iron (Fe) and aluminum (Al) oxides and hydroxides (Casagrande, 2007).

In this work, the potentials for the use of, nano-sized aluminum oxide as adsorbents for copper ion removal from solution was investigated and the resulting isotherm compared using the error non-linear method of two widely used isotherms, Langmuir and Freundlich, were examined. Several factors influence the adsorbability of ions on aluminum oxide, including salt concentration, pH of the solution, degree of loading on the adsorbent, presence of ligands and complex formation (Kara, 2007). Moreover, its use over comes some experimental difficulties associated with experiments using natural suspended particles, which may represent an assemblage of components with different properties, including different surface groups.

2. Materials and methods

2.1. Instrumentations

Three equipments were used in this research:
1. An atomic absorption spectrometer (AAS) with copper hallow cathode lamp and air acetylene flame was used to determine the copper concentrations.
2. A pH meter was used for pH measurement. The meter was standardizedusing buffer solutions with pH values (6, 7and 8).
3. A mechanical shaker was used for agitating the samples.

2.2. Adsorption batch experiments

In order to eliminate/reduce Cu^{2+} from aqueous solutions, nano-sized aluminum oxide (20 nm) was investigated for adsorption of Cu^{2+} in various pH. Copper sulfate salts, was used to make a standard solution for adsorption experiments. Each flask contained 0.05 g of adsorbent and 5 ml of Cu^{2+} solution series (0 - 80 mg L^{-1}), and ionic strength was fixed with 0.01 M $CaCl_2$. The amount of uptake was calculated by the difference between the initial and the final concentrations in solution.

2.3. Adsorption isotherm

Numerous isotherm equations have been reported, and two major isotherms, the Langmuir and Freundlich isotherm, are tested to fit the experimental data.
The Langmuir adsorption isotherm can be expressed as:

$$q = bK_LC_e/1+C_e$$

Where b is the adsorption capacity, K_L is the Langmuir constant and C_e equilibrium concentration.
The Freundlich isotherm is in the form of:

$$q_e = K_f\, C_e^{1/n}$$

Where K_f is the extent of the adsorption and n is the degree of nonlinearity between Cu^{2+} concentration, and adsorption.

3. Results and Discussion

3.1. Effect of initial concentration

Figure 1 shows that adsorption capacity increasing from 0 to 3200 mg/kg as the metal concentration increases from 0 to 80 mg/L. The trend is that of the result of the progressive increase in the electrostatic interaction between the copper ions and the absorbent active sites. Moreover, this can be explained by the fact that more adsorption sites were being covered as the metal ions concentration increases (Bencheikh et al., 2005). Besides, higher initial concentrations lead to an increase in the affinity of the copper ions towards the active sites (Duvnjak et al., 2003). The decline in the adsorption capacity is due to the availability of smaller number of surface sites on the adsorbent for a relatively larger number of adsorbing species at higher concentrations (Ahmed et al., 2000). Similar results were observed by (Han et al., 2005) for the adsorption of copper (II) and lead (II) on chaff.

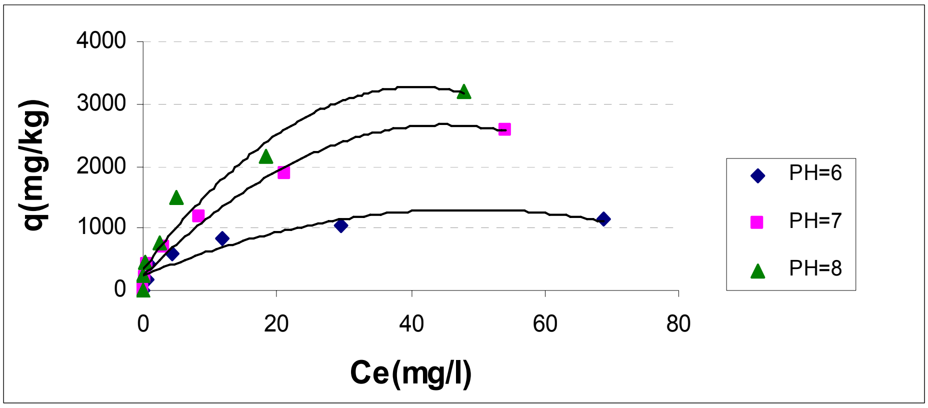

Fig.1. Effect of Initial Concentration Cu (mg/L) on Copper Adsorption

The shape of adsorption is good agreement with several previous studies of copper adsorption onto iron oxyhydroxides (Balistrieri and Murray, 1982; Ali and Dzombak, 1996; Christl and Kretzschmar, 2001).

3.2. Effect of initial pH

The acidity of solution pH is one of the most important parameters controlling the uptake of heavy metals from wastewater and aqueous solutions by metal oxides (King et al., 2006). Our results show that the Cu adsorption by nano Al_2O_3 is very pH dependant, as pH increasing from 6 to 8 the adsorption of Cu significantly increased (Fig 2)

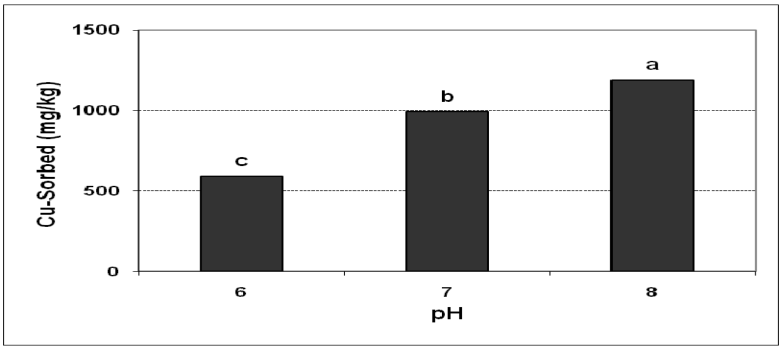

Fig.2.Effect of pH on copper adsorption capacity, q_e (mg/kg) by 0.05g adsorbent aluminum oxide

The minimum adsorption observed at low pH 6 (1133 mg/kg), It would be plausible to suggest that at lower pH value, the surface of the adsorbent is surrounded by hydronium ions (H+), thereby preventing the metal ions from approaching the binding sites of the sorbent (Acar et al., 2006). This means that at higher H^+ concentration, the aluminum oxide surface becomes more positively charged such that the attraction between aluminum oxide and metal cations reduced (Waheed Akhtar et al., 2004). In contrast, as the pH increases, more negatively charged surface becomes available thus facilitating greater copper removal. It is commonly agreed that the sorption of metal cations increases with increasing pH as the metal ionic species become less stable in the solution (Gezici et al., 2007).

3.3. Adsorption isotherm

Adsorption isotherms are fundamental and play an important role in the determination of the behavior and maximum capacity of adsorption. It also provides a panorama of the course taken by the system under study in a concise form, indicating how efficiently an aluminum oxide will adsorb

and allows an estimate of the economic viability of the aluminum oxide commercial applications for the specified solute.
In order to adapt for the considered system, an adequate model that can reproduce the experimental results obtained, equations of Langmuir, Freundlich, have been considered. The experimental data on the effect of an initial concentration of metal on the aluminum oxide of the test medium were fitted to the isotherm models and graphical representations of these models are presented in figures (3, 4 and 5).

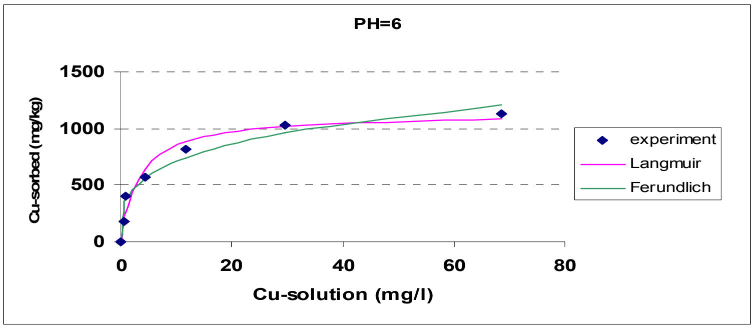

Fig. 3: Copper ions adsorption isotherms on nano-sized γ-Al_2O_3 in pH 6

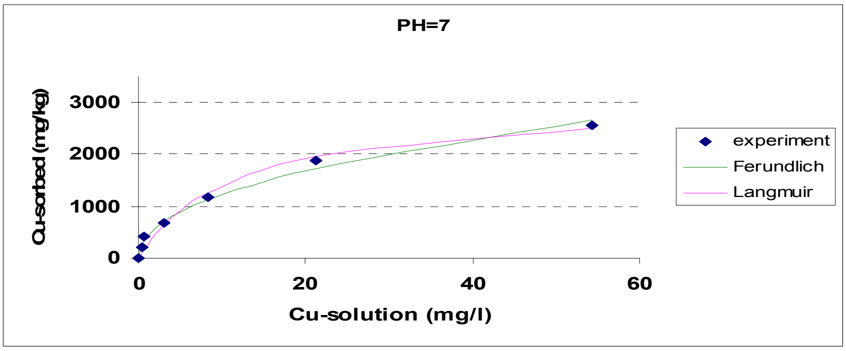

Fig. 4: Copper ions adsorption isotherms on nano-sized γ-Al_2O_3 in pH 7

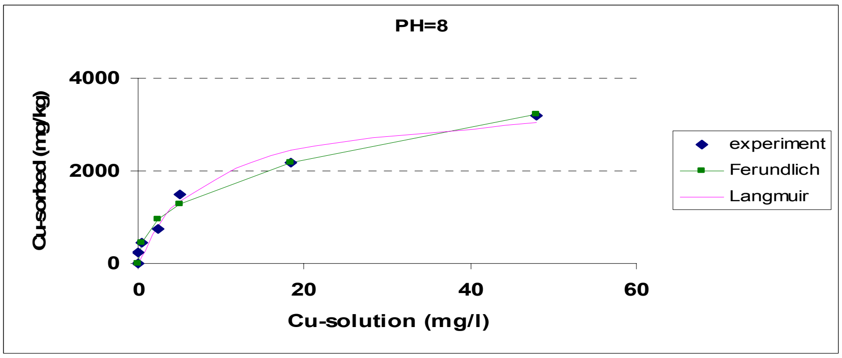

Fig. 5: Copper ions adsorption isotherms on nano-sized γ-Al_2O_3 in pH 8

Table1. The value of parameters for each isotherm model used in the studies

PH	Isotherm Model	Parameter	R^2
6	Langmuir	K_L=0.3 b=1132 (mg/kg)	0.97
	Freundlich	K_f=374 n=0.27	0.95
7	Langmuir	K_L=0.082 b= 3070 (mg/kg)	0.99
	Freundlich	K_f=415 n=0.46	0.99
8	Langmuir	K_L=0.11 b=3607 (mg/kg)	0.97
	Freundlich	K_f=660 n=0.40	0.98

It is observed that the both Langmuir and Ferundlich models better fitted with the results of experiment while the correlation coefficients (R2) of the Freundlich model are higher than those of the Langmuir (Table 1). Moreover, the maximum monolayer adsorption capacity (b) was obtained in pH 8 (3607 mg/kg). It is observed that nano-aluminum have high efficiency in removing of Cu2+ from aqueous solution and removal efficiency (RE) of copper increased with increasing pH and initial concentration of Cu2+. It suggests that adsorption functions of nano-aluminum oxide give great potential (RE) for applications in heavy metal removal from contaminated water.

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Efficiency of nano-sized γ-Al_2O_3 in removal of phosphorus from contaminated water

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Abstract
Phosphorus (P) is an essential nutrient for growth of living organisms and it is a limiting factor for plants and animals that live in water ecosystems. Increasing of P in the aquatic environment cause to eutrophication and reduce water quality. P polluted soil and water through different ways, including agriculture, human industries, mining activities, the application excessive phosphorus fertilizers. Surfaces of metal oxides are important PO_4 adsorption sites due to the presence of a multiply charged cation, high positive surface charge densities at near-neutral pH, and the propensity to hydroxylate in aqueous systems. To evaluate the efficiency of nano-sized aluminum oxide in P removal, a laboratory batch experiments carried out with different initial concentration of P (0-60 mg L^{-1}) and three pH (4, 7, and 10) with 0.1 g of nano γ-Al_2O_3. Final P concentration determined after shaking 24 h in centrifuge extraction and phosphorus adsorption was calculated from initial and final concentration differences. The results showed high efficiency and capacity of nano aluminum oxide in P removal from aqueous solution. Removal efficiency (RE) decreased with increasing pH and initial P concentration, as RE was 100% in initial P concentration below 10 mg L^{-1} at pH 4 while reduced to %74 in 60 mg P L^{-1} at pH 10. Experimental data were fitted to Langmuir and Freundlich adsorption models, and Freundlich (R^2=0.97-0.99) was a little better fitted than Langmuir for P sorption by nano γ-Al_2O_3 at three pH (R^2=0.94-0.97).
Keywords: phosphorus, nanoparticles, aluminum oxide, eutrophication

Introduction

Phosphorus (P) is an essential nutrient for the growth of organism most ecosystems, but superfluous phosphorus can also cause eutrophication and hence deteriorate water quality. Phosphorus is released into aquatic environments in many ways, of which the most significant are human industrial, agricultural, and mining activities. Although phosphorus removal is required before discharging wastewater into bodies of water, phosphorus pollution is nevertheless increasing. Therefore, there is currently an urgent demand for improved phosphorus removal methods which can be applied before wastewater discharge (Yan, 2010).
Phosphorus removal from wastewater has been widely investigated and several techniques have been developed including adsorption methods, physical processes (settling, filtration), chemical precipitation (with aluminum, iron and calcium salts), and biological processes that rely on biomass growth (bacteria, algae, plants) or intracellular bacterial polyphosphates accumulation (Bashana, 2004). Recently, the removal of phosphate from aqueous solutions via adsorption has attracted much attention. The key problem for many phosphorus adsorption methods, however, is finding an efficient adsorbent. Several low-cost or easily available clays, waste materials and by-products such as zeolite, palygorskite, fly ash, blast furnace slag, hydroxides sludge, oxide tailings, activated aluminum oxide and ferric hydroxide, have all been studied extensively and systematically (Yan, 2010). Al_2O_3 widely used for cation removal is also known to have affinity towards phosphate anions.
In present study we intend to evaluate sorption behavior of phosphate anions on nano γ-Al_2O_3 and the effect of pH of solution and initial phosphorus concentration on P adsorption and removal efficiency of nano γ-Al_2O_3 for phosphorus sorption.

Materials and methods

A laboratory batch experiments carried out with different initial concentration of P (0-60 mg L^{-1}) and three pH (4, 7, and 10) with 0.1 g of nano γ-Al_2O_3. The suspensions were continually shaken at room temperature (25 $^{\circ C}$) and 180 rpm for 24 h to ensure approximate equilibrium. The suspensions were centrifuged with D7200 Universal in 6000 rpm for 1/2 h. The supernatant was filtered by a 0.42 μm filter paper and then was analyzed for P using a spectrophotometer 2100 UV at length wave 880 nm (Murphy and Riley, 1962)) and the amount of P adsorbed on the nano particles was calculated as follows:

$$q = (C_o - C_e) * V/m$$

Where, C_0 and C_e are the concentrations of the initial and the final metal ions (mg/L), respectively; V is the volume of the solution (L); and m is the anatase weight (g) in dry form. The amount of removing efficiency (%RE) was determined as follows:

$$\%RE= ((C_0 - C_e)/C_0)*100$$

Where, C_0 and C_e are the concentrations of the initial and the final metal ions (mg/L), respectively.
The experimental data were fitted to three two-parameter equations (Langmuir and Freundlich). These models are the most widely used in literatures to describe the relationship between equilibrium P adsorption (q_e) and final concentrations (C_e) at equilibrium. The Langmuir equation [Eq. (1)] is the most important model for mono-layer adsorption. It is based on the assumptions: adsorption can only occur at a fixed number of definite localized sites, each site can hold only one adsorbate molecule, and all sites are equivalent and no interaction between adsorbed molecules:

$$q_e =(K_L C_e q_{max})/ (1+ K_L C_e) \qquad (1)$$

where q_{max} is the maximum quantity of phosphate ions per unit weight of nano γ-Al_2O_3 to form a complete monolayer on the surface (mg/g), C_e is the equilibrium concentration of phosphate (mg/L) and K_L is a constant related to the affinity of binding sites with the phosphate ions (L/mg). It should be noted that q_{max} represents a practical limiting adsorption capacity corresponding to the surface of sorbent fully covered by sorbate ions. This quantity is particularly useful in the assessment of the adsorption performance, especially in cases where the sorbent does not reach its full saturation as it enables the indirect comparison between different sorbents.
The Freundlich model [Eq. (2)] is the most important multi-site sorption isotherm for heterogeneous surfaces. Even though the model originates from empirical expressions, it has also been derived by assuming an exponential decay energy distribution function:

$$q_e =K_F C_e^n \qquad (2)$$

where K_F is an indicator of adsorption capacity (L/mg), C_e is the equilibrium concentration of phosphate (mg/L) and n indicates the effect of concentration on the adsorption capacity and represents the adsorption intensity (dimensionless).

Results and Discussions

Effect of initial concentration and pH on P sorption

The sorption of phosphate anions on nano γ-Al_2O_3 at pH 4, 7 and 10 can be seen from Figure 1, which shows that with increases in concentration of phosphate, sorption on nano γ-Al_2O_3 increases sharply and reaches a maximum similar to reported previously for phosphate sorption on goethite (Peleka and Deliyanni, 2008). Removal efficiency (RE) decreased with increasing pH and initial P concentration, as RE was 100% in initial P concentration below 10 mg L^{-1} at pH 4 while reduced to %74 in 60 mg P L^{-1} at pH 10. It is also interesting to note that pH has a major effect on the sorption of phosphate anions depicting a strong dependency on pH (Fig. 1). As shown in Fig. 1 with increasing pH from 4 to 10, P sorption decreased and sorption curve in pH 10 lied horizontally than others. Similar trend was reported elsewhere (Churms, 1966). It is demonstrated that a greater affinity of aluminum oxide for the phosphate anions at low pH values due to protonation of aluminum oxide surface below the point of zero charge pH (pH_{PZC}) (Sperling et al., 1992). Thus, at low pH the magnitude of the positive charge is greater and there are greater interactions of phosphate anions with the solid surface (Hingston et al., 1974; Sperling et al., 1992). In contrast to high pH i.e., close to PZC the surface charge on nano γ-Al_2O_3 is minimum; therefore, the extent of phosphate sorption is low and with increasing pH to above ZPC, negative charges increased on the surface due to deprotonation; therefore, PO_4^{3-} sorption decreased.

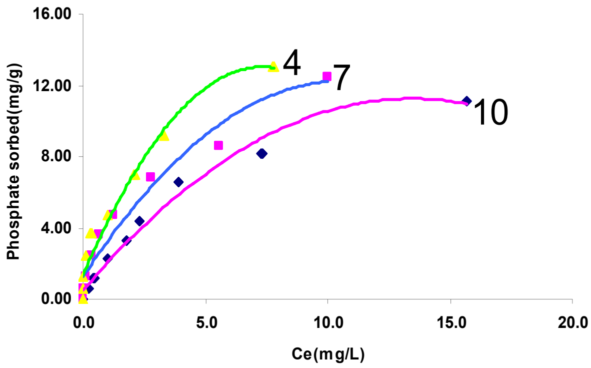

Fig. 1: Effect of initial concentration on sorption of phosphate anions at pH 4, 7, 10 on nano γ-Al_2O_3

Sorption isotherm modeling

Analysis of equilibrium data is important for developing an equation that can be used for design purposes. Classical adsorption models, such as the Langmuir and Freundlich, have been extensively used to describe the equilibrium established between adsorbed ions on the nano metal oxides and ions remaining in solution (C_e) at a constant temperature. Experimental data were fitted to Langmuir and Freundlich adsorption models, and Freundlich (R^2=0.97-0.99) is better fitted than Langmuir for P sorption by nano γ-Al_2O_3 at three pH (R^2=0.94-0.97). The Langmuir and Freundlich adsorption coefficients are presented in Table 1 according to non-linear fitting by SOLVER program (Fig. 2).

Table 1: The coefficients of Freundlich and Lagmuir models for phosphates sorption onto nano γ-Al_2O_3 at three pH

pH	Langmuir constants			Freundlich constants		
	$q_{max(mg/g)}$	$K_{L(L/mg)}$	R^2	$K_{F(L/mg)}$	n	R^2
4	16.2	0.47	0.94	5.61	0.42	0.98
7	14.9	0.36	0.95	4.23	0.47	0.99
10	14.7	0.19	0.97	2.73	0.53	0.97

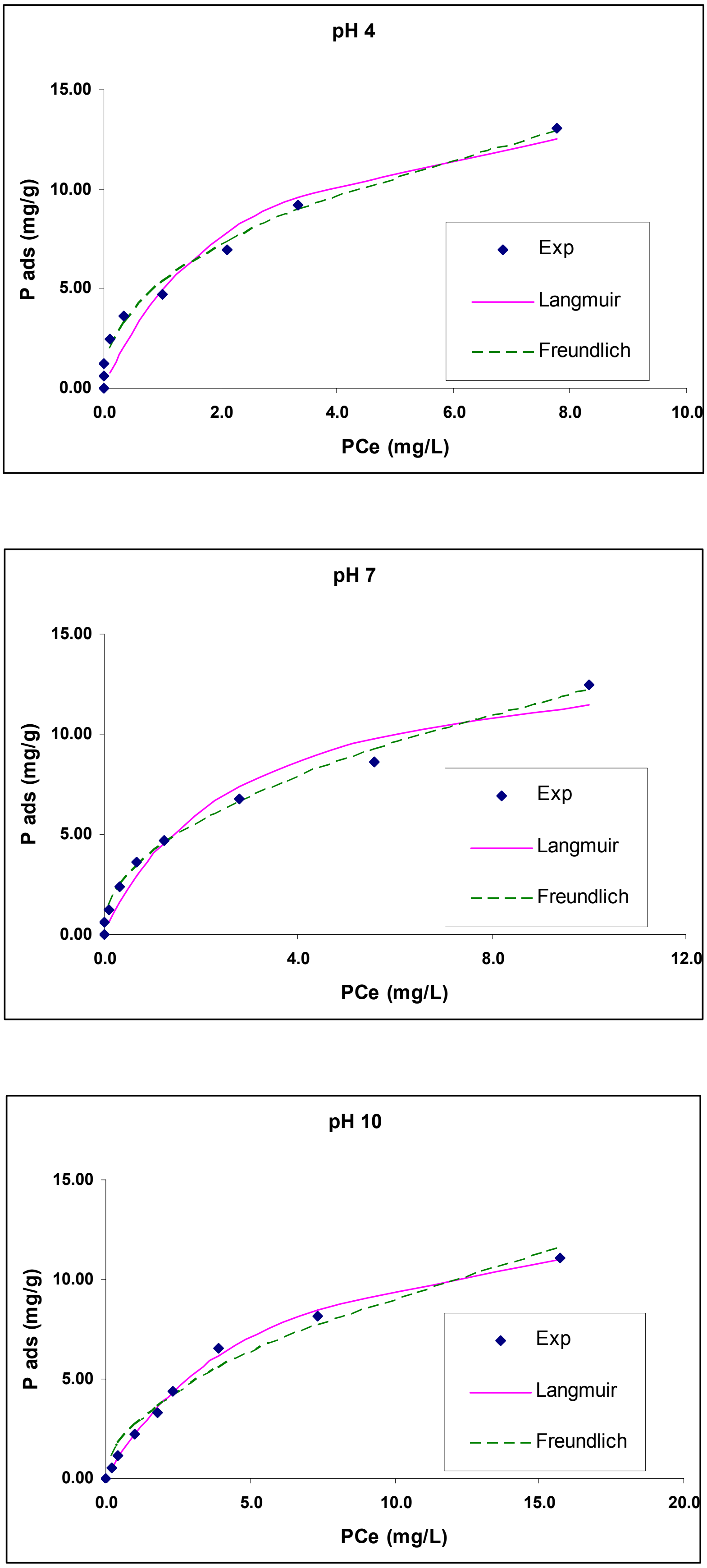

Fig 2. Comparison of experimental and estimated data by different isotherms at 3 ranges of pH

According to the regression coefficients in Table 1, both models were good fitted and could adequately predict the experimental data as shown in Fig. 2. Regarding Langmuir model the monolayer sorption capacity, q_{max}, was found to decrease from 16.2 to 14.7 mg P g^{-1} as the pH increased from 4 to 10, indicating that the sorption of phosphates decreased with increasing pH. The q_{max} value is the maximum value of q_e, which is important to identify which sorbent show the highest uptake capacity. In addition, the values of constant K_L correspond to the concentration at which phosphate ions in the amount of q_{max} are bound and indicate the affinity for the binding of phosphate ions, which deceased by increasing pH due to deprotonation of nano γ-Al_2O_3 surface. Similar trend observed for K_F values (adsorption capacity) in Freundlich model.

Conclusions

It is concluded that nano-sized aluminum oxide had high capacity and efficiency for phosphate ions sorption and its removal from aqueous solution. Adsorption process is pH dependant and removal efficiency (RE) decreased with increasing pH and initial P concentration, as RE was 100% in initial P concentration below 10 mg L^{-1} at pH 4 while reduced to %74 in 60 mg P L^{-1} at pH 10. Both Langmuir and Freundlich adsorption models were good fitted and could adequately predict the experimental data.

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Effect of initial concentration and pH on adsorption of Cd^{2+} by nano- Al_2O_3

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Abstract:

The mobilization of Cd^{2+} in the water and wastewater by industrial activities and use of phosphorus fertilizers is of serious concern due to the toxicity of this metal in humans. In this study, the equilibrium adsorption isotherms of Cd (II) ions from aqueous solution studied by using nano-sized Al_2O_3 with surface area of 200 m^2/g . The adsorption of Cd^{2+} by Al_2O_3 nanoparticles was measured as a function of pH (6, 7, 8) and initial concentration at 298 ^{0}K, and ionic strength of background electrolyte fixed in 0.01 by $Ca(NO_3)_2$. Adsorption isotherms were obtained using concentration of the Cd^{2+} ions ranging (0-80) mg L^{-1}. The results showed that adsorption by Al oxide increased steadily as the pH was increased due to the effect of pH on the amount of surface charge of nanoparticles. Also, the amount of Cd^{2+} adsorbed increased as the initial concentration increased, but removal efficiency of nanoparticles decreased by increasing the initial concentration of Cd^{2+} where the maximum adsorption efficiency (%56) reached on 2.5 mg L^{-1}. The sorption of Cd^{2+} behaviour by nano Al_2O_3 was fitted to various models. The adsorption data showed good fit to both Langmuir and Freundlich models (R^2= 0.93-0.98). Due to high capacity obtained for Cd^{2+}, nano Al_2O_3 can be regarded as a potential adsorbent for treatment of waste water.

Keywords: Cadmium, contaminated water, waste water, nano paticles, metal oxide (Al_2O_3)

Introduction

Today most of the countries are facing drinking water problems and conditions are very severe especially in developing countries. It has realized that heavy metal ions, such as cadmium, mercury and lead, contained in waste water from various sources of industries are detrimental to human health, due to their extended persistence in biological systems and tendency to bio- accumulate as they move up the food chain. Cadmium, as one of the principle toxic heavy metals, is strictly controlled in drinking according to the water quality criterion. The maximum of the acceptable concentration of cadmium should be equal to or less than 0.005 $mg \cdot L^{-1}$ (Liu Guanghui, 2008).

The removal of toxic contaminants from industrial wastewaters is one of the most important environmental issues.(Choi, 2006). A number of technologies have been used to remove heavy metals from wastewater streams such as precipitation, evaporation and membrane processes (Choi, 2006). However, these technologies are most suitable in situations when the concentrations of heavy metal ions are high. They may be either ineffective or expensive when heavy metals are present in the wastewater at low concentration, or when very low concentration of heavy metals in the treated water are required (Yu, 1999). Among various techniques, the adsorption process is used exclusively in water treatment and many studies have been carried out to find inexpensive and chemico - physically feasible adsorbent (Choi, 2006). With the advent of improved technology in the recent years, more emphasis has been placed in the field towards the invention of micro and nanotechnology to remove of toxic metals pollutants from wastewater (Balaji, 2009). The high surface area to mass ratios nano particles can greatly enhance the adsorption capacities of sorbent materials (Dhermendra, 2008).

Fe/ Al oxides such as goethite, hematite, and gibbsite are important compounds of acid soils in tropical and subtropical regions (Padomaabham, 1983). It was noted that Cu (II) could be adsorbed specifically by goethite and gibbsite (Padomaabham, 1983). Laboratory experiments have demonstrated that Al_2O_3 nano particles are strong sorbents for heavy metals. The removal of heavy metals by nano particles has shown promising results with nano- crystalline titanium dioxide (Pena, 2005), nano scale zero valent iron (Hardiljeet, 2011), and so on. Recently it has been discovered that it is possible to remove As species with magnetite (Fe_3O_4) nano particles (Yean, 2005), and it is very effective in removal of Cr (vI) (Wang, 2009).

The objective of this study is to determine the adsorption behaviors of Cd on nano sized Al_2O_3 and the effects of pH and initial concentration of Cd^{+2} on Cd sorption by Nano-sized Al_2O_3.

Materials and Metods:

Al_2O_3 nano particles were used as a adsorbent to remove heavy metal Cd^{+2} in this study. The contents of Cd^{2+} ions remaining in the solution after ion-exchange experiment were measured by atomic absorption spectrometry (AA- 6300 Shimadzu atomic absorption spectrometry). BET surface areas are 200 m^2/g for Al_2O_3 nano particles. $Cd(NO_3)_2$ (Merck Co) were used to prepare various concentration of stock solution. Sorption experiments were carried out at 3 pH (6, 7, 8) and the initial total metal ion concentration range was (0, 2.5, 5,10, 20, 40, 80) mg/L with the $Ca(NO_3)_2$ solution with ionic strength of 0.01 as a back ground and sorbent concentration was 0.1g/L into 5 ml of glass flask. HCl 0.1 M and NaOH 0.1 M were used to adjusting the pH. Then the flasks were shaked for 24 hours at 298 K until the equilibrium reached at given temperature and initial concentration, Then the samples were centrifuged at 10,000 rpm for 15min and then filtered and the filtrate was analyzed to determine the contents of Cd ion remaining in the filtrate by atomic absorption spectroscopy (AA- 6300 Shimadzu atomic absorption spectrometry). Sorption isotherms have been fitted with Temkin, Langmuir and Freundlich equation.

The Langmuir equation was chosen for the estimation of maximum adsorption capacity corresponding to complete monolayer coverage on the adsorbent surface. It is well known that the Langmuir equation is intended for a homogeneous surface. A good fit of this equation reflects monolayer adsorption. Once a Cd molecule occupies a site, no further adsorption will take place at the site. The Langmuir equation can be represented as follows:

$$q_e = (K_L C_e q_m)/(1+K_L C_e)$$

where Ce (mg. L^{-1}) and qe (mg. g^{-1}) are the liquid phase concentration and solid phase concentration of adsorbate at equilibrium, respectively; q_m (mg. g^{-1}) and K_L (L. mg^{-1}) are the Langmuir isotherm constants; q_m is the maximum adsorption capacity and K_L related to the binding energy of the metal ion to the active site.

The Freundlich equilibrium isotherm equation was also used to describe perimental adsorption data. This isotherm is an empirical equation which is used for the description of multilayer adsorption with interaction between adsorbed molecules. The Freundlich isotherm is the earliest known relationship describing the adsorption equation. Model predicts that the Cd concentrations on the material will increase as long as there is an increase of the Cd concentration in the solution. Usually it applies to adsorption onto heterogeneous surfaces with a uniform energy distribution and reversible adsorption. The application of the Freundlich equation suggests that adsorption energy exponentially decreases on completion of the adsorbtional centers of an adsorbent. The equation is given as:

$$q_e = K_F C_e^{1/n}$$

where q_e is concentration of Cd adsorbed (mg. g^{-1}), C_e (mg. L^{-1}) is the equilibrium Cd concentration in the aqueous solution, and K_F and n are constants that can be related to the adsorption capacity (mg/l) and the adsorption intensity, respectively.

The Temkin isotherm model assumes that the adsorption energy decreases linearly with the surface coverage due to adsorbent–adsorbate interactions. The Temkin isotherm model is given by the equation:

$$q_T = A + K_T \ln C_e$$

where K_T is the Temkin constant related to the heat of sorption (J mol^{-1}) and A is the Temkin isotherm constant (L. g^{-1}) .(Elebi, 2007)

The amount of adsorbed metal was calculated using the equation:

$$q = (C_o - C_{eq}) / C_s$$

where C_0 and C_{eq} [mg.l^{-1}] are the concentrations of the metal ion in initial and final solutions and C_s [g.l^{-1}] is the amount of sorbent.

Results and discussion:

Effect of pH on Cd adsorption

In order to investigate the effects of pH on Cd^{+2} adsorption on Al_2O_3, a batch experiment was carried out in 3 range of pH (6, 7, and 8). The amount of uptake on various pH was plotted and showed in Fig. 1. The results showed that pH has a significant effect on the adsorption of Cd^{+2} on alumina particles, as adsorption by Al oxide increased steadily as the pH was increased. At lower pH, the reaction solution would be positively charged, thus making H^+ ions compete effectively with positive Cd^{+2} ions, which may result in a decrease in the amount of Cd adsorbed (ZHU Zhi-liang et al 2006). As shown in Fig. 1, the extent of Cd^{+2} removal was minimum at the initial pH 6, because the presence of H^+ ions can inhibit the adsorption of cadmium ions. However Cd adsorption increased with increasing of pH from 6 to 8 significantly. Because by increasing the pH of the solution the surface of the alumina particles contains large number of hydroxyl groups (–OH) that may provide negative charge to the adsorbent which enhanced the adsorption of positively by forming the hydroxy complexes of heavy metals. The effect of pH on adsorption of Cd(II) (Fig. 1) was similar to those described by previous authors including ZHU Zhi-liang et al 2006. As the pH values higher than 9.0, Cd(II) ions precipitated out because of the high concentrations of –OH ions in the aqueous solution.

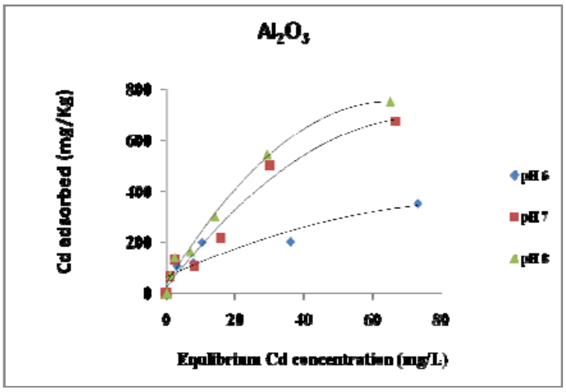

Fig. 1: Effect of pH on Cd(II) adsorption at 25 ċ on Al_2O_3 at pH 6, 7 and 8

Effect of initial Cd^{+2} concentration on adsorption

The initial Cd^{+2} concentration is another important variable that can affect the adsorption process. The results indicated that the adsorption efficiency decreased when the initial concentration increased, and the maximum removing of Cd^{+2} ions carried out at concentration of 2.5 mg /L and decreased with increasing the concentration of Cd^{+2} ions in solution, due to the inhibition of available active sites on the surface of nano Al_2O_3. The amounts of removing efficiency and Cd^{+2} adsorption at different concentrations of Cd, and pH were shown in table 1. These results were an agreement with reports by other authors (Mataka, 2010 & ZHU Zhi-liang, 2007).

Table 1: The effects of initial concentration of Cd on adsorption and removing efficiency

Cd_i (mg/l)	Cd ads (mg/Kg)			RE (%)		
	pH			pH		
	6	7	8	6	7	8
0	0	0	0	-	-	-
2.5	58	65	70	46	52	56
5	103	128	138	41	51	55
10	116	103	160	23.3	20.6	32
20	198	212.5	300	19.8	21	30
40	200	500	541	10	25	27
80	350	675	750	8.75	16.88	18.7

Adsorption isotherm modeling

The equilibrium relationship between the quantity of adsorbate per unit of adsorbent (q_e) and its equilibrium solution concentration (C_e) at a constant-temperature is known as the adsorption isotherm. Adsorption isotherms describe how pollutants interact with sorbent materials and so, are critical in optimizing the use of adsorbents. Hence relationship between the removal ability of the material and the concentration of the contaminant solution could be illustrated by adsorption isotherms. In order to optimize the design of an adsorption system to remove Cd^{+2} from solutions, it is important to establish the most appropriate correlation for the equilibrium curve. Langmuir, Freundlich and Temkin isotherm models were employed to investigate the adsorption process (Table 2). Model fits to equilibrium adsorption results of Al_2O_3 were assessed based on the values of the determination regression coefficient (R^2) of the plot. The adsorption data showed good fit to both Langmuir and Freundlich models than Temkin.

Table 2: Adsorption parameters of the Langmuir and Freundlich and Temkin isotherms at room temperature for the adsorption of Cd(II) on Al_2O_3 nano particles

pH	Langmuir			Freundlich			Temkin		
	q_m (mg/Kg)	$K_{L(L/mg)}$	R^2	n	K_f	R^2	K_T	A	R^2
6	392 c	0.061 a	0.90	0.43 a	54 c	0.95	68 c	15 a	0.90
7	700 b	0.064 a	0.91	0.46 a	83 b	0.95	134 b	15 a	0.88
8	800 a	0.058 a	0.96	0.46 a	91 a	0.98	149 a	7 b	0.92

The results indicated that by increasing pH, adsorption capacity (K_f) significantly increased but the adsorption intensity (n) is not significantly changed. In Langmuir model q_m (the monolayer maximum adsorption) increased with increasing pH from pH 6 to 8, and K_T (coefficient of Temkin models) showed similar trend.

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Arsenic removal by iron oxide mineral: Limonite

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Abstract

Arsenic is usually found in the environment combined with other elements such as oxygen, chlorine and sulfur. Arsenic combined with these elements is called inorganic arsenic. It occurs naturally in soil. The concentration of arsenic in soil varies widely, generally ranging from about 1 to 40 parts of arsenic to a million parts of soil (ppm). The natural occurrence of arsenic in groundwater is directly related to the arsenic complexes present in soils. Arsenic in groundwater causes serious toxicity and potential health risks for human beings and other living organisms. Due to the high toxicity of As, remediation of As contaminated groundwater, is necessary to protect the environment and the public health. Iron oxide is promising adsorptive material for removing inorganic arsenic due to its high surface area and positive charge. In this work the adsorption of As(III) and As(V) on limonite has been studied. The kinetic of the adsorption process was tested for the pseudo-first order and pseudo-second order reactions and intra-particle diffusion models. The rate constants of adsorption for all these kinetic models were calculated. The comparison among the kinetic models showed that the pseudo second-order model best described the adsorption kinetics. Langmuir, Freundlich and Dubinin-Radushkevitch isotherm models were applied to the experimental equilibrium data and the isotherm parameters were determined. The experimental data have been best fitted with Langmuir isotherm model. The results of present investigation showed that limonite is useful adsorbent for the removal of arsenic.

Keywords: Arsenic removal, groundwater, adsorption, iron oxides, limonite

1. Introduction

Arsenic (As) contamination in groundwaters presents a hazard in many countries (Bhattacharya et.al., 2002). It was found that the chronically poisonous effects on humans can appear at even lower concentrations which have widely been found in potable groundwater (Guo et.al., 2009). If groundwater is used for drinking, people are exposed to arsenic (UNICEF, 2008). The major arsenic species present in natural waters are arsenate ions: H_3AsO_4, $H_2AsO_4^{-}$, $HAsO_4^{2-}$, and AsO_4^{3-} (oxidation state V) and arsenite ions, H_3AsO_3, $H_2AsO_3^{-}$ and $HAsO_3^{2-}$ (oxidation state III). However, As(V) ions are most prevalent in oxygenated water while As(III) is found in anaerobic conditions like in well water or in groundwater (Gomes et.al., 2007).. Arsenic cannot be easily destroyed and can only be converted into different forms or transformed into insoluble compounds in combination with other elements, such as iron (Choong et.al., 2007).

The groundwater contains more than 50 µg/L of arsenic (Mcarthur et.al., 2001). Following the accumulation of evidence for the chronic toxicological effects of as in drinking water, recommended and regulatory limits of many authorities are being reduced. The WHO (World Health Organization) guideline value for as in drinking water was provisionally reduced in 1993 from 50 µg/l to 10 µg/l. The US-EPA (United States Environmental Protection Agency) limit was also reduced from 50 µg/l to 10 µg/l in January 2001 following prolonged debate over the most appropriate limit. The EC (European Commission) maximum admissible concentration (MAC) for As in drinking water is also to be reduced to 10 µg/l in 1998 (Smedley and Kinniburgh, 2001).

The lowering of As drinking water standard requires effective and cheap technologies for As removal from the As drinking water. Among a variety of technologies (including precipitation coagulation, membrane separation, ion exchange, lime softening and adsorption), adsorption and coagulation are believed to be the cheapest as removal methods (Guo and Zhao, 2010).

In this work the adsorption of As (III) and As (V) on limonite has been studied. The present work reports the results for kinetic and equilibrium adsorption studies of As(V) and As(III) onto limonite. Kinetic and equilibrium isotherm models were used to establish the rate of adsorption, adsorption capacity, and the mechanism for As adsorption. Results of this work will be useful for future scale up using this limonit as adsorbent for the removal of As from groundwater.

2. Materials and Methods
2.1. Materials

The natural Fe-mineral (limonite) used in this study. The mineral samples were ground and sieved to produce various particle size fractions. Particle size fraction of 0.25–0.50 mm used in this study. All chemicals used were of analytical grade. All glass and plastic equipment used in bench-scale testing was cleaned and acid washed using %10 nitric acid, triple-rinsed with deionized water. Stock solutions of arsenate and arsenite containing 5000 mg As(V)-As(III)/L were prepared using $Na_2HAsO_4.7H_2O$ and $NaAsO_2$. The stock solutions were stored at 4°C in the refrigator. An arsenate and arsenite solutions at an initial experimental concentration of 500 µg As(V)-As(III)/L were prepared on the day of experimentation by diluting the stock solution with deionized water. Hydrochloric acid and sodium hydroxide solutions were prepared for pH adjustment.

2.2. Experimental
The adsorption experiments were conducted at 30°C. A constant mass of solid (0.1 g) was put in contact with 50 mL of arsenate and arsenite solutions at 500 µg of As(V)-As(III)/ L . The batch experiments were performed in a mechanical shaker at an agitation speed of 150 rpm. The pH of suspension was adjusted to value 7 by adding either HCl or NaOH (1 M, 0.1 M/0.01 M). After the required reaction time, the supernatants were filtered through 0.45 mm pore size membran filters. Arsenic concentrations in the supernatants were measured by Inductively Coupled Plasma-Mass Spectrometry. Kinetic studies were conducted with 500 µg As(V) or As(III)/L concentration and between 30 seconds and 96 hours. Isotherm studies were conducted with initial As(V) or As(III) concentrations between 100 and 1000 µg/L, and a contact time of 84 h.

2.3. Analytical methods
Solution pH was monitored by a standard pH meter (Sartorius, PB-10). As was analyzed by ICP-MS için (Perkinelmer Optima 2100 DV ICP-OES- USA). The chemical compositions of limonite were analyzed using x-ray fluorescence (XRF). Table 1 shows the chemical compositions of limonite. Morphological analysis of used adsorbent was performed by scanning electron microscopy (SEM). Fig. 1 shows the SEM micrograph for limonite.

Table 1. The chemical compositions of limonite

Adsorbent	Elements	(%)	Elements	(%)
Limonite	Al	2.00	O	32.348
	Ba	0.187	P	0.025
	Ca	1.102	Pb	0.038
	Cl	0.021	S	0.026
	Cu	0.076	Si	2.127
	Fe	55.542	Sr	0.009
	K	0.190	Ti	0.021
	Mg	0.937	Zn	0.160
	Mn	5.155	Zr	0.003
	Ni	0.033	-	-

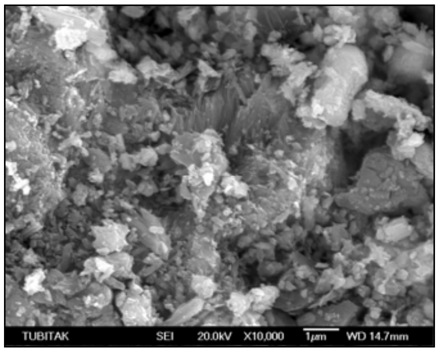

Fig. 1. SEM image of Limonite

4. Results and discussion
4.1. Adsorption kinetics
In order to investigate the adsorption processes of As(V) and As(III), three kinetic models (pseudo-firstorder, pseudo-second-order and intra-particle diffusion models) were used. Kinetic studies were conducted between 30 minutes and 96 hours. Equilibrium is reached in 84 h.

4.1.1. Pseudo-first-order model

The pseudo-first-order equation (Weng et.al., 2007) based on the adsorption capacity has the form of

$$\frac{dq}{dt} = k_1(q_e - q) \quad (6)$$

where qe is amounts of adsorbed As onto Limonite at equilibrium (µg/g); k1 the pseudo-first-order rate constant (min^{-1}). If the initial adsorbed concentration at t = 0 has qt = 0, and if at t = t, the adsorbed concentration is qt = qt, then Eq. (6) becomes

$$q_t = q_e(1 - \frac{1}{10^{(k1/2.303)t}}) \quad (7)$$

4.1.2. Pseudo-second-order model

The pseudo-second-order equation (Weng et.al., 2007) can be expressed as

$$\frac{dq}{dt} = k2(qe - q)^2 \quad (8)$$

where k2 is the pseudo-second-order rate constant (g/µg-min). By integration, Eq. (8) becomes

$$qt = \frac{k2.qe^2.t}{1 + k2.qe.t}$$

4.1.3. Intra-particle diffusion study

The possibility of intra-particle diffusion was explored by using the intra-particle diffusion model (Mall et.al., 2007)

$$qt = kid.t^{1/2} + I \quad (9)$$

where $k$id is the intra-particle diffusion rate constant (µg/g $min^{1/2}$) and I (µg/g) is a constant that gives idea about the thickness of the boundary layer, i.e., larger the value of I the greater is the boundary layer effect.

The best fits of these models are shown in Fig. 1 a, b and c. Parameters for the model results are summarized in Table 2.

It was found that the adsorption kinetics was best described by the pseudo-second-order model with relatively low standard deviations and high correlation coefficient R^2 values. The calculated correlation coefficients are closer to unity for pseudo-second-order kinetics than that for the pseudo-first-order kinetic model. Therefore, the sorption can be approximated more appropriately by the pseudo-second-order kinetic model than the first-order kinetic model for the adsorption of As ions onto Limonite. From Fig. 2-c, it may be seen that there are three separate regions the first straight portion is attributed to the macropore diffusion (phase I) and the second and third linear portion to micro-pore diffusion (phase II-III). Phase (III) may be attributed to a very slow diffusion of the adsorbates from the surface film into the micro-pores, which are the least accessible sites of adsorption.

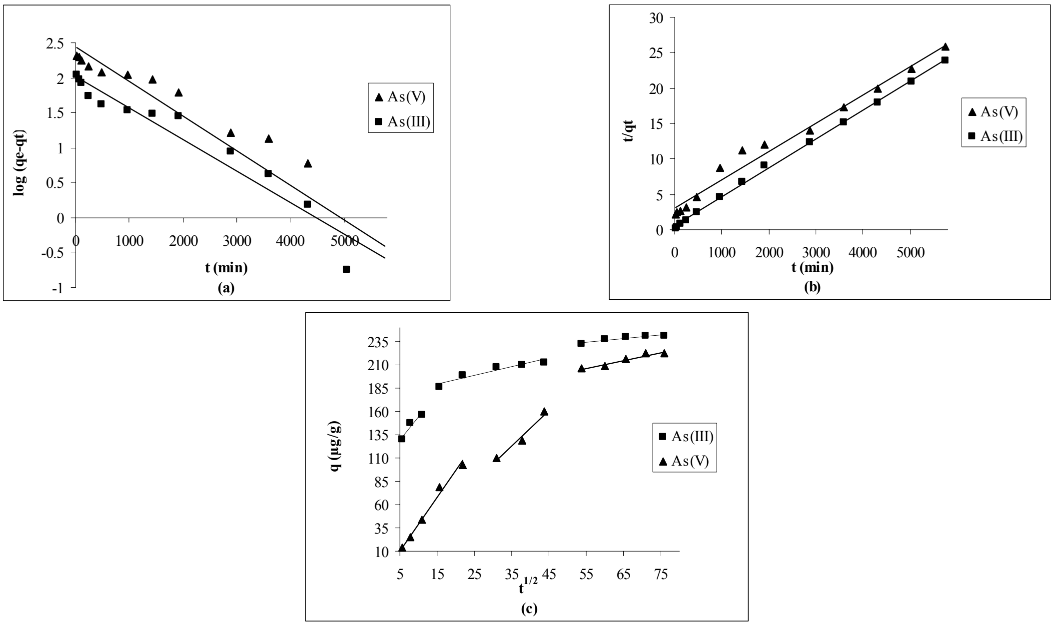

Fig. 2. Pseudo-first-order (a), Pseudo-second-order (b) and intra-particle-diffusion (c) kinetic plots for removal of As(V) and As(III) by Limonite (temperature=30°C, initial AS concentration=500 µg/l, limonite dosage=0.1 g/50 ml, 150 rpm).

Table 2. Kinetic parameters for the removal of As(V) and As(III) by limonite

	Pseudo-first-order model			Pseudo-second-order model				Intra-particle diffusion model					
	k_1 min^{-1}	q_e (µg/g)	R^2	k_2' (g/µg min)	q_e (µg/g)	h (µg/g min)	R^2	k_{i1} (mg/g $min^{1/2}$)	R^2	k_{i1} (mg/g $min^{1/2}$)	R^2	k_{i1} (mg/g $min^{1/2}$)	R^2
As(V)	0.0002	283,1	0.84	$5,28*10^{-6}$	250	0.33	0.98	5.645	0.98	3.793	0.96	0.833	0.94
As(III)	0.0002	107,1	0.93	$3,43*10^{-5}$	250	2,14	0.99	4.821	0.91	0.92	91	0.4	0.89

4.2. Adsorption isotherms
4.2.1. Langmuir model

The Langmuir model (Hamdaoui and Naffrechoux, 2007) assumes uniform energies of adsorption onto the surface and no transmigration of adsorbate in the plane of the surface. The Langmuir equation may be written as

$$qe = \frac{qm.b.Ce}{1 + b.Ce} \qquad (1)$$

where *q*e is the amount of solute adsorbed per unit weight of adsorbent at equilibrium (µg g−1), *C*e the equilibrium concentration of the solute in the bulk solution (µg L^{-1}), *q*m the maximum adsorption capacity (µg g−1), and *b* is the constant related to the free energy of adsorption (L $µg^{-1}$). Eq. (1) can be linearized to different linear forms as shown in Figure 3.

4.2.2. Freundlich model

The Freundlich equation (Hamdaoui and Naffrechoux, 2007) can be written as

$$qe = K_F.Ce^{1/n} \qquad (2)$$

where *K*F is a constant indicative of the relative adsorption capacity of the adsorbent and *n* is a constant indicative of the intensity of the adsorption. The Freundlich expression is an exponential equation and therefore, assumes that as the adsorbate concentration increases, the concentration of adsorbate on the adsorbent surface also increases. The linear form of the Freundlich isotherm is shown in Figure 3.

4.2.3. Dubinin–Radushkevich isotherm

The Dubinin–Radushkevich (Kumar et.al., 2010) has the following form

$$qe = qm.e^{-\beta e^2} \tag{3}$$

where qm is the Dubinin–Radushkevich monolayer capacity (μg/g), β a constant related to sorption energy, and ε is the Polanyi potential which is related to the equilibrium concentration as follows

$$\varepsilon = R.T.\ln(1+\frac{1}{Ce}) \tag{4}$$

where R is the gas constant (8.314 J/mol K) and T is the absolute temperature. The constant β gives the mean free energy, E, of sorption per molecule of the sorbate when it is transferred to the surface of the solid from infinity in the solution and can be computed using the relationship:

$$E = \frac{1}{\sqrt{2\beta}} \tag{5}$$

The graphical representations of isotherm models are presented in Fig. 3 a, b and c. All of the constants are presented in Table 3. Since the value of R^2 nearer to 1 indicates that the respective equation better fits the experimental data. The experimental data yielded excellent fits with in the following isotherms order: Langmuir, Dubinin–Radushkevich and Freundlich based on its R^2 values. Langmuir isotherm model assumes a monolayer surface coverage limiting the adsorption due to the surface saturation, while Freundlich isotherm model is an empirical model allowing for multilayer adsorption.

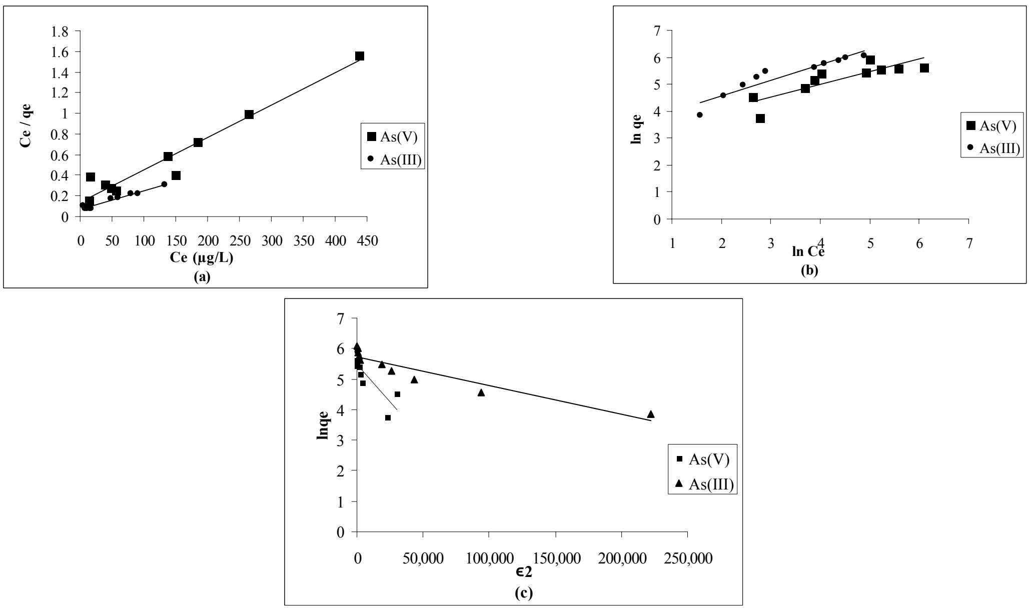

Fig. 3. Langmuir (a), Freundlich (b) and *D–R* (c) ısotherm plots for removal of As(V) and As(III) by Limonite (temperature=30°C, time= 5760 min., limonite dosage=0.1 g/50 ml, 150 rpm).

Table 3. Isotherm parameters for removal of As(V) and As(III) by limonite

Isotherm	Constants	Results for As(V)	Results for As(III)
Langmuir adsorption isotherm	Q^o (μg/g)	322,58	555,55
	b	0.0174	0.027
	R^2	0.9725	0.9612
	RL	0.103	0.068
Freundlich adsorption isotherm	K_F(μg/g)/(μg/l)$^{1/n}$)	19.49	30.47
	n	2.05	1.74
	R^2	0.723	0.8859
D - R adsorption isotherm	K	0.0176	0.0131
	X_m	0.03	0.05
	E (kJ/μg)	5.3	6.17
	R^2	0.86	0.93

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Kinetics of Pb Desorption from Two Calcareous Soil Textural Classes as Influenced by Organic and Mineral Pb Sources

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Abstract
Adsorption and desorption are among the most influential soil processes controlling mobility of heavy metals including lead (Pb). A laboratory study was performed to investigate the influence of soil texture (sandy loam and clay loam), Pb sources ($Pb(NO_3)_2$ and enriched compost) and levels on the extraction of this metal by diethylenetriamine tetraacetic acid (DTPA) and its desorption patterns at ten shaking periods. The soil textural classes were polluted with five Pb levels and incubated for a month. Soil samples were extracted using DTPA for shaking periods of 1 to 1440 min. Kinetic models commonly used in nutrients release study including zero-, first-, second- and third-order, parabolic diffusion, Elovich and power function equations were used in the study. Results showed that Power function model ($q=at^b$) described pattern of Pb desorption better than other kinetic models. Results showed that Pb extraction rates increased as Pb levels increased, and was more considerable in sandy loam soil with Pb-mineral source in comparison with clay loam soil and Pb-organic source. The *a* value (Pb desorption magnitude constant) was the highest in sandy loam soil amended with mineral source of Pb. The lowest value of *a*; however, was observed in clay loam soil receiving organic source of Pb. The *ab* coefficient (initial desorption rate of Pb) was higher in sandy loam soil in comparison with clay loam soil demonstrating higher initial release rates of coarse textural soil. Addition of mineral source of Pb increased *ab* value in comparison with organic source in both soil textural classes.
Keywords: kinetics, desorption, lead (Pb), calcareous soil

Introduction
Knowledge on the kinetics of adsorption and desorption reactions of heavy and trace metals in soils are important in controlling mobility and bioavailability of these metals in the environment and prediction of ions availability in soils. The bioavailability of Pb in soil is highly dependent on the sorption and desorption characteristics of soil (mouni et al., 2009). Zinati et al., (2004) reported that the application of compost in agricultural soils, improving soil physical and chemical conditions and act as a source of plant nutrients. Composts usually contain higher heavy metal concentration than most agricultural soils. Soils with different properties have various capacities for heavy metals retention (Rajaei et al., 2006). Maftoun et al., (2004) reported that Cd sorption in calcareous soils is depending on a wide range of soil physical and chemical properties. This research was initiated to investigate the influence of Pb levels and sources on kinstics of Pb desorption in two calcareous soil textural classes.

Methods and materials
A bulk sample from 0–30 cm soil surface was collected from Bajgah Agricultural Experiment Station of Shiraz University, Shiraz, Iran. The soil was air-dried and passed through a 2-mm sieve before analysis. Soil textural class, determined by the hydrometer method (Bouyoucos, 1962) was clay loam. A portion of pure quartz sand was acid-washed and added to the soil to obtain sandy loam textural class. Some chemical and physical soil characteristics were measured according to standard procedure (Table 1). Compost as mineral source of Pb, was air-dried, and passed through a 2-mm sieve. Some of compost characteristics were determined (Table 2). For raising the total Pb content of compost to such a level that, when added to soil at 3% would rich the soil Pb to 50, 250, 500 and 1000 mg Pb kg^{-1}, enough $Pb(NO_3)_2$ was added to compost and kept it moistened and incubated for 1 month at room temperature to equilibrate. Ten g of each soil textures, in triplicate were extracted by 20 mL of DTPA extractant and shaken for periods of 1, 5, 15, 30, 120, 240, 450, 840 and 1440 min. At the end of each desorption period, the soil suspensions were centrifuged for 15 min at 2500 rpm, and the supernatants were collected for measurement of Pb concentration with atomic absorption spectrophotometer. Seven kinetic models were used in this study (Table 3). The best kinetic models selected by high coefficient of determination (R^2) and low standard error of estimate (SE).

Table 1- Selected chemical and physical properties of the soil textural classes.

Property	Clay loam	Sandy loam
Sand%	29.84	71.72
Silt%	34	15.38
Clay%	36.16	13
CEC ($cmol_+kg^{-1}$)	200	110
CCE%	32	14
OM%	1.2	0.75
pH	7.4	7.5
DTPA extractable Pb (mg kg^{-1})	n.d	n.d

n.d: not detected

Table 2- Selected chemical and physical properties of the compost

Characteristics	compost
DTPA-extractable of Pb (mg kg^{-1})	1
DTPA-extractable of Fe (mg kg^{-1})	2408
DTPA-extractable of Cu (mg kg^{-1})	1.8
DTPA-extractable of Mn (mg kg^{-1})	247.95
DTPA-extractable of Zn (mg kg^{-1})	83.2
1M NH_4OAc-extractable K (mg kg^{-1})	3450
NaOAc- extractable P (mg kg^{-1})	2187.42

Table 3- kinetic models used in Pb release study

Model	Equation	Parameter
Zero order	$q_t=q_0-k_0t$	k_0, Zero-order rate constant (mg Pb kg^{-1} min^{-1})
First order	$Lnq_t=lnq_0-k_1t$	k_1,first-order rate constant (min^{-1})
Second order	$1/q_t=1/q_0-k_2t$	k_2, second-order rate constant[(mg Pb $kg^{-1})^{-1}$]
Third order	$1/q_t^2=1/q_0^2-k_3t$	k_3, third-order rate constant [(mg Pb $kg^{-1})^{-2}$ min^{-2}]
Parabolic	$q_t=q_0+kpt_0^{.5}$	k_p, diffusion rate constant
Exponential	$q_t=at^b$	*a*, Pb sorption magnitude constant [(mg Pb kg^{-1}(min^{-1})b] and *b* sorption rate constant
Simple Elovich	$Q_t=1/\beta_s \ln s\beta_s+1/\beta_s \ln t$	α_s,initial Pb sorption constant (mg Pb kg^{-1} min-1) and, β_s, Pb sorption rate constant [(mg Pb kg^{-1}) $^{-1}$]

q_0 and q_t are the amount of Pb desorption (mg Pb kg^{-1}) at time zero and t, respectively.

Result and discussion

Results showed that the effects of texture, Pb source and levels were significant on Pb desorption. The amount of desorption in sandy loams was significantly higher than clay loam soil (Table 4), which is probably due to the low levels of organic matter in sandy loam soil as compared to clay loam soil. The results indicated that the desorption was higher in mineral source than the organic source (Table 4). Oh et al (2010) reported that Pb desorption decreased with an increase in CEC and organic carbon. Since the sorption of heavy metals including Pb increased with higher levels of CEC and organic carbon. Strawn and sparks (2000) reported that soils treated with compost and sewage sludge had a high capacity in immobilization of heavy metals specifically Pb, which is because of possessing high OM and CEC. Results indicated that with an increase in Pb concentration, the desorption increased at all times (Table 5). Dutta et al., (2011) reported similar results the highest Pb desorption rate was observed in sandy loam and at 1000 mg Pb level. This seemed to be due to the fact that in sandy loam the active fractions of soil are low and Pb was retained in soil with much less energy. Karaka (2004) found a negative correlation between OM and the extractable levels of Cu and Cd, which was due to high CEC of OM and their high potential in forming complexes with metals. With increasing Pb levels in the mineral source desorption also increased. In the organic source a similar trend was also observed

Table 4- The amount of Pb desorption at different shaking periods.

Time	Soil texture		Pb Source	
(min)	Clay Loam	Sandy Loam	Inorganic Source	Organic Source
1	37.83b*	61.34a	36.68b	62.49a
5	50.36b	74.79a	45.91b	79.24a
15	70.10b	94.11a	60.51b	103.70a
30	83.54b	110.49a	72.62b	121.41a
60	107.43b	137.33a	101.47b	143.29a
120	136.34b	165.57a	138.49b	163.41a
240	171.51b	201.22a	161.37b	211.35a
450	204.12b	230.43a	193.23b	241.32a
840	224.53b	246.78a	213.98b	257.33a
1440	244.75b	273.85a	235.87b	282.72a

*Means followed by the same letters at each column and row are not significantly different according to LSD test at the level P<0.05.

Table 5- Effect of Pb levels on Pb desorption.

Time	Pb levels (mg kg^{-1})				
(min)	1	50	250	500	1000
1	0.71e*	11.23d	38.78c	82.44b	114.76a
5	0.75e	12.88d	46.39c	97.89b	154.96a
15	0.79e	14.61d	66.73c	115.6b	212.8a
30	0.84	16.49d	74.06	131.08b	262.62a
60	0.87e	19.69d	99.55c	161.09b	330.71a
120	0.9e	23.81d	123.12c	203.32b	403.6a
240	0.92e	27.11d	142.37c	248.1b	513.32a
450	0.94e	32.2d	163.09c	293.42b	596.71a
840	0.95e	36.84d	716.97c	324.93b	638.58a
1440	0.95e	41.95d	201.52c	369.71b	682.36a

*Means followed by the same letters at each column and row are not significantly different according to LSD test at the level P<0.05.

Correlation coefficients between the amount of desorbed Pb and the different lengths of shaking time indicated that the amounts of Pb desorption was increased in all treatments as the length of shaking periods increased. Desorption rates was higher at the initial periods and followed by a slower rate. Dong et al., (2009) reported that with increasing the shaking time the concentration of extracted elements increased in a way that the extraction was higher at first times and reached equilibrium in 10 hours and followed by a slower rate of extraction. In both textures the two-constant-rate equation was the best fitting equation to describe Pb desorption, data not shown. Most researchers have reported that equations having high R^2 and low SE are highly desirable (Elkhatib 2007). Reyhanitabar et al., (2010) studied the different kinetic models in desorption of Zn in calcareous soils and reported that the two-constant equation was the best option in Zn desorption. The value of constant a, b and ab for two constant-rate are shown in (Table 6). The results showed that mean *a* value in sandy loam and mineral source at 1000 mg Pb was the highest and at 1 mg reached the lowest. The *a* value at 1000 mg in sandy loam and the mineral source was 2.64 times higher than in sandy loams receiving organic source. In clay loam there was no significant difference between the two sources. In sandy loam and clay loam, the *a* value increased as pb level increased. The *a* value in sandy loam soil was twice of clay loam, which proved that the Pb desorption in sandy loam was higher than clay loam soil. Mean value for mineral source treatmant was 2.4 times higher than organic source treatmant. The data indicated that the highest *b* constant in clay loam and the organic source at 1000 mg Pb showed a 7.86 fold increase as compared to

Table 6- Effects of Pb levels, texture and sources on values of rate constant for two- constant rate model

Pb level (mg/kg)	Sandy loam Mineral source	Sandy loam Organic source	Mean	Clay loam Mineral source	Clay loam Organic source	Mean
			a			
1	0.6n*	0.59n	**0.6E**	0.6n	0.59n	**0.59E**
50	5.73k	4.19l	**4.96E**	4.29l	3.22m	**3.75E**
250	24.76e	16.46h	**20.61CD**	8.52j	4.23l	**6.38DE**
500	57.88b	18.01f	**37.95AB**	29.32d	13.07i	**21.2C**
1000	79.17a	24.56e	**51.86A**	33.81c	17.26g	**25.54BC**
Mean	**33.63A**	**12.76B**	**23.19A**	**15.31B**	**7.67B**	**11.49B**
			b			
10	0.047p	0.038r	**0.042D**	0.046p	0.042q	**0.044D**
50	0.181n	0.196l	**0.188C**	0.186m	0.21j	**0.198C**
100	0.204k	0.218i	**0.211C**	0.286d	0.32b	**0.305A**
200	0.168o	0.25g	**0.213C**	0.22h	0.281e	**0.252B**
400	0.211j	0.29c	**0.251B**	0.27f	0.33a	**0.3A**
Mean	**0.16B**	**0.2AB**	**0.18B**	**0.202AB**	**0.23A**	**0.22**
			ab			
10	0.028p	0.023p	**0.026E**	0.028p	0.025p	**0.027E**
50	1.04m	0.82n	**0.93E**	0.8n	0.67o	**0.74E**
100	5.07g	3.59j	**4.33CD**	2.44k	1.37l	**1.9DE**
200	9.75b	4.65h	**7.2B**	6.55e	3.68i	**5.11BC**
400	16.77a	7.16d	**11.97A**	9.17c	5.7f	**7.44B**
Mean	**6.53A**	**3.25B**	**4.89A**	**3.8AB**	**2.29B**	**3.04B**

*Means followed by the same letters at each column are not significantly different according to LSD test at the level P<0.05.

the mineral source at 1 mg Pb level. The results showed that applying different Pb levels increased the *b* value significantly. The clay loam texture caused a 22.22% increase in the *b* value compared to the sandy loam soil. The data showed that the *b* value for organic source was about 22% higher than mineral source. By increasing the Pb levels the *b* value increased in both sources. The Sandy loam texture with the mineral source increased the *ab* (initial adsorption rate of Pb) coefficient almost twice compared with the sandy loam with the organic source. The *ab* coefficient was approximately twice in sandy loam compared to clay loam soil, which is due to the low active fraction of soil in sandy loam soil. The data showed that mean *ab* coefficient in mineral source was about 86% more than the organic source since organic matter bonds with Pb more efficiently than the mineral soil fractions. Reyhanitabar et al., (2010) reported that *a* and *b* constants changed with soil properties.

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The effect of Zeolite on germination and growth of Agropyron (*Agropyron smithii 1.*) and sunflower (*Helianthus annusl)* in petroleum Contaminated soil of Esfahan oil refinery

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Abstract

Soil contamination by hydrocarbon is a serious treat for environment and this contamination in soil maybe toxic to human, plants and soil microorganism and poses treat to ground water and food chains. Therefore remediation of this compound is vital. Phytoremediation is becoming a cost effective technology for the clean up of sites polluted with petroleum hydrocarbons. Difference in the ability of various crops to germinate and growth in contaminated soils should be better explored to choose the most appropriate plant species in the development of any Phytoremediation process. Zeolite is benefit material for soil amendment, these material increased of efficiency. In this study we investigated the phytoremediation efficiency of two plant including Agropyron (*Agropyron smithii 1.*) and sunflower (*Heslianthus annusl*. Soil with four contamination levels were collected from Esfahan refinery zone (C0 : uncontaminated soil, C1 :1:1 w/w, un contaminated: contaminate soil, C2 :1:3 w/w, uncontaminated: contaminated soil, C4 :contaminatedsoil) and three levels of Zeolite (without Zeolites, 0.5 and 5%). Presence of total petroleum hydrocarbons (TPHs) in the soil had no effect on seed germination of agropyron and sunflower. According to these results the dry matter yield of agropyron was higher than sunflower (%45) ,also dry matter yield in Zeolite treatments (0.5 and 5%) were higher than control (23 and 33% respectively) . The results showed that agropyron is suitable choices for growth and germination of investigated petroleum contaminated soil. Zeolite is benefit for germination and dry matter yield of investigated petroleum contaminated soil.

Keyword: Zeolite, Phytoremediation, TPH_S, Germination

Introduction

Total Petroleum hydrocarbons (TPH_s) are one of the most common group of Persistent organic contaminants in the environment and are known to be toxic to the environment and are known to be toxic to many organisms. There are many sources of TPH contamination in soils including petroleum extraction, transportation, refining and consumption (US.EPA.,2000).Petroleum Hydrocarbons in soil maybe toxic to human getting in contact with such soil, plants and microorganism and also pose arist of grandwater contamination (Besalatpour et al., 2008).There is serious concern about the environmental presence of TPH_S, especially their potential for bioaccumulation in food chains (Jian et al., 2004). Thus remediation of these compounds from contaminated soil is vital. There are different method can be used for remediation of contaminated sites such as Physical, chemical and biological method, different approaches to reduce this problem have failed so far (BesalatPour et al., 2008).To find more environmentally and economically acceptable options, biological methods such as Phytoremediation have been investigated. Phytoremediation has long been recognized as a cost- effective method for removal of these pollutants from soil (Aprill and sims, 1990).Several studies have aimed at quantifying the effectiveness of specific Plants in stimulating the degradation of Petroleum contaminants in soils (Palmroth et al., 2002).The presence of Petroleum hydrocarbons in soils is a negative factor for plant growth and development and this compound make stress for plant (li et al., 1997).Their harmful effects include inhibition of seed germination (Adam and Duncan, 2002).Lack of germination due to the lake of viable seeds (ogboghodo et al., 2004).The Plants that should be chosen carefully so that they provide a maximum root surface area (April and sims, 1990).Grasses have been used for germination of contaminated soils due to their fibrous root systems with extensive surface area for microbial colonization (Adam et al., 2002).To a achieve a optimum condition for growth of chosen plant we use zeolite. Zeolites are a naturally occurring mineral group consisting of 50 mineral Types and it is widely distributed in huge deposit in different regions of Iran (Hejazi and Ghorbani., 1994). Zeolites are micro porous material that play important roles in Ion, exchange, catalysis and adsorption sciences and the molecular sieving Properties of zeolites are exploited in many industrial applications (Choudary and newalkar,2010).The main properties of zeolite is high cation exchange capacity (CEC)arising from the substitution of Al for SI in the silicon oxide tetrahedral units that constitute the mineral structure (Vaughan., 1978).Natural zeolite are particularly useful in agriculture because of their large porosity high exchange capacity and their selectivity for NH_4^+ and K^+ cations as well as their physical stability (Vaughan., 1978).There are several petroleum refineries in Iran, such as the Esfahan oil refinery, unfortunately environmental Pollution is increasingly becoming a great concern in these regions, So that in some Parts ecosystem is subject to serious challenge. For reduce environmental hazards of Petroleum hydrocarbons around the Esfahan oil refinery, in this study we use two plant that were compare the ability of these to germinate and in petroleum contaminated soil and we use tree level of zeolite to investigated the effect of zeolite on germination and dry biomass.

Material and methods

Bulk samples (300kg) of uncontaminated and petroleum- contaminated surface (0-40 cm) soil were collected from farmland and oil waste land fills around the Esfahan oil refinery. Soils were air dried and passed through a 4-mm sieve. Four contamination levels were prepared by mixing contaminated and uncontaminated soil at various weight soils. Including C_0 (uncontaminated soil), C_1 (1:1 w/w), uncontaminated: contaminated soil. C_2 (1:3 w/w, uncontaminated: contaminated soil). The soil samples were land farmed in an open field for 2**1** days. The process for land farming included irrigation (near 70% field capacity and aeration the contaminated soil every 3 days by hand using a garden hoe after that we used zeolite at various weight ration including (without zeolite (z_0), 0/5 (z_1), 5% (z_2) the type of zeolite that we used in this study was clinoptilolite. The size of zeolite was smaller than of 106 μm.Selected physical and chemical properties of the uncontaminated and contaminated soil samples are shown in Table 1.

Table1.Physical-chemical properties and TPH concentration of the experimental soils (C_0,C_1,C_2,C_3)

treatment	pH $CaCl_2$	EC $dS.m^{-1}$	Clay	O.M	N Total	P Available	K Available	Fe DTPA	Cu DTPA	Mn DTPA	Zn DTPA
			%			mg.kg^{-1}					
C_0	7.8	5.27	15	2	0.05	60	72	35	1.3	10	6.8
C_1	7.2	8.93	15	6.7	0.08	41.9	117	19.7	1.5	46	14
C_2	7	12.35	11	10.08	0.09	52.28	127	28.75	7.5	60	28
C_3	6.8	15	11	13.4	0.14	56.49	143	35	10	86	30

Total petroleum Hydrocarbons were extracted from 10 gr soil sample by soxhlet for 24-hour solvents used in the trace analysis quality were n-hexane-dichloromethane (150 ml) to clean vials in the pure analysis quality (Christopher et al., 1988).The TPH_S concentration in different contamination treatments are shown in Table 2.

Table2. Concentrations of measured TPH_S in different contamination treatments.

Treatment	mg kg^{-1}
C_0	<100
C_1	52000
C_2	76000
C_3	111600

Concentration of PAH_s were evaluated in soil extracts using gas- Chromatography (GC)with a Delsi DI 200 chromatograph equipped with a direct injection port and FID detector both set at 340°c, Carrier gas was helium under 0/08 MP, column was a CP sill 5CB (chrom pack) capillary column (50m bg 0/32 mm, film thickness-0/25 μm); The temperature was increased from 100 to 320°c at a rate of 3°c min^{-1}. Selected PAH_S in (c_2) treatment is shown in table 3.

Table 3. PAH_S treatment

PAH	$Mgkg^{-1}$
Naphthalene	42
Acenaphthylenc	18
Acenaphthene	12
Flourene	12
Phenanthrene	34.3
Anthracene	12
Fluoranthen	12
Pyrene	18.3
Benzo (a) anthracene	18
Chrysene	12
Benzo (b) fluoranthene	8
Benzo (k) fluoranthene	18
Benzo (a) pyrene	8
Indeno (1, 2, 3, … , cd) pyrene	8
Dibenzo (a, h) anthyacene	13.8
Benzo (g, h, i) perylene	14.4

For greenhouse experiment we use two plant including: Agropyron (*Agropyron smittii L.*) and sunflower (*Heliantus annus L.*). Seeds were planted at 1 to 2 cm depth in plastic pots having 150 mm diameter and 250 mm high with about 3kg soil in each replicates A disc of filter paper was placed at the bottom of each pot to prevent soil loss through the drainage holes. Soils moisture was kept near 70% field capacity by watering the pots from the top during the experimental period in each trial. The plants were grown for 8 weeks in the greenhouse without fertilization. At the end of the experiment, all plants were harvested and separated into shoots and roots. Both shoots and roots were washed first with tap and then with distilled water, dried in an oven and weighed for determination of dry biomass. The experiment was set up in a completely randomized factorial design Result were analyzed by Analysis of variance using the SAS computer program (SAS institute, 1988).

Results

The effect of petroleum contaminants an dry biomass of the studied plants are shown in figures (1).

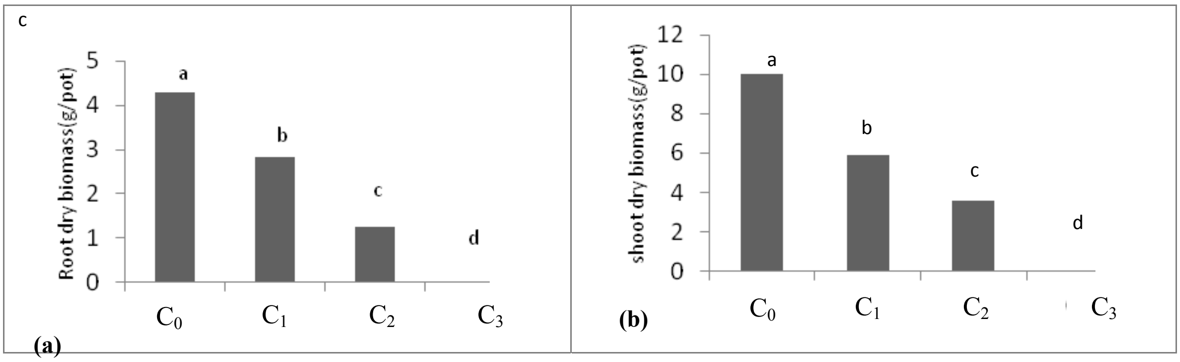

Fig 1. The effect of petroleum contaminants on root dry biomass (a) and shoot dry biomass (b) of the studied plants.

Figure 1 shows the root and shoot dry biomass produced by studied plants at soil TPH concentration of < 100 mg kg^{-1} (C_0), 52000 mg kg^{-1} (C_1) and 76000 $mgkg^{-1}$(C_2) and 111600 $mgkg^{-1}$ (C_3).

According to this result the shoot was found decrease in the root and shoot dry biomass in the C1 shoot treatments as compared to the C_0 treatments and in the C_2 treatments as compared to the C_1 treatments, the dry biomass decrease in the C_3 treatments as compared to the C_2 treatments.

The effect of studied plants on dry biomass are shown in figure (2).

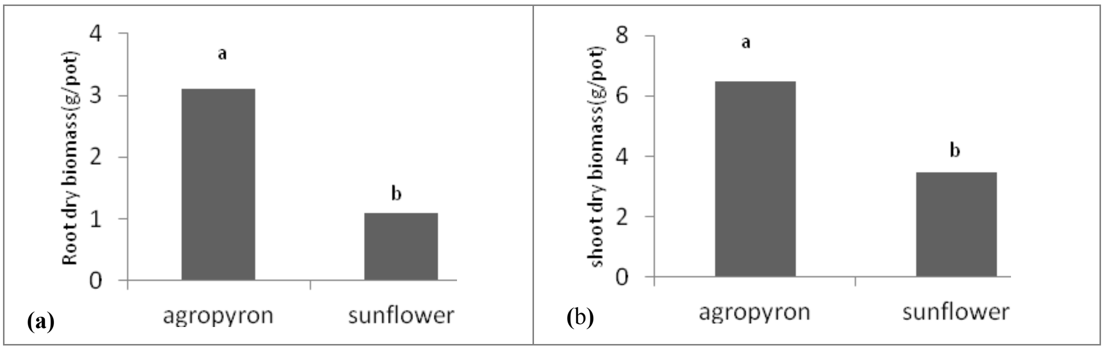

Figure 2. The effects of studied plants on root dry biomass (a) and shoot dry biomass (b)

Figure 2 shows the root and shoot dry biomass produced by agropyron is more than sunflower. The effect of zeolite level on dry biomass of the studied plants are shown in figures (3).

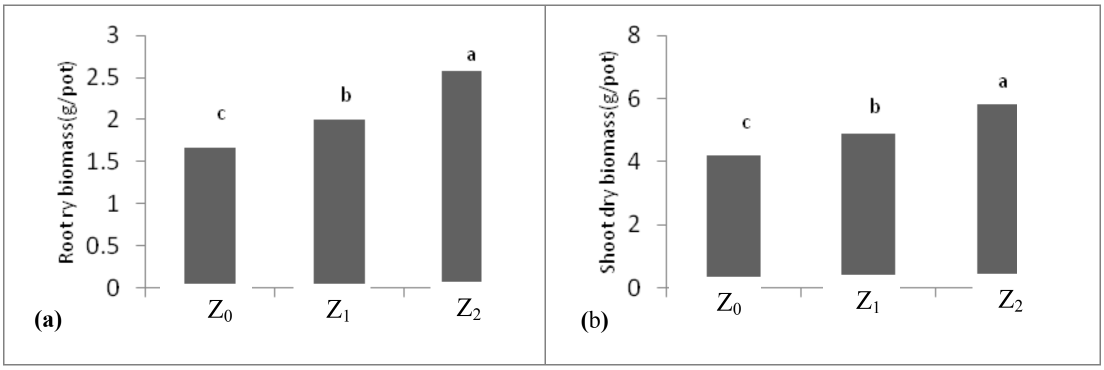

Figure 3. The effect of zeolite treatments on root dry biomass (a) and shoot dry biomass (b) of the studied plants.

Figure 3 shows that the root and shoot dry biomass produced by studied plants at zeolite level of (Z_0) treatment, (Z_1) treatment (0/5%) and (Z_2) treatment (5%).According to this result was found increased of root and shoot dry biomass in the Z_2 treatments as compared to the Z_1 treatments and was found increased of root and shoot dry biomass in the Z_1 treatment as compared to the Z_0 treatment.

Discussion

According to this result seed germination started with 4 and 5 day delayed in C_1 and C_2 treatment and germination started with 18 day delay in C_3 treatment and with increasing contamination level we observed that the dry biomass have decrease. Some study reported about germination and develop in the contamination treatment. For example chaineau et al (1997) Identified that increasing soil concentration of fuel oil prohibited 50% of seeds germinating. Dibble and Bartha (1979) reported that the slower rate of germination in the soil contaminated with petroleum contaminated may have been result of decrease oxygen availability and consequently, increase competition between the germination seeds and microorganisms. In addition, inhibition of plant growth on petroleum contaminated soil may also result from reduced water and nutrient availability due to hydrophobicity effects (kechavarzi et al., 2007).In this study we considerate the effect of natural zeolite on dry biomass of chosen plant this type of zeolite are widely distributed in huge deposit in different regions of Iran (Hejazi and Ghorbani, 1994).Natural zeolite are particularly useful in agriculture because of their large porosity; high cation exchange capacity and their selectivity for NH_4^+ and K^+ cation as well as their physical stability (Vaughan., 1978). The main use of natural zeolites in agriculture is, however, for ammonium NH_4^+ exchange, storage and slow release. It has been shown that zeolite, with their specific selectivity for NH_4^+ , can take up NH_4^+ from farmyard manure, composts or NH_4^+ from ammonium bearing fertilizers and store it temporarily in the internal coid spaces before slow release (Mumpton, 1999). The zeolites (clinoptilolite), when saturated with mono – valet nutrient cations, such as NH_4^+ and K^+ , have been reported to increase the solubility of phosphate rock (Gholizadeh, 2008). The mechanism proposed to account for the solubilization of phosphate rock is exchange – induced dissolution. It is proposed that the plant uptake of NH_4^+ or K^+ frees exchanges sites which are occupied by ca^{2+}, lowering the soil solution ca^{2+} concentration and inducing further dissolution of phosphate rock however, phosphate rock dissolution may also be the result of H^+ generation through nitrification or H^+ efflux.

The effect of petroleum contamination on dry biomass of the studied plant show that the dry biomass of agropyron was higher than sunflower. Because of agropyron is a kind of grasses plant. Grass root system has maximum root surface area (Aprill and sims, 1990). Grasses have been used for germination of contaminated soil due to their fibrous root systems with extensive surface area for microbial colonization (Adam et al., 2002).Various plants have been identified for their potential to tolerate of contaminated sites with petroleum hydrocarbons. In the majority of studies grasses and legume have been singled out for their potential in this regard (Reilley et al., 1996). Native plants are better in terms of growth and reproduction under environmental stress than other plants (yoon et al., 2006). Furthermore, it seems that seed resistance to phytotoxic properties of oils is mainly attributed to the structure of the cell wall (Terje, 1984).

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Effect of Cadmium levels on growth and water relations of Spinach at three soil moisture regimes

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Abstract
Cadmium (Cd) are highly toxic heavy metals. Soil factors such as pH, cation exchange capacity, $CaCO_3$ content and moisture have a significant influence on Cd uptake by plants. Moreover, soil moisture deficient has a great impact on crop yield in Iran. Therefore, the effect of three moisture regimes (2, 4, and 7 days irrigation interval) and six Cd rates (0, 5, 10, 20, 40, 80 mg/kg as $CdSO_4$) on the growth and water relation of spinach was studied in greenhouse experiments. Treatments were arranged in a factorial manner in completely randomized design with three replicates. Leaf and stem dry weights of both crops were reduced significantly with a rise in Cd levels. Leaf water potential of spinach was not affected by Cd addition. However, stomatal resistance increased and transpiration decreased with increasing Cd rates. In the present study, stem was more tolerance to Cd than leaf. However, soil moisture deficit had a more depressing effect on the stem than on the leaf growth.

Keywords: cadmium, moisture regimes, Spinach, water relations.

Introduction

Heavy metals occur naturally in soils as trace elements. High concentrations are confined primarily to certain minerals usually present in forms which are not easily available but maybe sensitive to higher concentrations. The release of heavy metals in biologically available forms, as a results of human activity, may damage or alter both natural and man made ecosystems .The application of sewage sludge or smelter flue-dust or industrial emission can be contaminated soil with heavy metals. Elevated heavy metals concentration in soil can be negative effect on crop growth. Vegetables take up Hg, Pb, Cd, Zn, and accumulate them in the edible and inedible parts with various concentrations. The intake of edible parts of vegetables is an important path for heavy metals in the soil to harm human health (Zheng et al., 2007). Cadmium is of concern to agronomists because it is adsorbed in excessive amounts by a number of edible crops (Bingham et al., 1976).

Cadmium is a non-essential metals and a powerful enzyme inhibitor (Lockwood, 1976).cadmium, a trace metal of current intense environmental concern, occurs in ores used in the production of phosphorus fertilizer (Lockwood,1976).

soil are subjected to drying and rewetting cycle in the field, depending on climate, topography and exposure. This enhances temporal variability of soil chemical properties, e. g. ion equlibria, and influences decomposition and oxidation processes, which could interact with the uptake of elements by plants. Alternative wet and dry condition would also enhance formation of less hydrated and more crystalline compound of lower solubility.

Material and methods

Surface soil (0-30 cm) was collected. The name of this soil in soil taxonomy is "Fine, mixed (calcareous) mesic, Typic Calcixerepts ". The collected soil were air dried, crushed to pass through 2-mm sieve and Soil texture was determine by Hydromerter ,organic matter with oxidation with chromic acid and titration by fero ammonium sulfate , soil pH of saturated paste with a pH meter capability of electrical conductivity of saturated paste with electrical conductivity meter total nitrogen with kejeldal method, cation exchangeable capacity with sodium acetate calcium carbonate ,available phosphorous ,trace element was extracted with DTPA and determined by atomic absorption specronoc.Table1 shows physicochemical properties of the soil.

Table1: Properties of the soils used in the greenhouse experiment

Property	Range
Sand (%)	14.10
Silt(%)	45.50
Clay (%)	40.40
FC(%v/v)	29.70
PWP(%v/v)	13.04
Bulk density (g cm^{-3})	1.418
OM (%)	1.18
TN(%)	0.038
pH	7.84
EC (ds m^{-1})	0.495
CCE(%)	40.10
P soluble in Sodium Bicarbonate(mg Kg^{-1})	13.5
Cu (extracted with DTPA (mg Kg^{-1}))	1.83
Zn (extracted with DTPA (mg Kg^{-1}))	0.94
Fe (extracted with DTPA (mg Kg^{-1}))	6.8
Mn (extracted with DTPA (mg Kg^{-1}))	9.4
Cd (extracted with DTPA (mg Kg^{-1}))	0.27

Treatments were arranged in a factorial manner in completely randomized design with three replicates. 216 kg lots of soil were treated with Cd SO4 at rates equivalent to 0, 5, 10, 20, 40, 80 mg Cd kg-1 soil, and uniform application of N-P fertilizer (urea, KH2PO4) and sulfate of Zn, Cu, and Fe (Fe EDDHA) was made to each soil. According the test sample, the soil did not need any more K fertilizer. The treated soil was mixed thoroughly, divided among 54 pots (4 kg). 15 seed of spinach *(Spinosa oleracea)* were planted in each pot then given deionized water to field capacity and weighted daily for retaining the moisture in field capacity. Three weeks after emergence of the seedling, plants were decreased to 5 plants in each pot, and treated 3 moisture regime (2, 4 and 7 days irrigation interval). Evapotranspiration determined with difference between weighted before and after each interval irrigation and estimated volume of irrigation water for next period. For determination of evaporation, same pot without plant put between planted pot and evaporation estimated in each irrigation interval separately. Transpiration was calculated from differential evapotranspiration and evaporation. Average of soil moisture before irrigation was determined and then soil matric potential calculated by moisturing characteristic of soil curve (table2).

Table2: Minimum of soil moisture, decreasing of available water, minimum of Matric potential in interval irrigations

Interval irrigations	Minimum of soil (v/v) moisture	decreasing of available water	Minimum of Matric potential (Bar)
2	0.207	54	-1.63
4	0.176	72	-2.98
7	0.149	88	-6.83

equivalent of curve was:

$$\theta = 0.135 + \frac{0.300}{[1 + (5.62*10^{-5}h)^{0.62}]^{688}}$$

After 10 weeks leaf potential of spinach was determined by pursuer bomb. Stomatal Resistance determined. After harvesting leaf area determined with Leaf Area Meter, then leaf and stem separated and washed with deionized water, oven- dry weighted were determined. Data analyzed by MSTAT-C and SPSS software.

Results and discussion

Leaf and stem weight showed a significant negative response to Cd addition (Fig.1). The highest doses of Cd applied (80 mg kg^{-1}) decreased the yield significantly to about 54 and 34 percent for leaf and stem respectively. It may be attributed to the decrease essential nutrient at higher doses (Khan and Khan, 1983). Results showed that dry weight reduced with increasing interval irrigation. Table 3 show with increasing in interval irrigation decreased tolerance to cadmium in leaf and stem. Based on these results, yield decrements of 50% for leaf and stem deceased with Cd and interval irrigations.

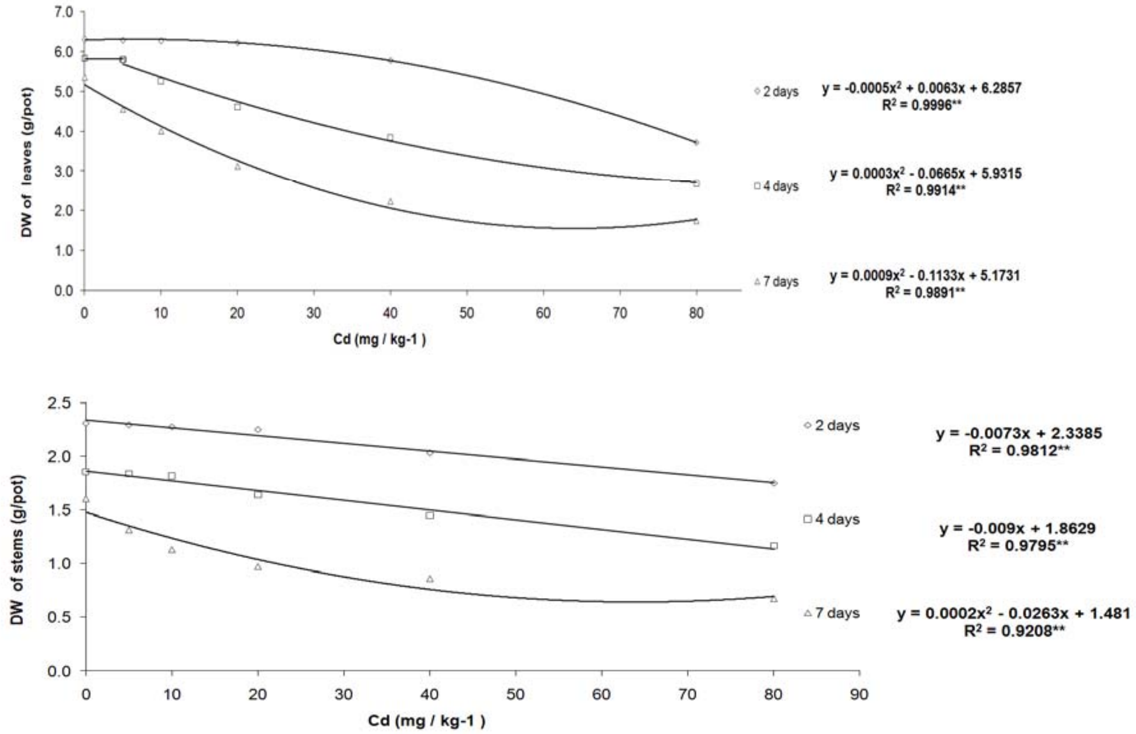

Fig.1. Effect of Cadmium levels and soil moisture regimes on dry weights (DW) of Spinach leaves and stems

Table 3: yield decrements of 50% for leaf and stem

interval irrigation (days)	stem	leaf
2	172	116
4	104	68
7	32	27

Stem was more tolerance to Cd than leaf. There was not any visual symptom like chlorosis in plant. The only effect was growth decreasing.Cadmium is a one essential heavy metal and a powerful enzyme inhibitor (Lockwood, 1976).

Transpiration rate certainly has an impact on plant heavy metal uptake. Heavy metals uptake has been examined for several crops and under various levels of soil or growth solution concentration (Grifferty and Barrington, 2000). Water stress and Cadmium were found to affect significantly plant transpiration. The highest cadmium decreased 17 percent of plant transpiration. In others, Cd toxicity appeared to induced reduced transpiration (haag-Kerwer et al., 1999; Paivok, 2002;Vaselove et al.,2003; Menon et al., 2005).Cd interfere with closure stomatal then affected on transpiration (Pearson and Kirkham, 1981).

Increasing Cd levels had a significantly positive effect on stomatal resistance but these effects appeared from 20 mg Cd/ kg^{-1} and up (Fig.2). Increasing stomatal resistance was 31 percent at the highest range of Cd. Effect of cadmium on decreasing stomatal conduct of cucumber reported by Burzynski and Klobus (2004).

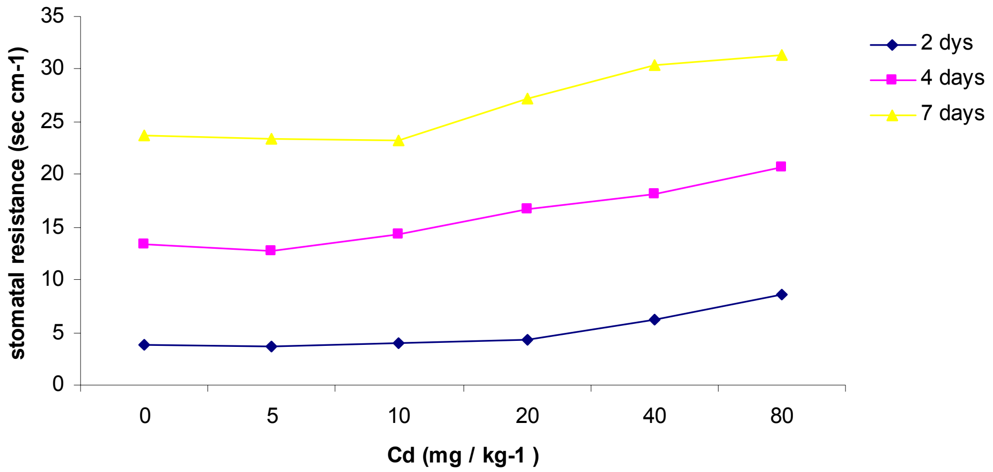

Fig. 2: Effect of Cadmium levels and interval irrigation on spinach stomatal resistance (sec cm^{-1})

Cd treatment did not effect on the water potential although the stomatal resistance increased. Relation water potential and stomatal conduct can be different in plant species and environmental conditions. Stomatal resistance may be as a controller of the water potential. The level of heavy metal translocated to chloroplast is estimated to be only 1% however, a much variation between species which needs to be investigation. Both permanent stomatal closure and increasing ethylene may be responsible for senescence induction by cadmium (Fuhrer, 1988). In an effort to estimate the water stress experienced by crop plants, Idso *et al.* (1981) suggested a 'crop water stress index'derived from the increase in average canopy temperature induced by stomatal closure in water stressed crops.Because the plant water potential (Ψ) is an adequate expression of plant water balance at any time (Karamanos, 2003), it could be a useful and objective indicator of the tensity of water stress in plant. Thus, initially, the stomatal closure seemed neither due to a general leaf turgor loss nor to a change in bulk leaf ABA levels. This early increase of stomatal resistance may be due to either a small pool of active ABA not detectable when bulk leaf ABA is analyzed or other metabolic changes. In addition to ABA, other compounds may be involved in stomatal closure (Mansfield ,1986; Poschenrieder,1989 and et al). Provided that toxic amounts of Cd reach to the guard cells, an interference of Cd with both K and Ca seems very likely (Poschenrieder,1989 and et al.).

Leaf area showed a decreasing significant effect to Cd addition (table 4).

Table 4: Effect of Cadmium levels and interval irrigation on spinach leaf Area (cm^3/pot)

Cadmium levels (mg kg^{-1})	interval irrigation (days) 2	4	7
0	2029	1514	1020
5	1997	1436	877
10	1954	1373	743
20	1851	1221	586
40	1482	1004	418
80	1376	860	335
Mean	1782	1235	663

	LSD(0.05)
Cadmium	30
interval irrigation	30
interval Cadmium× irrigation	51

Interaction between water stress and Cd addition was positive and the least of leaf area were revealed at Cd_6 and 7 days interval irrigation and it was 57 percent compare to Cd_0 and 2 days interval irrigation. Overnell (1975) reported that cadmium reduced the concentration of ATP and chlorophyll in many species and decreased oxygen production. Cadmium may inhibit leaf cell expansion growth through alterations of the plant water balance (Poschenrieder, 1989 and et al.)
Linear correlation analysis highly significant between DTPA-extractable and Cd concentration of leave and stems and in these studies , highly significant correlation were found between total cadmium added to soil and cadmium soil DTPA-extractable. Then we can say with DTPA-extractable, we can predict cd concentration in spinach. Shuman (1986) mentioned that, the better extraction is DTPA since it produced correlation between the plant metal content and DTPA extractable metal. DTPA was able to extracts more than 90 % Cd, corresponding total DTPA extractable metals from the soil. Leaf and stem weight and leaf area showed a significant negative response to Cd addition. Based on these results, yield decrements of 50% for leaf and stem deceased with Cd and interval irrigations. Stem was more tolerance to Cd than leaf. Water stress and Cadmium were found to affect significantly plant transpiration. The highest cadmium decreased 17 percent of plant transpiration. Increasing stomatal resistance was 31 percent at the highest range of Cd. Cd treatment did not affect the water potential although the stomatal resistance increased. Stomatal resistance may be as a controller of the water potential.

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Study on nitrification potential following sewage sludge and manures application

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Abstract:
Mineral forms of nitrogen (N) are unstable in soils and nitrate (NO_3) leaching result in contamination of soils and ground waters. Nitrification from organic residues including sewage sludge as well as manures is important processes which influence the amount of soluble N and other nutrients bioavailibity in soils. This study was done to evaluation of organic residues effect on growth, N content in rhizosphere of corn plants in greenhouse experiment using sewage sludge (SS), poultry (Pl), cattle (Ct) and sheep (Sh) manures. The results showed plants acquired significantly more N, P, K, Ca, Mg, Fe, Mn, Zn and Cu in SS treatment than other manures. The highest mineralized nitrogen in rhizosphere and non-rhizosphere soil were achieved in Pl (214.8 mg kg^{-1}) and SS (227.5 mg kg^{-1}), respectively. Mineralized N in particular NO_3 was observed at high concentration in rhizosphere compared to non-rhizosphere soil, which indicate higher microbial activity including nitrificators and high mineralization processes in rhizosphere. Net nitrogen mineralization (N_m) in rhizosphere was higher than that in non-rhizosphere which the microbial population in corn rhizosphere was 3.7 times higher than non-rhizosphere soil. Treatments applied with SS showed the highest microbial activity compared to other organic residues. At rhizosphere, net nitrogen mineralization was occurred in Pl and SS treatments whereas Ct and Sh applied soil showed nitrogen net immobilization. Except for SS, all applied residues showed net immobilization in non-rhizosphere soil. The highest total mineralized N were as fallows in treatments: Pl> SS> Sh> Ct. Therefore, the results showed that application of sewage sludge and poultry manure may cause soil and water resource pollution trough NO_3 increment.
Keywords: sewage sludge, manures, nitrate, rhizosphere

Introduction

Organic farming systems mainly rely on organic fertilizers such as compost, slurry, and animal and green manure to maintain nitrogen (N) nutrition of the crops. Unlike in most mineral fertilizers, the N contained in organic fertilizers only gradually becomes available after application, through a decomposition process. It is one of the key challenges of organic farming to match N release from organic fertilizers and crop demand for N. However, predicting N release from organic fertilizers is complicated because it is influenced by the physico-chemical properties of the added organic material, abiotic factors like pH, temperature, water and clay content of the soil, and the characteristics of the soil microflora involved in the decomposition process (Net et al., 2010). On the other hand, addition of organic waste such as manures and sewage sludge has been proposed as one method of maintaining levels of organic matter in agricultural soils (Garcia et al., 1992; Boyle and Paul, 1989). In addition, land application of appropriately treated organic wastes has frequently been demonstrated to be a safe and effective means for recovery of plant nutrients (particularly N and P) while simultaneously improving soil physical and microbiological properties (Ayuso et al., 1996; Hernandez et al., 1991; Parr et al., 1989). However, the application rate of sewage sludge to land must be determined on the basis of crop N requirement to avoid potential hazards associated with excessive NO3- in soil (Navarro Pedreno et al., 1996; Paul and Clark, 1989).

Assessment of the relative amounts of N that will become available when sewage sludges are applied to soils will allow use of these materials in a more efficient and economical way, and minimize potential NO_3 losses to groundwater (Hernandez et al., 2002). Many N availability indices, most of them based on methods developed for determining N availability in soils, have been proposed (Bremner, 1965; Fox and Piekielek, 1978; Stanford, 1982). However, procedures involving the determination of N mineralised during controlled incubation are, generally, considered the most satisfactory methods available for evaluating N availability (Stanford et al., 1974). Mineralization of the organic N in sludges is a complex process affected by several factors such as soil type, soil pH, temperature, moisture and rate and type of sludge. Different authors have indicated that mineralization kinetics vary according to the type of sewage sludge stabilization process, aerobic or anaerobic (Ryan et al., 1973; Epstein et al., 1978; Hattori and Mukai, 1986).

Serna and Pomares (1992) in a non-leached incubation experiment observed that aerobically treated sewage sludges gave higher mineralization rates than anaerobically treated wastes.

It is widely accepted that the reactions of N mineralization generally follow a first-order kinetic (Stevenson, 1965; Stanford and Smith, 1972; Serna and Pomares, 1992). In an incubation experiment, Stanford and Smith (1972) estimated the N mineralization potential (N_0) and rate constant (k) of the kinetic equation to be first order, using a model based on a logarithmic function. This model was improved by Smith et al. (1980) who found that a non-linear least-squares equation gave more accurate estimations of k and N_0.

Different studies have been conducted on the influence of sludge characteristics on potential N mineralization (N0) (Magdoff and Chromec, 1977; Epstein et al., 1978; Serna and Pomares, 1992) but studies on the influence of soil type on potential sludge N mineralization in freshly amended soils are scarce. Information on N mineralization in various soil-organic systems is needed for a better prediction of crop N requirements.

The objectives of this study were: (i) to determine the influence of different manure type and sludge application on the extent of N mineralization in a calcareous soil freshly amended with sludge, sheep, cattle and poultry manures.

Materials and Methods

This study was performed on a silty clay loam soil, collected from Nazlu region (38º11'N, 44º46'E), West Azerbaijan province located in North-West of Iran. For this purpose surface layer (0–15 cm) samples of calcareous soil were collected and transferred to greenhouse. Some physical and chemical properties of soil have shown in Table 1. The sewage sludge (SS) was prepared by sewage refinery unit of Maragheh city and manures including sheep (Sh), cattle (Ct), poultry (Pl) manures kindly were supplied by Department of Animal Science, Urmia University (Table 2). The sludge was added and mixed to experimental pots (containing 3 kg soil) at the rates of 20g sludge kg^{-1}soil. Amended and control soils were placed at greenhouse condition. The soil moisture content was maintained around FC. Mineralised NO_3^- and NH_4^+ were determined at the starting of the experiment and at the end of experiment.

Table1. Physico-chemical properties of soil used in experiment

CCE	OC	NH_4^+	NO_3^-	K	P	N	EC	pH	Texture	clay	silt	sand
g kg^{-1}	g kg^{-1}	mg kg^{-1}				%	dS m^{-1}			g kg^{-1}		
110	6.7	87.5	70	284	16.2	0.11	1.27	7.57	SiCL	300	520	180

CCE: Calcium carbonate equivalent
OC: Organic carbon
EC: Electrical conductivity
SiCL: Silty caly loam

Table2. Some characteristics of applied manures and sewage sludge

Manure type	pH	EC	N	P	K	OC	C:N	Fe	Mn	Cu	Zn
		dSm^{-1}	%					mg kg^{-1}			
Sheep man.	7.46	29.8	2	0.89	3.07	52	26	949	45	11	43
Cattle man.	7.73	13.6	2.4	1.02	0.81	61	25.4	1611	72	4	54
Poultry man.	5.14	11.8	2.9	1.39	0.77	36	12.4	955	209	36	298
Sludge	7.3	6.7	4.2	1.71	0.26	58	13.8	1447	207	83	559

EC: Electrical conductivity
OC: Organic carbon

Each pot was planted with 10 corn (*Zea mays* L.) seeds, which finally thinned to 4 seedlings per pots. ad newly developed two true leaves and on average, based on a sub-sample of 68 seedlings, contained 1.24 mg N. Corn has relatively high capacity for N uptake and was thus used to remove mineral N from the pot so as to prevent N losses by denitrification or leaching and to transform the N into a form that can be readily analyzed at the end of the experiment. In the greenhouse, all pots were arranged in a randomized block design, blocked by pot replicate. All pots were frequently weighed and gravimetric water content adjusted to 170 mg g^{-1} dry soil (approximately 70% of water holding capacity). No drainage water from the pots was observed as their watering completely controlled at short times intervals. The duration of the experiment was 63 days from planting. During this period, the average temperature in the greenhouse was 27°C (min, 23.1°C and max, 32.2°C). At the end of experiment shoot and roots of corn plants harvested and washed with deionized water, then the dry weight and amount of N and other nutrients were determined with standard methods. Soil samples from rhizosphere and non-rhizosphere of corn plants collected by method of Shen et al. (2003). Total N, NO_3^- , NH_4^+, P, K, OC, pH and EC were assessed in soil samples. To evaluate the net N mineralization the model proposed by Net et al. (2010) was used. The proposed equation is the following:

$N_m = N_{PLANT} + Nmin_{END} - Nmin_{START}$,

where N_m is the amount of N mineralised associated with sludge during experiment time; N_{PLANT} is plant N uptake which calculated as the difference between total plant N content at the end and start of the pot experiment. $Nmin_{END}$ and $Nmin_{START}$ are the soil mineral N content at the end and start of pot experiment, respectively.

Data were analysed using the analysis of variance for a completely random design. Comparisons between means were made using Duncan's multiple range tests.

Result and Discussion

The results showed plants acquired significantly more N, P, K, Ca, Mg, Fe, Mn, Zn and Cu in SS treatment than other manures. The most immediate impact of organic waste application is on the availability of nitrogen to the subsequent crop, as a consequence of mineralization–immobilization processes (Hadas et al., 2004). Other authors have tested the release of N from organic wastes mineralization, to ryegrass, maize and wheat (Evers, 2002; Vasconcelos et al., 1999). Because 50−90% of the N in sludge is organic (Sommers, 1977) it is necessary to know the rate of N mineralization in order to predict N availability during a crop season. In a residue-amended soil the total amount of available N can be described as the sum of the inorganic N and mineralized organic N in the soil and the added residue. This amount of available N must be considered as potentially available N because N may be removed from the available N pool by denitrification, leaching or ammonia volatilization. The contents of different mineral N, total N mineralized, net mineralization or immobilization at rhizospheric soil are given in Table 3. The highest and lowest mineralized nitrogen in rhizosphere soil were observed in Pl (214.8 mg kg^{-1}) and Ct (134 mg kg^{-1}). At rhizosphere of SS and Pl amended treatments net N mineralization was occurred, however net N immobilization was seen in Sh and Ct amended soils (Table 3). No significant difference was seen regarding $N\text{-}NH_4^+$ in rhizospheric soil. Nitrate was higher in Pl added soil compared to other organic amended soils.

Mineralized N in particular NO_3 was observed at high concentration in rhizosphere compared to non-rhizosphere soil, which indicate higher microbial activity including nitrificators and high mineralization processes in rhizosphere (Tables 3, 4).

Table 3. Comparison of different mineral N, total N mineralized, net mineralization or immobilization at rhizospheric soil

Manure type	N_{plant}	$N\text{-}NH_4$	$N\text{-}NO_3$	Mineralized N	TN_{min}	N_m or N_i
	mg kg^{-1}					
Sludge	101.2	48.2	32.1	80.5	181.7	+24.2
Sheep man.	52.5	49.6	32.7	82.2	134.8	-22.8
Cattle man.	57.5	49	27.4	76.4	134	-23.6
Poultry man.	83.8	47.6	83.5	131.1	214.9	+57.4
Control	68.3	48.5	29.7	78.2	146.5	-11
$LSD_{0.05}$	27.1	ns	37.1	41.2	47.2	18

N_{plant}: N absorbed by plants (N uptake)
TN_{min}: Total N mineralized (sums of column 1 and column 4)
N_m/N_i: Net mineralization or net immobilization (positive and negative data show N_m and N_i, respectively)
All data corrected for inorganic N initially added in manures and sludge

At non-rhizosphere the mineralized N was higher than that of rhizosphere in all organic manures treatments. The highest and lowest mineralized N belonged to SS (227.5 mg kg^{-1}) and Ct (107 mg kg^{-1}), respectively. SS applied treatments showed higher amount of N-NO_3 compared to other manures (Table 4). Except for SS, all applied residues showed net immobilization in non-rhizosphere soil. In the other hand at non-rhizospher, nitrification exceeds ammonification which may indicate higher activity of nitrificators.

Table. Comparison of different mineral N, total N mineralized, net mineralization or immobilization at non-rhizosphere soil samples

Manure type	N-NH_4	N-NO_3	Mineralized N	TN_{min}	N_m or N_i
			mg kg^{-1}		
Sludge	47.8	179.7	227.5	227.5	+70
Sheep man.	51.3	59.5	110.8	110.8	-46.3
Cattle man.	43.1	64.7	107.3	107.3	-50.2
Poultry man.	35.2	102.7	137.9	137.9	-19.6
Control	33.8	33.8	67.7	67.7	-89.9
$LSD_{0.05}$	ns	37.1	41.1	47.2	18.1

TN_{min}: Total N mineralized
N_m/N_i: Net mineralization or net immobilization (positive and negative data show N_m and N_i, respectively)
All data corrected for inorganic N initially added in manures and sludge

Net nitrogen mineralization (N_m) in rhizosphere was higher than that in non-rhizosphere which the microbial population in corn rhizosphere was 3.7 times higher than non-rhizosphere soil. Treatments applied with SS showed the highest microbial activity compared to other organic residues (data not shown). At rhizosphere, net nitrogen mineralization was occurred in Pl and SS treatments whereas Ct and Sh applied soil showed nitrogen net immobilization. The highest total mineralized N were as fallows in treatments: Pl> SS> Sh> Ct. Therefore, the results showed that application of sewage sludge and poultry manure may cause soil and water resource pollution trough NO_3 increment.

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Sunflower Plant Capability for Phytoremediation of Ni Polluted Soils in Presence of Plant Growth Promoting Bacteria (PGPB)

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Abstract
This experiment was conducted to evaluate the capability of sunflower for phytoremediation of Ni polluted soils in presence of plant growth promoting bacteria (PGPB). The experimental treatments were arranged as factorial based on a completely randomized design with three replications. Each plot consisted of a pot with different concentrations of Ni [0(control), 150, 300 and 450 mg /kg]. The bacterial treatments consisted of control (no bacteria), Bacillus mycoides, Micrococcus roseus and Bacillus mycoides+ Micrococcus roseus. The results of the experiment showed a significant ($P<0.01$) interaction effect of Ni concentration × bacteria inoculation on shoot dry matter, total dry matter, Ni concentration in both shoot and root zones of sunflower plants. The Ni concentration up to 300 mg kg^{-1} had no significant decreasing effect on shoot and total dry matter in sunflower, however, when Ni concentration in the soil reached 450 mg/kg, this parameter significantly decreased compared to control. The highest shoot and total dry matter was obtain when the plants inoculated with bacterial treatments (B.mycoides+M.roseus) and the lowest amounts of dry matter was observed in control treatment while the Ni concentration in the soil was 450mg/kg. The highest translocation factor was observed in Ni control treatment when plant inoculated with bacteria (B.mycoides+M.roseus). The highest Ni concentration in plant was obtained at 450 mg/kg when plant inoculated with B.mycoides+M.roseus. The results of the experiment confirmed the sunflower capability to absorb more Ni from the soil when the plant growth is enhanced by PGPB application.
Keywords: Sunflower, phytoremediation, PGPB, Ni

Introduction
Soil contamination by heavy metals is one of the major environmental problems in world, posing significant risks to public health and ecosystems (Sheng et al, 2008). Most plants are sensitive to excessive accumulation of heavy metals in rhizosphere. When the heavy metal ions, accumulate at an elevated levels in the soil, are excessively absorbed by roots and translocated to shoot, leading to impaired metabolism and reduced growth (Bingham et al., 1986). Also high concentrations of these metals in soils result in decreased soil microbial activity and soil fertility, and yield losses (McGrath et al., 1995). Phytoremediation is the use of both green plants and their associated rhizosphere microbes to remove pollutants from the environment or to render them harmless (Salt et al., 1998). It is a low cost and eco-friendly means, which can be applied for the cleanup of contaminated soils (Salt et al., 1998; Gardea-Torresdey et al., 2005). promotion of plant growth under stress conditions is critical to the optimum performance of phytoremediation of metal accumulating crops (Belimov et al, 2004). Rhizosphere microorganisms, which are closely associated with roots, have been termed plant growth promoting rhizobacteria (PGPR) (Glick, 1995). PGPRs by facilitating the uptake of certain nutrients (Cakmakci et al. 2006), induction of resistance in plants against fungal, bacterial and viral diseases (Khan et al. 2002), synthesizing phytohormones precursors (Ahmad et al. 2008), vitamins, enzymes, siderophores, antibiotics (Glick, 2001) and inhibiting ethylene synthesis improve plant stress tolerance to drought, salinity and metal toxicity, leading thereby to increased plant growth. The objective of this research was to investigate the effectiveness of Helianthus annuus L. for the phytoremediation of Ni in presence of plant growth promoting bacteria.

Material and methods
A greenhouse pot culture experiment was performed to study the effect of Ni in presence of plant growth promoting bacteria on sunflower growth characteristics and itscapability for Ni uptake.The experimental treatments were arranged as factorial based on a completely randomized design with three replications. Treatments consisted of four levels of Ni [0(control), 150, 300 and 450 mg Ni kg^{-1} soil as nickel chloride] and bacteria inoculation [Control (no bacteria), Bacillusmycoides, Micrococcus roseus and co-inoculation (Bacillus mycoides+ Micrococcus roseus)]. For each pot,

3.5 kg oven dried soil (48 h at 70° c) was weighted and contaminated according to treatments via spraying uniformly. Seeds were surface sterilized by soaking for 10 min in 1.5% sodium hypochlorite and then thoroughly rinsed with sterile distilled water. Then inoculated seeds were sown and irrigated up to field capacity until the experiment was terminated. After 8 weeks, roots were carefully removed from the soil, gently shakened, and washed to eliminate any attached soil. In order to assess Ni concentration, the plant material was dried at 60◦C for 72 h and weighed half gram of dried plant tissue was digested in 20 ml diacid mixture of concentrated H2SO4:HClO4 (4:1) and the final volume made to 25 ml. Nickel in digested solution was determined using Atomic Absorption Spectrophotometry (Lindsay and Norvell, 1978). In order to assess amount of metal transfer from root to shoot, translocation factor was determined by dividing metal concentration at shoot by its concentration at root (Marchiol et al., 2004). Ni uptake values in both shoot and total plant biomass, was calculated by: biomass × Ni concentration. The analysis of variance of the data was done by SAS, 9.1 software. The means were separated by Duncan's multiple range test (DMRT) at the 5% probability level.

Results and Discussion

Shoot and Total Plant Biomass

Shoot and total plant biomass decreased by Ni application and increased with bacterial treatments (Fig 1 and 2). The highest shoot and total plant biomass was obtained at 150 mg kg^{-1} of Ni with M. roseus inoculant. Also the lowest values for these parameters were observed at 450 mg kg-1 of Ni and no bacteria inoculation. Ahmad et al (2011) that Nickel stress significantly reduced sunflower shoot dry weight. Free-living bacteria as well as symbiotic bacteria can enhance plant growth by providing bio-available phosphorus, nitrogen and sequestering trace elements. The availability of siderophores, plant hormones (auxins, cytokinins and gibberellins), and lowered plant ethylene levels are the other means by which PGPR stimulate the plant growth (Kumer et al., 2009). It has been reported that application of rhizobacterium (Bacillus subtilisSJ-101) in soils amended with nickel, significantly increased Brassica Juncea growth while protecting the plants from nickel toxicity (Zaidi et al. 2006). Similarly, Cadmium resistant Bacillus mycoides and Micrococcus roseus protected Amaranth (Amaranthus retroflexus L.) grown in soils supplemented with Cadmium (Motesharezadeh et al. 2008).

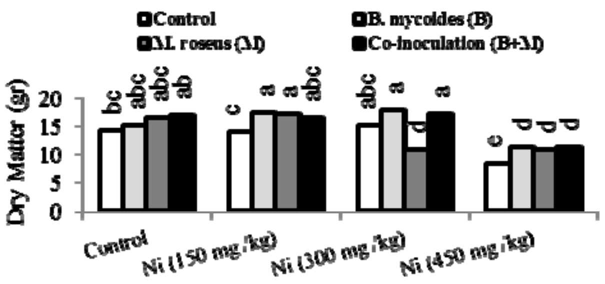

Fig 1. interaction effects of Ni and PGPBs on shoot biomass

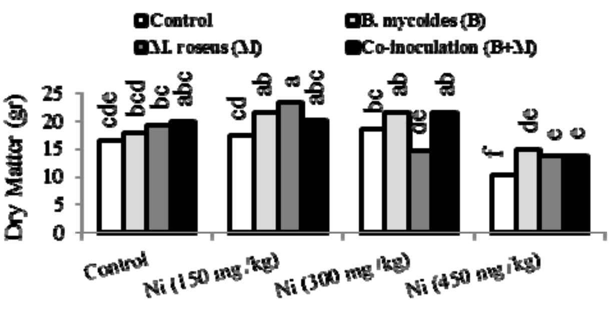

Fig 2. interaction effects of ni and PGPBs on total plant biomass

Root and Shoot Ni Concentration

Ni concentration in Root and shoot was significantly affected by different treatments. Root and shoot Ni concentration consistently increased as Ni concentration increased in soil. Increment of Ni in plants aerial part in response to soil Ni content has been reported in literature (Ahmad et al, 2011). Results showed that bacterial inoculants, especially inoculation by B. mycoides decreased Ni concentration in both root and shoot compared to non-inoculated plants. The highest and lowest Ni concentrations in both root and shoot, were observed in non-inoculated plants sown in 450 mg kg^{-1} of Ni and inoculated plants by B. mycoides, in control (no Ni contamination) pots, respectively (Fig 3 and 4). The overall uptake of Ni by plants depends on the concentration of Ni, plant metabolism, acidity of soil solution and presence of other metals and organic matter composition (Chen et al. 2009). Abou-Shanab et al (2003) studied the effects of PGPB on nickel extraction by Alyssum murale and demonstrated that Ni uptake increased by PGPB application. They suggested that the bacteria facilitated the release of Ni from the non-available phase in the soil, thus enhancing the availability of Ni to A. murale.

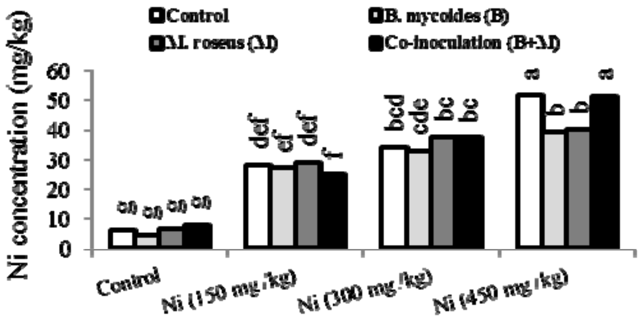

Fig 3. interaction effects of Ni and PGPBs on shoot Ni concentration

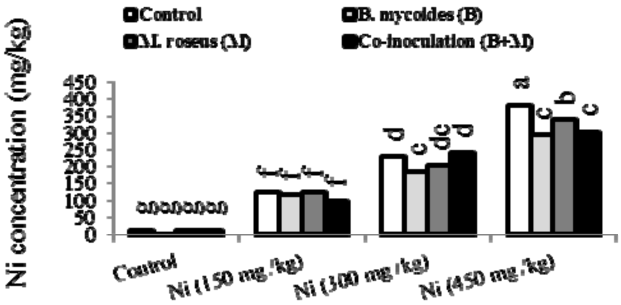

Fig 4. interaction effects of Ni and PGPBs on root Ni concentration

Translocation Factor (TF)

Ni translocation factor (TF) was significantly affected by the application of Ni, bacterial inoculation and interaction effect of Ni application× bacterial inoculation. It was observed that Ni translocation factor decreased consistently with Ni contamination increment in the soil. Generally, Ni translocation factor in inoculated plants, especially co-inoculation treatment (B. mycoides + M. roseus) was more than non-inoculated plants (Fig. 5). In this study, TF values was less than 1 and therefore sunflower plant can not be used for phytoremediation though "phytoextration" mechanism, although it can be used in phytoremediation process via "phytostabilization" mechanism. Phytostabilization is defined as immobilization of a contaminant in soil through absorption and accumulation by roots, adsorption onto roots or precipitation within the root zone of plants. In this case, prevention from expansion of contaminants by its stabilization in root zones. Motesharezadeh et al (2008) reported that sunflower plant can be used for phytoremediation of Cadmium contaminated soils through phytostabilization mechanism.

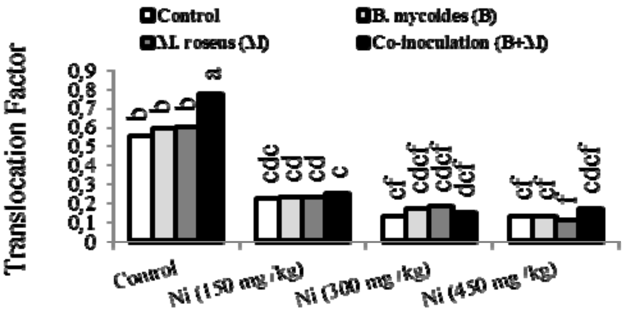

Fig 5. interaction effects of Ni and PGPBs on Ni translocation factor

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The Effect of Plant Growth Promoting Bacteria on Fe and Zn Absorption and Accumulation in Sunflower Plant under Ni Pollution Stress

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Abstract
This experiment was conducted to evaluate the sunflower capability to absorb Fe and Zn at different soil contamination levels of Ni and plant growth promoting bacteria application. The experiment was set up as factorial based on a completely randomized design with three replication. Each plot consisted of a pot with different concentrations of Ni [0(control), 150, 300 and 450 mg /kg]. The bacterial treatments consisted of control (no bacteria), Bacillus mycoides, Micrococcus roseus and Bacillus mycoides+ Micrococcus roseus.The results showed that as the Ni contamination in soil increased the concentration of this element in shoot and root increased while the Zn concentration in both organs followed a decreasing was observed in Bacillus mycoides treatment at soils with 450 mg/kg. The highest concentration of Zn in shoot and root was observed at Micrococcus roseus and Bacillus mycoides + Micrococcus roseus bacterial treatments. Application treatment of Bacillus mycoides caused the highest accumulation of Fe in root and shoot of sunflower. The results of this experiment indicated that the reactions of sunflower of absorb Fe and Zn in Ni contaminated soils will differ according to different bacterial treatments.
Keywords: Sunflower, Plant Growth Promoting Bacteria, Fe, Zn, Ni

Introduction
Biosphere Pollution by toxic metals has accelerated dramatically since the beginning of industrial revolution (Saghir Khan et al, 2009). It is well known that Ni has been classified among essential micronutrient and essential for plant growth in low concentrations (Welch, 1995). It remains associated with some metallo-enzymes, Nevertheless, beyond certain threshold concentration, it is toxic to plants (Pandey and Sharma, 2002). It has been reported that, excess Ni affects nutrient absorption by roots (Rahman,2005). Ahmad et al (2011) reported that application of Ni-stress decreased concentrations of micro- and macro-nutrients in leaf tissues of sunflower plants. Rhizosphere microorganisms, which are closely associated with roots, have been termed plant growth promoting rhizobacteria (PGPR) (Glick, 1995). PGPRs by facilitating the uptake of certain nutrients (Cakmakci., et al. 2006), induction of resistance in plants against fungal, bacterial and viral diseases (Khan et al. 2002), synthesizing phytohormones precursors (Ahmad et al. 2008), vitamins, enzymes, siderophores, antibiotics (Glick 2001) and inhibiting ethylene synthesis improve plant stress tolerance to drought, salinity and metal toxicity, leading thereby to increased plant growth. This experiment was carried out to study the effect of PGPBs application on Fe and Zn uptake by sunflower plant at Ni stress condition.

Material and methods
This experiment was carried out to evaluate Fe and Zn uptake by sunflower plant in Ni stress condition. Experimental treatments were arranged as factorial based on a completely randomized design with three replications. Treatments consisted of four levels of Ni [0(control), 150, 300 and 450 mg Ni kg-1 soil as nickel chloride] and bacteria inoculation [Control (no bacteria), Bacillus mycoides, Micrococcus roseus and co-inoculation (*Bacillus mycoides* + *Micrococcus roseus*)]. For each pot, 3.5 kg oven dried soil (48 h at 70° c) was weighted and contaminated according to treatments via spraying uniformly. Inoculated seeds were sown and irrigated up to field capacity until the experiment was terminated. After 8 weeks, to determination of Fe and Zn concentrations in root and shoot parts, the plant material was dried at 60◦C for 72 h and weighed half gram of dried plant tissue was digested in 20 ml diacid mixture of concentrated H_2SO_4:$HClO_4$ (4:1) and the final volume made to 25 ml. Fe and Zn in digested solution was determined using Atomic Absorption Spectrophotometry (Lindsay and Norvell, 1978). The analysis of variance of the data

was done by SAS, 9.1 software. The means were separated by Duncan's multiple range test (DMRT) at the 5% probability level.

Results and Discussion

Effect of Ni and PGPBs application on root Fe concentration and interaction effects of Ni and PGPBs application on shoot Fe concentration and root and shoot Zn concentration was significant. Higher Ni levels significantly decreased the concentrations of Fe in shoot and Zn in both shoot and root tissues. In contrast, Fe concentration in root increased consistently as Ni contamination level increased (Fig 2). According to Ahmad et al (2011), Ni stress decreased concentrations of micronutrients such as Fe and Zn in leaf tissues of sunflower plants. In addition, they reported that Ni stress severely affected root proliferation. Thus, reduced root development under high Ni application interfered with the uptake of different nutrients. Also Parida, *et al* (2003) reported that excess Ni levels in the soil decreased Zn contents in fenugreek plant tissues. It is thought that excess Ni^{2+} competitively inhibits the configuration of Fe^{2+}-ligand complexes, decreasing their mobility. For example, nicotianamine and citrate, which are the primary ligands responsible for the mobility of Fe in plants, can effectively chelate Ni^{2+} in plants (Saito et al. 2010). It has been reported that in several plant species, excess Ni inhibits the root to shoot translocation of Fe and causing overaccumulation of Fe in the root part (Yang et al. 1996). Furthermore, several Fe^{2+}-nicotianamine transporters in the Yellow Stripe-Like (YSL) family show Ni^{2+}-nicotianamine transport activity (Gendre et al. 2007), indicating that competition between Fe^{2+} and Ni^{2+} occurs within the membrane transport system.

The effect of bacterial inoculants on root and shoot Zn concentration did not follow any specific trend (Fig 4 and 5). Inoculation by *B. mycoides* significantly increased the Fe concentration in shoot tissue, however, significantly reduced root Fe concentration compared to control. Inoculation by *B. mycoides* significantly increased shoot Fe concentration to 300 mg kg-1 of Ni, but had not any positive effects at 450 mg kg-1 of Ni (Fig. 1). This may be due to adverse effects of Ni on bacterial population and activities. Supporting our findings, Motesharezadeh et al (2008) demonstrated that in sunflower and Amaranthus plants, Fe concentration in shoot increased when they were inoculated by *B. mycoides*.

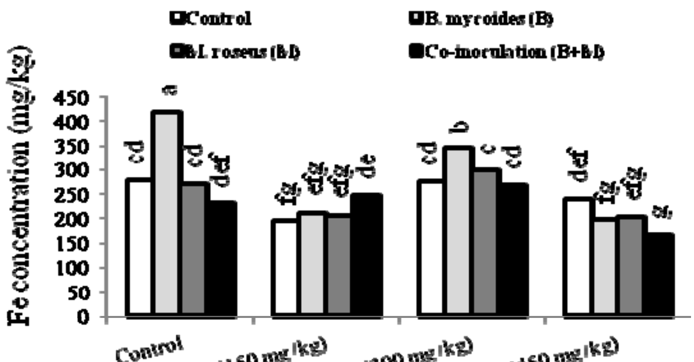

Fig 1. interaction effects of Ni stress and PGPBs on sunflower shoot Fe concentration

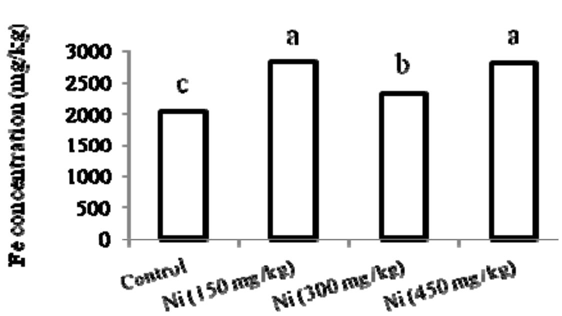

Fig 2. effect of Ni stress on sunflower root Fe concentration

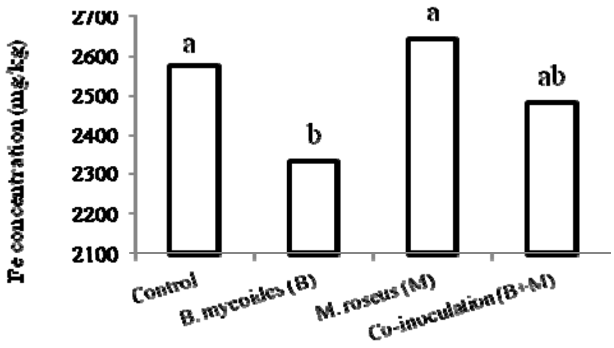

Fig 3. effect of PGPBs application on sunflower root Fe concentration

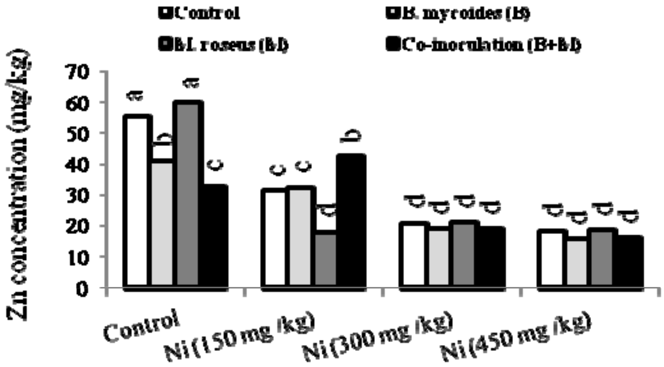

Fig 4. interaction effects of Ni stress and PGPBs on sunflower shoot Zn concentration

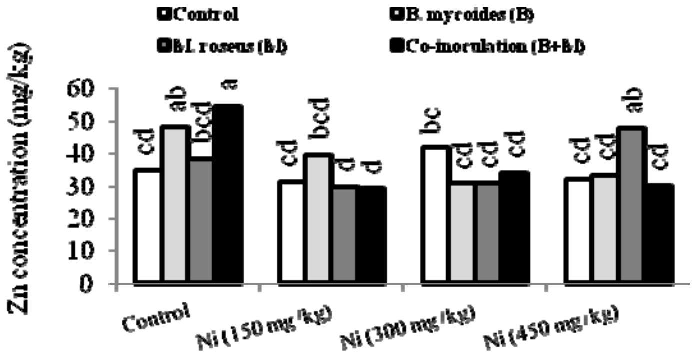

Fig 5. interaction effects of Ni stress and PGPBs on sunflower root Zn concentration

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Sorghum germination and early growth in presence of different Cd, Ni, Cu and Pb concentrations

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Abstract
This experiment was conducted to examine the effect of different concentrations of heavy metals (Cd, Ni, Pb and Cu) on germination and early growth of sorghum. The experiment was set up as a completely randomized design with three replications. Cd, Ni, Pb and Cu were applied at 15, 30, 45 and 60 ppm along with a control (no heavy metal). Presence of Cd, Ni and Cu significantly reduced shoot and root length and dry weight compared to control. However, Ni significantly increased root length and root/shoot ratio compared to control. The highest adverse effect of heavy metals of Cd, Ni and Cu on germination and early growth of sorghum was observed at 60 ppm concentration.
Kewwords: sorghum, germination, early growth, heavy metals, Cd, Ni, Pb, Cu

Introduction

As a result of human activities, such as mining and smelting, sewage irrigation, disposal of municipal wastes and so on, agricultural soil pollution by heavy metals has become increasingly serious throughout the world (Jun-yu et al, 2008). Growth inhibition is a general phenomenon associated with most of heavy metals (Reichman, 2002). The tolerance limits for HM toxicity are specific for every species and even for every variety of cultural plants (Jiang, Liu, 2000). Various authors (Jiang, Liu, 2000) have reported metal toxicity in plants. Jun-yu et al (2008) reported that rice seed germination and seedling growth indices significantly decreased at presence of Cadmium. Seed germination is the most resistant phase to heavy metals (Seregin and Kozhevnikova 2006). However, there have been many reports on the toxic effects of Ni on germination and seedling growth in plants. The germination of pigeonpea decreased by 20% in a 1.5 mM solution of 'and the germination percentage decreased in proportion to the concentration of Ni (Rao and Sresty 2000). Growth of *Zea mays* L. seedlings decreased with the increasing concentration of Ni as compared to control (Bhardwaj et al. 2007). Also, Bashmakov et al (2005) reported adverse effects of Cu and Pb on *Zea mays* L. seed germination and seedling growth.

Mterial and methods

to evaluate the influence of different concentrations of heavy metals include: Cd (Cadmium Nitrate), Cu (Copper Sulfate), Pb (Lead Nitrate) and Ni (Nickel Chloride) on seed germination and seedling growth of sorghum, an experiment was carried out on based on CRD with 3 replications. Experimental treatments included; Cd, Cu, Pb and Ni in four levels (15, 30, 45, 60 ppm) and Control (distilled water). sorghum seeds (50 seeds per Petri dish) after sterilization with Sodium hypochlorite were washed and placed on petri dishes. Petri dishes were filled with 8 ml of treatment solution levels and were placed inside the incubator with a temperature of 24 ± 2. After 7 days, root and shoot length, root and shoot dry weigh were measured. Seedling vigour index 1 and 2 were determined by the following formula (Reddy and Khan, 2001):

$$\text{Seedling Vigour Index 1} = \text{germination percentage} \times \text{seedling length}$$

$$\text{Seedling Vigour Index 2} = \text{germination percentage} \times \text{seedling dry weight}$$

The analysis of variance of the data was done by SAS, 9.1 software. The means compare were separated by Duncan's multiple range test (DMRT) at the 5% probability level.

Results

According to results of analysis of variance (Table 1), root and shoot length, root and shoot dry weight, root to shoot ratio and seedling vigour index 1 and 2 significantly affected by treatments. However, had not a significant effect on germination percentage.

Table 1. Analysis of variance for sorghum seed germination and seedling growth characteristics under heavy metals stress

S.O.V	df	RL	ShL	FGP(%)	RDW	SHDW	Sh/R	SVI1	SVI2
Treatment	16	40.6**	3.52**	33.4ns	0.0002**	0.00004**	0.77**	34.5**	0.00018**
Error	34	0.39	0.53	54.4	0.00002	0.000011	0.018	0.85	0.00007
CV (%)	-	12	11	9.5	10.9	6	19	10.4	11

- ns: Non-significant, *and**: Significant at 5% and 1% prabability levels, respectively
- RL (root length), Sh.L (Shoot Length), FGP (Final Germination Percentage), RDW (Root Dry Weigth), SHDW (Shoot Dry Weigth), Sh/R (Shoot to Root ratio), SVI1 (Seedling vigour Index 1) and SVI2 (Seedling Vigour Index 2)

Results showed that Pb at applicated levels had not adverse effects on root length and even stimulated root length at low concentrations. Highest root length was observed at 15 ppm of Pb. However, the lowest root length was related to 60 ppm of Cd and Cu (Fig. 1). Shoot length reduced by heavy metals application and lowest value of shoot length was related to Cu at 60 ppm (Fig. 1).

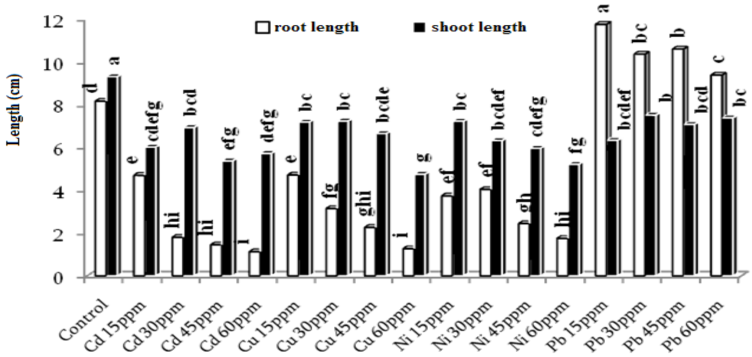

Figure 1. Effect of heavy metals on sorghum root and shoot length

Highest root and shoot dry weight were obtained at 30 ppm of Cu and 45 ppm of Pb, respectively. However, lowest root and shoot dry weight was observed at 60 ppm of Ni (Fig. 2). Cailin *et al* (2009) investigate the effect of Cd on wheat seedling growth and showed that Cd stress decreased root and shoot growth strongly. Also, it has been reported that, application of 100 and 200 µM Ni reduced the wheat shoot mass by 20% and 26% below the control level, respectively (Gajewska and Sklodowska 2008).

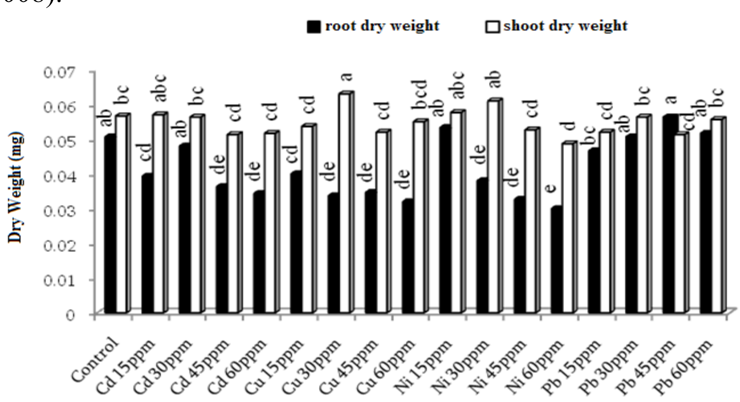

Figure 2. Effect of heavy metals on sorghum root and shoot dry weight

Root to shoot ratio in Pb treated seedlings, especially at 15 ppm, was higher than other treatments. Also, the lowest values of root to shoot ratio were observed at high levels of Cd and Cu (Fig. 3). Mahmood et al (2007) reported inhibitory effect of Cu on barley, wheat and rice seed germination, root length and root to shoot ratio.

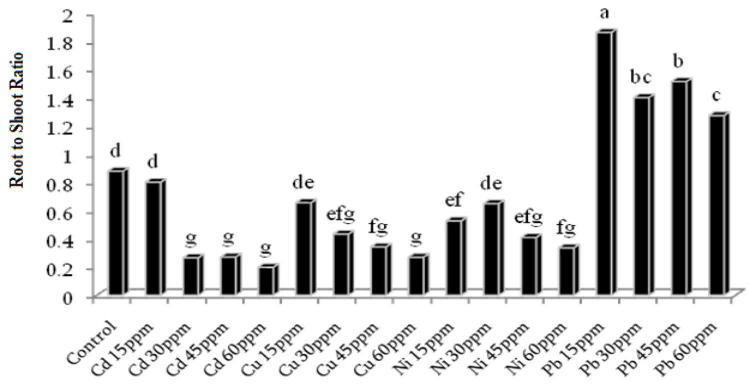

Figure 3. Effect of heavy metals on root to shoot ratio

Pb had not a significant effect on seedling vigour index 1, but, this parameter significantly reduced at high levels of Cd, Cu and Ni (Fig. 4). Jun-yu et al (2008) investigate the effect of Cd stress on seed germination and seedling growth of rice and reported that Germination rate, Germination index and Vigour index significantly reduced at stress condition. Growth of *Zea mays* L. seedlings decreased with the increasing concentration of Ni as compared to control (Bhardwaj et al. 2007).

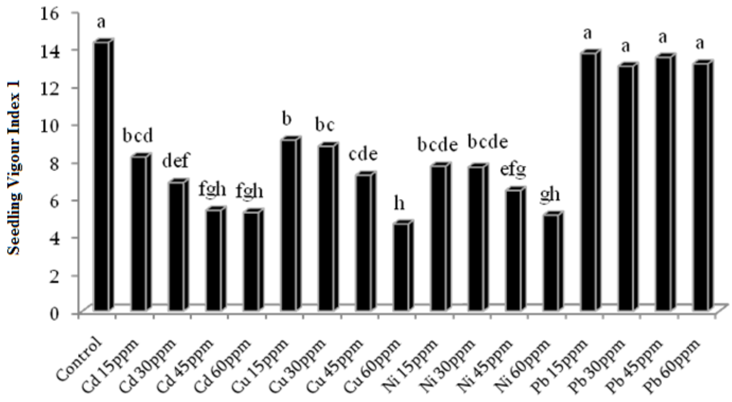

Figure 4. Effect of heavy metals on seedling vigour index 1

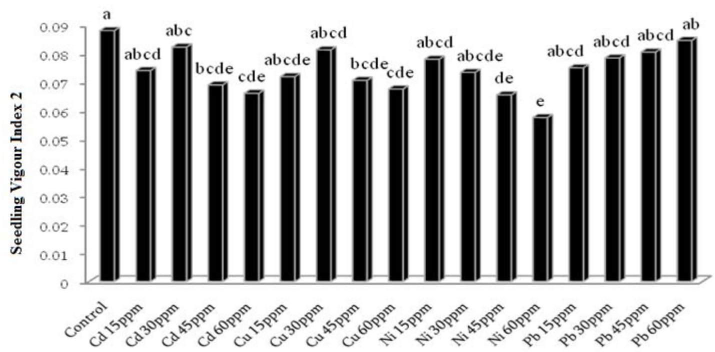

Figure 5. Effect of heavy metals on seedling vigour index 2

Discussion

It has been demonstrated that (Ivanov et al., 1998) heavy metals inactivate catalytic and regulatory proteins by interacting with sulfhydryl groups. Besides, heavy metals are capable to interact with membrane components altering its permeability, membrane potential and enzymatic activity. The adverse effects of heavy metals especially Cu have reportedly caused structural and morphological changes of roots as well as inhibition of root hair growth of seedlings (Fernandez & Henriques, 1991). Adverse effects of Cu on roots are related to severe reduction in the elongation growth of the longest root as well as root plasma membrane permeability of the seedlings was reported (McBride, 2001). The reason for the above toxic effect of Ni on seed germination and seedling growth might be due to ill effect generated by Ni on varied metabolic processes, poor elasticity of cell walls and disturbed cell proliferation (Seregin and Kozhevnikova, 2006) as well as the suppression of the activity of hydrolytic enzymes (Walker et al. 1985).

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The spectrophotometric determination of atrazine in soil extracts

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Abstract
Several instrumental methods including HPLC, GLC, TLC and LC have been adapted to be used for atrazine determination. Nevertheless, some of which are not available direct or cost effective. Hopefully, there is one another simple spectrophotometric method proposed by Kesari and Gupta (1998). In this method, Atrazine is first reacted with pyridine and converted into a quaternary pyridinium halide, subsequently it adds a hydroxyle group in the presence of alkali to form a carbinol base. In the second step a glutaconic dialdehyde is formed due to breaking of the hetrocyclic linkage of the carbinol. Finaly, the glutaconic dialdehyde couples with p-aminoacetophenone in acidic medium to form a yellow-orange plymethine dye which is determined by spectrophotometer at 470 nm. The preliminary experiments showed that atrazine determination in soil extracts using the above method is not possible. An explanation is the interference between dissolved organic matter and atrazine. In this research, the spectrophotometric method was carried out by standard-addition approach in three soil samples with different pH and organic matter content. The results showed that this method can be successfully applied for the determination of atrazine in contaminated soils. Furthermore, 5 – 50 mg atrazine per soil kg (but not lower) is the best working range for the method.
Keywords: atrazine, spectrophotometric method, standard-addition

1. Introduction

Atrazine (2-choloro-4-ethylamin0-6-isopropylamino-1,3,5-triazine) as a selective herbicide from the group of s-triazines is the most widely used pre- and post-emergency herbicide to control grassy and broadleaf weeds of corn and sorghum over the last 30 years (Kovaios et al., 2006; Lavignac et al., 2006; Xiaozhen et al., 2005; Lesan and Bhanadari, 2003). Atrazine has been recognized as a contaminant in many environmental compartments, particularly in groundwater, due to its widespread use, relatively long half-life ranging from 60 to >100 days in agricultural soils and moderate mobility(Siczek et al., 2008; Smalling and Aelion, 2006). Atrazine has a maximum concentration limit of 0.1 μg L^{-1} (European Union) or 3 μg L^{-1} (U.S. Environmental Protection Agency) for drinking water (Rousseaux et al., 2003).

Several instrumental methods including GLC, HPLC, TLC and LC are adopted for atrazine determination. These methods are often costly and unattainable. Therefore, simple and reliable methods such as spectrophotometric methods are of interest. The spectrophotometric method as a simple but sensitive technique has been introduced by Kesari and Gupta (1998).In this method, atrazine is first reacted with pyridine and converted into a quaternary pyridinium halide, subsequently a hydroxyle group is added in the presence of alkali to form a carbinol base. In the second step, a glutaconic dialdehyde is formed due to breaking of the hetrocyclic linkage of the carbinol. Finally, the glutaconic dialdehyde couples with p-aminoacetophenone in acidic medium to form a yellow-orange plymethine dye.

In this research, a spectrophotometric method modified by standard-addition approach used for determining the concentration of atrazine in extracts of three soil samples with different pH and organic matter content.

Standard addition is a technique that uses when the matrix of a sample interfere with analyte determination. It can uncover interferences and qualify dubious test results. The technique of standard addition consists of adding a series of increasing volumes of stock solution (V_{std}) to a constant volume of unknown (V_{unk}) solution. Finally, the flasks are made up with solvent to the mark line and then tested as usual (Zellemer, 1998).

2. Materials and Methods

2.1 Soil samples

Three soil samples (0-20 cm) were collected from different areas (received no atrazine) and were air-dried and screened through a 2 mm sieve.

2.1. Atrazine determination
Determination of atrazine concentration was performed according to the method of Kesari and Gupta (1998).

2.2 Atrazine
Analytical grade atrazine (99.2%) was provided by Crop Protection Organization of Iran-Tehran and employed for preparation of analytical standards. Also, a commercial formulation of atrazine (80%; Gia Co., Iran-Tehran) was purchased to carry out the experiments.

2.2. Reagents
Pyridine (C_5H_5N,99.9%), p-Aminoacetophenone (C_8H_9N0,99.8%) and Sodium hydroxide (NaOH) were purchased from Merck (Germany).

2.3 Pyridine reagent
12 ml of water was mixed with 8 ml of concentrated hydrochloric acid and 18 ml of pyridine was mixed into it.

2.4 p-Aminoacetophenone reagent
1 g of p-Aminoacetophenone was solved in 100 ml of hydrochloric acid 1:4.

2.5 Sodium hydroxide solution
A 2 M solution was prepared.

2.6. preliminary experiments
A comparative study using different volume amounts of reagents was carried out to investigate their efficiency on the color development reaction. Three series of reagent volumes employed in this experiment are presented in Table 1.

Table 1
Three series of reagent volumes

Reagent	The volume of reagent (ml)		
	Series 1	Series 2	Series 3
Pyridine	0.2	0.5	0.8
Sodium hydroxide (2 M)	1	4	7
p-Aminoacetophenone	2	5	8

2.7. Sorption isotherm experiments
A total of 70 ml of 0.01 M $CaCl_2$ solutions containing 0.5,1,2,3,4,5 mgL^{-1} atrazine was added to a series of 7 g of soil samples in centrifuge tubes and shaken in a reciprocal horizontal agitator in dark for 24 hours. In order to eliminate the microbiological activity, 1.84 mmol kg^{-1} $HgCl_2$ was added to it (Rodriguez-Cruz et al., 2006). After filtration, the concentration of atrazine was determined in soil extracts. Because the equilibrium concentrations of atrazine were found to be greater than the initial concentrations, the standard addition method was used for determining the concentration of atrazine. For this purpose, 10 ml of soil extracts was placed in each of five flasks. Then, 0, 2, 4, 8 and 10 ml of stock solution (15 mgL^{-1} atrazin in 0.01 M $CaCl_2$) were added to them. The pH of the solution was adjusted between 2 and 2.5 by 2 M HCl. Immediately; 0.5 ml pyridine reagent was added and placed on a water bath for 20 min. The flasks allowed to cool at room temperature and 4 ml of Sodium hydroxide reagent was added. Finally, 5 ml of p-aminoacetophenone solution was added and allowed to complete the colour development for 5 min.The total volume of each flask was made up to 30 ml by $CaCl_2$ 0.01 M. The concentration of atrazine was determined by spectrophotometer instrument (Apple model) at 470 nm.

3. Results and Discussion

3.1 Soil properties

Some physical and chemical properties of the soils are given in Table 2. As one can see the selected soils have a wide variation in properties.

Table 2
The general physical and chemical soil properties

Soil	Soil Texture	% Sand	% Silt	% Clay	pH	% OC
1	Sandy loam	53	32	15	4.4	4.3
2	Silty clay	16	42	42	5.7	1.2
3	Loam	40	40	20	7.9	0.5

3.2 preliminary experiments

As shown in Fig. 1, when the amount of reagent added decreased the absorbance decreased. However, due to no significant difference between the results of series2 and series3 and considering the economic considerations, the series2 (0.5 ml of pyridine reagent , 4 ml of 2 M sodium hydroxide solution and 5 ml of p-Aminoacetophenone reagent) was selected for the subsequent experiments.

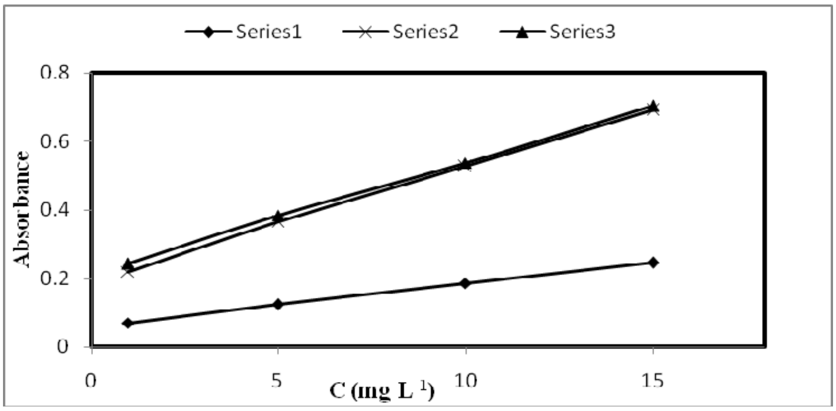

Fig. 1. The effect of different reagent amounts on color development reaction.

3.2 Sorption isotherm experiments

Determination of equilibrium concentrations of atrazine showed that these concentrations sometimes are greater than the initial concentrations. This can be attributed to the soil organic matter (SOM) dissolution and interference contribution of dissolved organic carbon (DOC) in color development. In this situation, the standard addition approach is the only reliable way to conduct the determination using spectrophotometric methods. The results of conventional method differed from those of the standard addition-based method by more than 10 percent. This indicated that our data obtained from the former method was corrupted by interferences. Determination of atrazine equilibrium concentrations using the latter method is presented in Fig 2. The traditional way to obtain the concentration of analyte from these plots is a straight line fit to the data. Nevertheless, no reliable data was found after fitting. Instead, a degree two polynomial equation provided acceptable results. According to the results the best working range of initial atrazine concentration for the method is 5 – 50 mg atrazine per soil kg (but not lower). A linear form of the sorption isotherms was fitted to the data and the results are present in Fig. 3. As one can see a typical sorption isotherm was found. Linear distribution coefficients (K_d) increased from 3.72 to 27.33 L kg^{-1} with increasing the percentage of soil organic carbon from 0.5 to 4.3%. This may be due to the main role of soil organic matter to adsorb atrazine. Park et al.(2003) reported similar results. The range of K_d in their work was 1.5 to 43.1 L kg^{-1}. The results of our research showed that the standard addition-based approach can be successfully applied for determining of atrazine by spectrophotometric methods in contaminated soils.

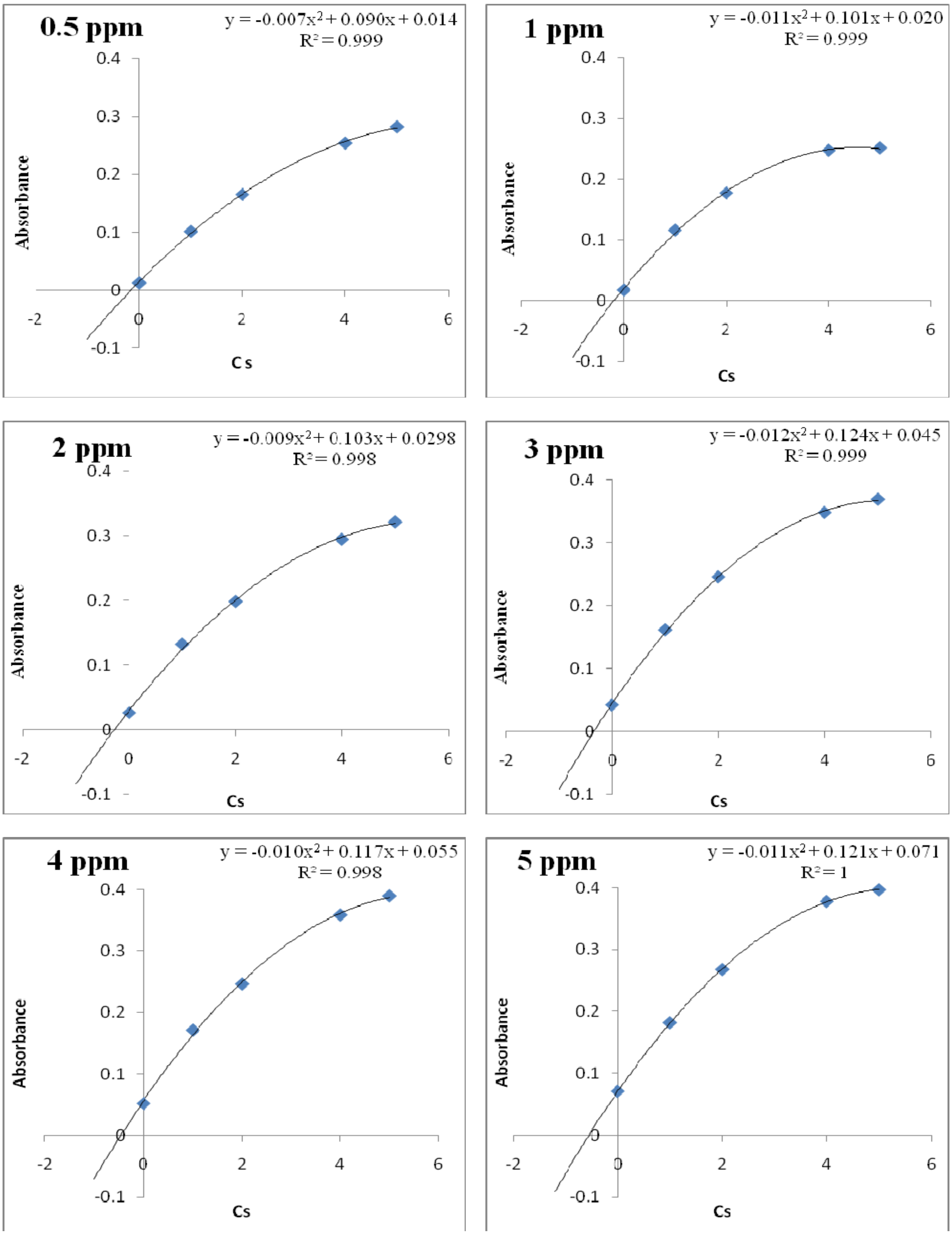

Fig. 3. Determination of atrazine equilibrium concentrations in soil 2 by standard addition approach.

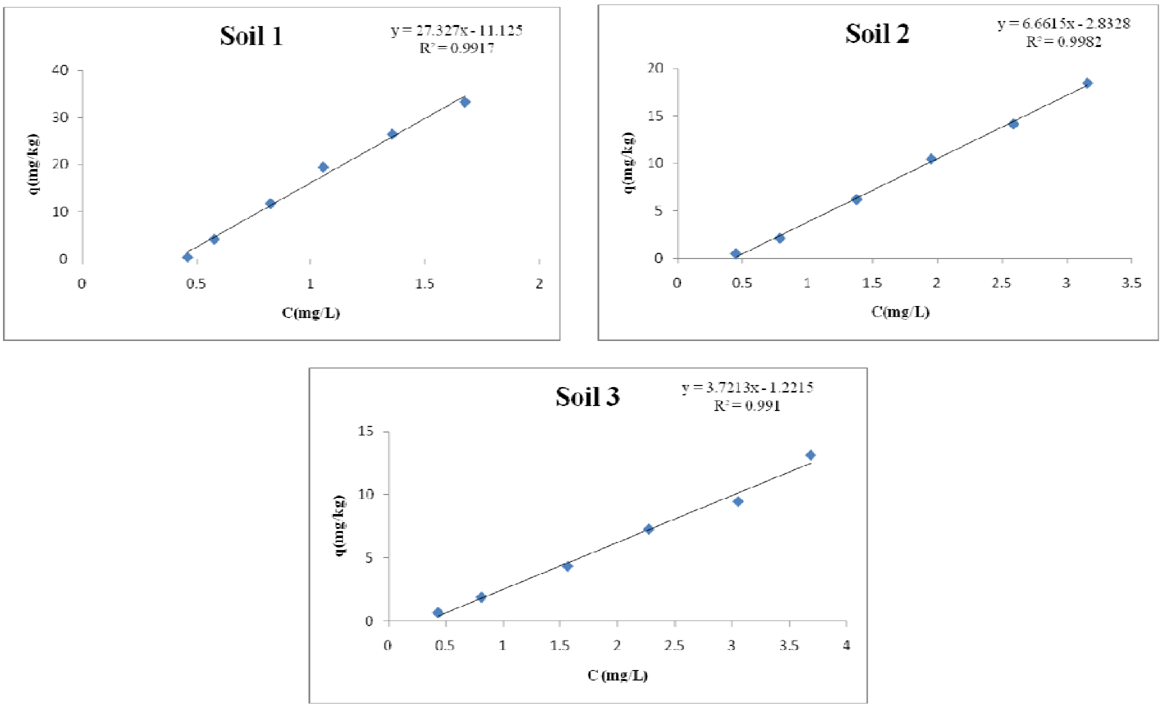

Fig. 4. Linear sorption isotherms of atrazine for three soils.

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Effect of former mining activity on Zn and Pb contents in natural soils from Cartagena-La Unión mining district, SE Spain

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Abstract: Seven tailing ponds and two natural areas (soils developed on ferruginous limestone and soils developed on laminated limestone) from a Pb/Zn mining district from southeast Spain were sampled and geochemically characterized. The objectives of this study were to evaluate the degree of pollution of the area using the enrichment factor, the geo-accumulation index and a combined pollution index; to determine the origin of Pb and Zn in the natural areas by mean of sequential extraction procedures, since wind erosion may have transferred metals from bare tailing ponds to the natural soils; and to assess the potential environmental risk of the area. Results showed that the tailing ponds and natural soils are highly polluted by Pb and Zn. Soils developed on ferruginous limestone are more polluted than soils formed from laminated limestone, and the surface samples show higher enrichment of metals than subsurface samples. According to the results of mineralogy, statistical analysis, concentration of heavy metals in soil profiles and sequential extraction analysis of Pb and Zn we can conclude that the high levels of Pb and Zn in soils developed on laminated and ferruginous limestones may be due to a geogenic and antropogenic origin. Due to the high concentrations of water-soluble Zn and bio-available Pb and Zn in most of the tailing ponds, it is recommended to carry out reclamation actions in order to reduce the Zn solubility and decrease the bioavailability of Pb and Zn for plants.
Keywords: Tailing ponds, heavy metals, soil contamination, sequential extraction, natural soils

Introduction

Lead and zinc extractions were intense for more than two thousand years in the mining district of Cartagena-La Unión (southeast of Spain), ceasing its activity in the late 20th century. As a result of the extraction activities, large amounts of wastes were generated, which are still part of the landscape of the area. These wastes gave rise to the so-called tailing ponds that are still bringing with it significant environmental risks.
Different studies have been developed in the Cartagena - La Unión mining district where pollution has been assessed by the presence of heavy metals in tailing ponds (Acosta et al., 2011). However, scarce information is available on metals in natural soils close to these tailing ponds, including total concentration of metals and their distribution in the different solid phases.
When a metal reaches soil surface, depending on soil properties and characteristics, it can be bound to different soil constituents, which includes water-soluble, exchangeable, organic associated, carbonate associated phases, bound and occluded in oxides and secondary clay minerals phase, and residual within the primary mineral lattice phase (Li and Shuman, 1996). In general, metals tend to be fixed in the soil when the pH is alkaline, however the risk is much higher in tailing ponds, owing to their low pH, high metal content, low organic matter and scarce or null vegetation. In addition, the oxidation of pyrite transforms the sulfide to sulfate, causing the solubilization of metals, which are likely to be washed with rain water and increase pollution in the soils and waters of the surrounding (Pavetti et al., 2006).
The aims of this study are (1) to carry out the geochemical characterization of seven tailing ponds from a Pb/Zn mining extraction area and the surrounding natural areas; (2) to evaluate their degree of pollution; (3) to determine the origin of Pb and Zn in the natural areas; and (4) to assess the potential environmental risk in the studied area.

Materials and Methods

Study area, soil sampling and analytical methods

The study was carried out in a zone called “El Avenque” stream, in the mining district Cartagena-La Unión (Murcia Region, SE Spain) (Figure 1). The weather of the area is semiarid Mediterranean, with an average annual temperature of 18 °C and an average annual rainfall of 275 mm. Seven tailing ponds located in this stream and two natural areas developed on ferruginous and laminated limestones were selected (Figure 1).

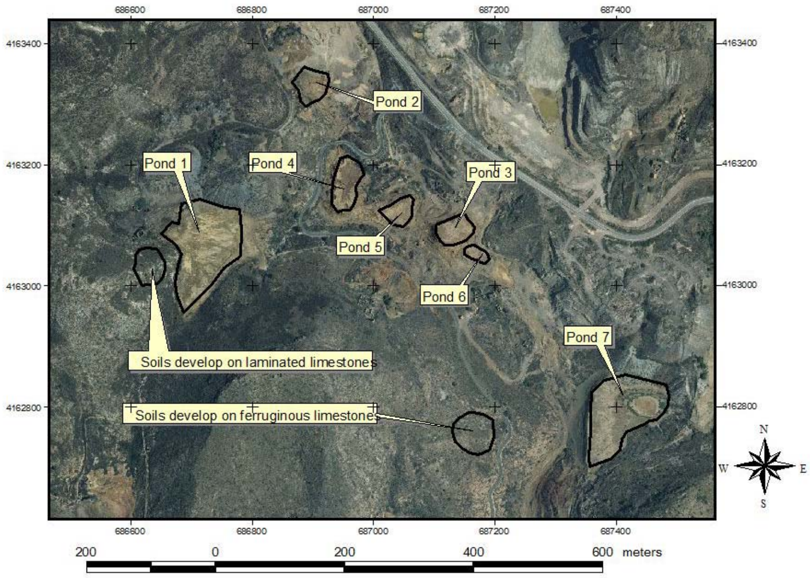

Figure 1. Study area

Surface waste and soil samples (0–15 cm) from the tailing ponds and natural areas were collected according to a regular sample grid.

All samples were air-dried for 7 days in the laboratory, passed through a 2-mm sieve, homogenized. Soil analyses for this study were determined as follows: pH and electrical conductivity (EC) were measured in a 1:1 and 1:5 deionised water/soil ratio solution, respectively, according to Peech (1965); Soil organic carbon (SOC); cation exchange capacity (CEC) was measured following the method of Chapman (1965); equivalent calcium carbonate was estimated using a Bernard calcimeter and particle size analysis (FAO-ISRIC, 2006).

Total Pb and Zn concentrations of bulk soil were determined after digestion in a microwave oven (EPA, 1996). The bioavailable metal fractions were determined using DTPA (1:2 soil-extractant ratio) for samples with pH>6, and EDTA (1:5 soil-extractant ratio) for samples with pH<6 (Lindsay and Norwell, 1978). Soluble heavy metals were determined using a 1:5 soil-deionized water ratio (Erns, 1996). Measurements of metals were carried out using atomic adsorption spectrophotometer (AAnalyst 800, Perkin Elmer). The used sequential extraction scheme was developed by Tessier et al. (1979). Fraction 1: exchangeable; b) Fraction 2: bound to carbonate and specifically adsorbed; c) Fraction 3: reducible; d) Fraction 4: oxidizable; e) Fraction 5: residual phase. Mineral compositions of samples were determined by X-ray diffraction (XRD).

Enrichment factor

The amount of anthropogenically-introduced metal into the soil was determined using an enrichment factor (EF_x). EF_x was calculated with respect to local background:

$$EF = \frac{[metal]_{sample}}{[metal]_{background}}$$

Where $[metal]_{sample}$ is the metal concentration in the sample (Pb and Zn), and $[metal]_{background}$ is the metal concentration proposed by Martínez-Martínez (2009) from soils of the Murcia Region..

Geoaccumulation index

Calculating the geoaccumulation index (Igeo) allows estimating the enrichment of metal concentrations above background concentrations:

$$Igeo = \log_2\left(\frac{[metal]_{sample}}{1.5x[metal]_{background}}\right)$$

where [*metal*] is the heavy metal concentration measured.

Combined Pollution Index

Abrahim (2008) presented an index which uses as many metals as the study may analyze:

$$CPI = \sum_{i=1}^{i=n} \left(\frac{[metal]_{sample}}{(metal)_{background}} \right) \Big/ n$$

where [*metal*] is the metal concentration in the sample, compared with the reference levels proposed by Martínez-Martínez (2009), and *n* is the number of analyzed elements.

Results and discussion

Geochemical characterization of tailing ponds and natural areas

Properties and characteristics of the tailing ponds were very heterogeneous among them (Table 1). pH values of the tailing ponds varying from ultra acid to neutral. Low pH in these soils was due to the oxidation of pyritic minerals. EC ranged from very slightly saline to slightly saline due to the presence of soluble sulfates. The CEC values ranged from very low to low which can be related to the very low clay and total organic carbon contents.

Table 1. Properties and metal concentrations of the seven selected tailing ponds

	Tailing pond 1		Tailing pond 2		Tailing pond 3		Tailing pond 4		Tailing pond 5		Tailing pond 6		Tailing pond 7	
	Mean (SD)	Range	Mean (SD)	Range	Mean (SD)	Range	Mean (SD)	Range	Mean (SD)	Range	Mean (SD)	Range	Mean (SD)	Range
pH_{H20}	2.88 (0.459)	1.9-3.4	4.78 (1.78)	3.4-7.5	6.36 (1.07)	4.7-7.9	4.19 (0.86)	3.4-5.9	5.91 (0.79)	5.1-7.1	4.46 (1.84)	3.1-7.7	6.87 (0.886)	4.3-7.7
EC (dS m^{-1})	3.71 (1.671)	1.3-6.8	2.38 (1.55)	0.4-4.7	3.75 (1.67)	2.2-5.9	2.56 (1.75)	1.5-6.8	2.99 (1.53)	2.1-64	3.70 (1.33)	2.3-5.1	5.08 (5.37)	1.1-20.9
CEC (cmol kg^{-1})	8.58 (2.67)	2.8-12.1	11.16 (4.02)	5.2-16.2	5.16 (2.13)	2.7-9.0	5.32 (1.62)	2.7-7.2	2.19 (0.87)	1.7-4.1	7.62 (1.86)	5.1-10.1	5.32 (1.33)	3.4-8.5
SOC (g kg^{-1})	3.44 (1.27)	1.4-5.8	4.40 (2.52)	2.5-8.2	6.42 (2.25)	3.3-10.3	2.27 (1.23)	0.6-4.2	5.01 (2.19)	1.9-8.0	4.96 (5.37)	2.0-14.5	7.93 (1.98)	4.4-11.2
Sand (%)	71.7 (21.77)	22.6-94.6	61.9 (19.52)	38.7-86.5	47.1 (31.64)	12.7-84.9	74.5 (24.3)	14.0-91.8	60.3 (23.59)	25.0-87.0	61.56 (28.37)	30.2-89.1	73.6 (18.57)	34.0-91.9
Silt (%)	20.8 (17.30)	2.0-61.1	32.5 (18.40)	11.2-57.6	47.4 (31.63)	10.4-82.6	17.6 8 (17.5)	6.1-62.0	35.7 (23.99)	7.7-71.0	30.21 (23.36)	10.5-65.8	21.3 (16.07)	5.4-54.7
Clay (%)	7.6 (5.55)	2.5-21.9	5.6 (2.92)	2.2-9.0	5.4 (0.79)	4.6-7.0	7.88 (7.07)	1.8-24.0	4.0 (1.84)	1.0-6.0	8.23 (11.69)	0.5-28.7	5.14 (2.86)	1.6-11.4
Total Lead (mg kg^{-1})	1761 (846.2)	855-3463	9861 (8324)	5087-24441	12775 (4631)	4781-18750	41636 (11111)	25424-55508	10758 (3922)	5903-17413	26220 (21314)	2152-48040	6913 (3092)	3727-13954
Total Zinc (mg kg^{-1})	1586 (294.1)	1161-2201	6847 (2671)	4078-10409	22799 (9086)	7836-36232	13288 (4614)	5537-19696	20781 (14451)	5200-40653	6154 (2802)	1863-8514	12715 (4326)	2898-20045
Bio. Lead (mg kg^{-1})	76.4 (205.0)	< D.L.-755	608 (215.6)	380-844	775 (1621)	3.5-4618	5298 (1341)	3205-8181	1046 (1570)	5.70-4343	1696 (2625)	76.4-6348	65.0 (85.6)	1.70-214
Bio. Zinc (mg kg-1)	146 (121.4)	13.0-369	502 (434.1)	110-1057	2449 (4131)	439-13841	3.83 (1.99)	2.10-7.70	3.39 (1.76)	1.10-6.10	1.25 (0.55)	0.60-1.90	497 (122)	262-753
Soluble Lead (mg kg^{-1})	1.55 (2.77)	0.10-10.60	6.55 (5.61)	0.40-11.10	3.96 (3.77)	0.20-11.50	21.83 (6.61)	3.90-27.60	7.64 (8.67)	0.40-21.40	14.44 (10.63)	1.80-29.90	0.38 (0.53)	< D.L.-1.90
Soluble Zinc (mg kg^{-1})	133 (100.5)	12.9-337	208 (283.8)	0.30-705.3	1463 (1975)	0.50-5432	0.46 (0.77)	0.10-2.90	0.37 (0.65)	0.10-1.80	0.47 (0.43)	0.10-1.20	148 (267)	0.80-858

Geochemical characteristics of the natural areas are more homogeneous than in the tailing ponds (Table 2). They had an average pH ranging from slightly alkaline to moderately alkaline due to the presence of calcium carbonate. EC demonstrated that these soils are non-saline. CEC was moderate, which is associated to a high total organic carbon content and clay.

Table 2. Properties and metals concentration of the natural soils

	Soils developed on ferruginous limestone		Soils developed on laminated limestone	
	Mean (SD)	Range	Mean (SD)	Range
Surface samples (0-15 cm)				
pH_{H20}	7.72 (0.16)	7.5 -8.0	7.9 (0.09)	7.8- 8.1
EC (dS m^{-1})	0.22 (0.04)	0.20-0.30	0.29 (0.09)	0.20-0.50
$CaCO_3$	76.59 (86.08)	13.4-224	359 (81.54)	249-496
CEC (cmol kg^{-1})	21.46 (4.61)	15.1-28.5	17.2 (3.91)	9.70-23.10
SOC (g kg^{-1})	34.56 (13.16)	16.7-54.1	43.8 (9.35)	27.70-57.00
Sand (%)	25.92 (7.58)	13.0-39.8	33.06 (5.84)	24.90-43.90
Silt (%)	43.52 (5.83)	35.6-54.0	41.53 (3.19)	35.80-45.70
Clay (%)	30.59 (7.59)	24.1-45.1	25.41 (5.87)	15.80-31.40
Total Lead (mg kg^{-1})	4737 (2944)	510-10725	2347 (828)	1205-3447
Total Zinc (mg kg^{-1})	4518 (2247)	1749-7645	1731 (1013)	720-3801
Bio. Lead (mg kg^{-1})	297 (258.3)	67-827	415.6 (199)	186-810
Bio. Zinc (mg kg^{-1})	362.0 (166.9)	133-543	189 (85.40)	84.90-306
Soluble Lead (mg kg^{-1})	6.58 (8.20)	<D.L.-23.81	6.69 (4.77)	<D.L.-12.79
Soluble Zinc (mg kg^{-1})	11.70 (12.87)	1.54-39.90	8.54 (6.83)	1.33-21.02

The mineralogical analysis of the collected samples in the tailing ponds showed, in most cases, the presence of quartz, magnetite, jarosite, goethite and gypsum. Oppositely, the main minerals composing the natural soils were, for the soil developed on ferruginous limestone: quartz (SiO_2), dolomite, kaolinite, hematite, calcite, muscovite and orthoclase and for the soil formed from laminated limestone quartz, dolomite, calcite, muscovite and montmorillonite.

Degree of pollution

The tailing pond 4 had the highest mean value of total Pb (41630 mg kg^{-1}), and the tailing pond 3 the highest mean value of total Zn (22799 mg kg^{-1}), while the lowest concentrations of both metals were found in tailing pond 1 (1761 mg Pb kg^{-1} and 1586 mg Zn kg^{-1}), (Table 1). The concentrations of Pb and Zn in natural soils were also very high (4737 mg Pb kg^{-1} and 4518 mg Znkg^{-1} in soils developed on ferruginous limestone, and 2347 mg Pb kg^{-1} and 1731 mg Zn kg^{-1} in soils formed from laminated limestone) (Table 2), indicating a high degree of pollution in these areas, what means that both areas are high enriched in these heavy metals.

Results obtained from the enrichment factors in the tailing ponds (Table 3) showed that all ponds have an important enrichment of Pb and Zn, exceeding the boundaries proposed by Lacatuso (1998) for considering a soil excessively contaminated (EF>16). In the same way, soils developed under ferruginous and laminated limestone are enriched in Pb and Zn at both depths, with EF>16 for all samples, except for subsurface samples of soil formed from laminated limestone (EF=7.9). The highest enrichments were found in surface samples from the ferruginous limestone area (EF=106).

Table 3. Enrichment factors. Geo-accumulation index and combined pollution index

	Lead		**Zinc**		
	EF	**GI**	**EF**	**GI**	**CPI**
Tailing pond 1	39.6 (19.0)	4.6 (0.7)	336 (354)	4.2 (0.3)	34.2 (10.3)
Tailing pond 2	221 (187)	6.9 (1.0)	124 (48.5)	6.3 (0.6)	173 (106.9)
Tailing pond 3	287 (104)	7.5 (0.6)	414 (165)	8.0 (0.7)	350 (111)
Tailing pond 4	935 (249)	9.2 (0.4)	241 (83.7)	7.2 (0.5)	588 (134)
Tailing pond 5	242 (88.0)	7.2 (0.5)	377 (262)	7.6 (1.2)	309 (148)
Tailing pond 6	589 (479)	7.7 (2.0)	112 (50.9)	6.0 (0.9)	350 (263)
Tailing pond 7	155 (69.4)	6.6 (0.6)	231 (78.5)	7.1 (0.7)	193 (69)
Ferruginous limestone					
Surface (0-15 cm)	106 (66.1)	5.8 (1.3)	82.0 (40.8)	5.6 (0.8)	94.2 (51.7)
Subsurface (15-30 cm)	77.6 (68.7)	5.1 (1.4)	48.8 (39.5)	4.5 (1.4)	63.2 (52.4)
Laminated limestone					
Surface (0-15 cm)	52.7 (18.6)	5.0 (0.6)	31.4 (18.4)	4.2 (0.8)	42.1 (17.2)
Subsurface (15-30 cm)	27.5 (13.0)	4.0 (0.7)	7.9 (6.1)	1.7 (1.7)	17.7 (9.3)

EF: enrichment factor. GI: geo-accumulation index. CPI: combined pollution index

Values obtained from the geo-accumulation index (GI) demonstrated that all tailing ponds, except tailing pond 1, were very highly polluted by Pb and Zn (GI>5) (Muller, 1981). Similarly, the soils developed from ferruginous limestone showed a high geo-accumulation index close to 5 for both metals; therefore, they should be considered as very highly polluted (Table 4). Oppositely, the GI of Zn in soils formed from laminated limestone were below 5, being in subsurface samples < 2, classified as moderately polluted by Zn (Muller, 1981).

Finally, results obtained from the combined pollution index (CPI) showed that all tailing ponds, except pond 1, widely exceeded the values proposed by Abrahim (2008) for considering a soil ultra high polluted. However, the values from natural soils were much lower, although they were still high (CPI>32). Therefore, it can be said that the soil developed on ferruginous limestone is more polluted than the soils developed on laminated limestone. In addition, and the surface samples showed higher enrichment of metals than the subsurface samples.

Identification of Pb and Zn origin in the natural areas

Samples taken from natural soils showed high levels of Pb and Zn (Table 2). Two possible hypothesis can explain the origin the Pb and Zn in these soils:

1. *Geogenic origin*: The mineralogical composition of the natural soils is very different to the minerals found in the tailing ponds, which seems to confirm that the aerial transport by wind of particles containing Pb and Zn is not the main source of these metals in the natural soils. Besides the principal component analysis (PCA) shows that the behaviour of Pb and Zn in soils developed on laminated and ferruginous limestones is different to the soils from tailing ponds (data not shown). In addition, the acidity and the increase of the SOC favour significantly the mobility of Pb and Zn. However, in the soils from tailing ponds is checked that an increase of the bioavailable Pb and water soluble Zn concentrations are only slightly influenced by the SOC.

2. *Antropogenic origin*: High significant correlation between total Pb and Zn exist in these natural areas (data not shown). And the same happens with the total content of these heavy metals in the tailing ponds. Therefore, the natural soils have been able to be influenced by mining activity

carried out in the proximities in the past. On the other hand, the Pb and Zn values obtained in the soil profiles developed on laminated and ferruginous limestones (data not shown) revealed that although the concentrations of these heavy metals in the parent material are significant, the surface soil horizons have very high contents of Pb and Zn, so they could have been influenced by wind transporting the heavy metals from mining areas.

In order to elucidate the hypotheses explained before, sequential extraction procedure was applied to the tailing pond and natural soils samples. The Pb and Zn retained in the five solid phases for tailing ponds and natural soils can be observed in Figure 2. Important differences were obtained for both metals in samples from tailing ponds and natural soils. For Zn in natural soils, the fraction with the highest concentrations and percentages was the residual phase. Oppositely, reducible phase was the most important one to retain Zn in the tailing ponds. In addition, carbonate fraction showed a significant importance for Zn in natural soils.

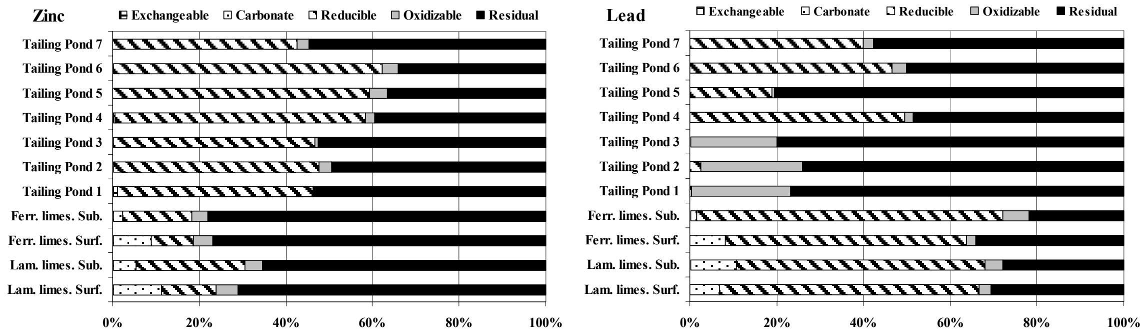

Figure 2. Distribution of Zn and Pb in the solid phases for tailing ponds and natural soils

For Pb, the highest concentrations and percentages were bound to Fe and Mn oxides for both natural soils. Oppositely, oxidizable and exchangeable fractions had the lowest concentrations and percentages (Figure 2). In addition, the percentage of Pb bound to carbonates was also significant, with a value close to 10 % of the total concentration, showing the accumulation of Pb in the sorbed-carbonate phase. Oppositely, in the tailing ponds the most important phase where the Pb is bound is the residual phase (Figure 2).

These differences found between tailing ponds and natural areas with regards to the distribution and behaviour of metals in the solid phases (Figure 2), support the previous hypothesis that the concentration of Pb and Zn in the natural areas has a geogenical origin.

Environmental risk of metal in the area

In the natural areas (Tables 2), water soluble Pb and Zn concentrations showed very low levels, being similar in both areas. In general, the concentrations of soluble Pb and Zn were higher in surface than in subsurface, which can be due to a more intensive weathering in the soil surface. Similarly, water soluble concentrations of Pb were low in the tailing ponds, showing the low solubility of this metal, and therefore its low mobility by water transport, demonstrating that Pb is not highly mobilized by water in the environment (Tables 1). Oppositely, soluble Zn concentrations were very high in most of the tailing ponds, up to 1463 mg kg^{-1}, which indicates that this metal can be transported by water and pollute the surrounding areas.

Concentrations of metals extracted by quelating agents (DTPA and EDTA) are much higher than those reported for water soluble metals (Tables 2 and 3), which suggests that high concentrations of these metals can be taken up by plants, creating risk for the food chain. In the case of the natural soils, concentrations in surface were higher than those reported in subsurface for both metals. In general, the bioavailable metals in the tailing pond samples were higher than those found in natural soils; therefore, the risk of metal uptake by plants is higher. Nevertheless, since most of the tailing ponds remain bare this is not a current risk, although it should be taken into account in future reclamation plans of these tailing ponds.

Conclusions

In the tailing ponds the pH values range from ultra acid to neutral. Low pH in these soils was due to the oxidation of pyritic minerals. The CEC, OC and clay contents were very low due to the origin of these mining wastes and the absence of vegetation cover. Oppositely, natural soils of the surrounding showed a pH values ranging from slightly alkaline to moderately alkaline due to the presence of calcium carbonate; CEC was moderate, which is associated to a high total OC and clay content.

Tailing ponds and natural areas were highly polluted by Pb and Zn. However, the used indexes indicated that the pollution in the tailing ponds was much higher than in natural soils. Soils from ferruginous limestone were more polluted than soils developed on laminated limestone, and the surface samples showed higher enrichment of metals than subsurface samples. The high levels of Pb and Zn in soils developed on laminated and ferruginous limestones may be due to a geogenic and antropogenic origin according the results on mineralogy, statistical studies, concentration of heavy metal in soil profiles and sequential extraction analysis of Pb and Zn. In this sense, it would be interesting to use other methods in future studies that allow us to elucidate whether the high concentrations of Pb and Zn in natural soils are due to a geogenic or anthropogenic origin, such as the study of different isotopes of Pb and Zn present in the soil samples.

In both natural areas and tailing ponds water soluble Pb concentrations showed very low levels showing that Pb is not highly mobilized by water in the environment. Oppositely, soluble Zn concentrations were very high in most of the ponds, which indicates that this metal can be transported by water and pollute the surrounding areas. However, high concentrations of Pb and Zn metals extractable by chelating agents suggest that they can be taken up by plants creating risk for the food chain. Therefore, it is recommended to develop reclamation actions as soon as possible in order to reduce the Zn solubility in the ponds and decrease the bioavailability of Pb and Zn for plants.

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Effect of camelid grazing in the soil fertility and vegetation at high altitude grassland in the Bolivian Andean

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Abstract: The high grasslands of Apolobamba Integrated Management National Area (Bolivian Andean) provide a natural habitat for a high number of wild and domestic camelids such as vicuna (*Vicugna vicugna*) and alpaca (*Lama pacos*) respectively. Because of the importance of the camelid raising for the Apolobamba´s inhabitant economy, it is fundamental to determine the natural resources condition and their availability for the camelid support. Therefore, the objectives of this research were to: (i) evaluate the soil fertility (ii) identify and characterize the vegetation species and plant communities and (iii) analyze the effect of the camelid populations in soil properties, plant coverage and species palatability. Soil and vegetation samplings were carried out in eight areas with different vicuna densities and soil properties, botanical taxonomy, plant coverage and palatability were determined. The physico-chemical soil results pointed out the good soil fertility and no camelid grazing affection. However, erosive processes could be taken place likely caused by the alpaca grazing. The botanical taxonomy pointed out that the most plentiful plant community was *Pycnophyllum* grassland although it was highly disturbed attributable to the domestic camelid grazing. The studied areas presented medium (30-50 %) and high plant coverage (above 50 %). The substitution of palatable plant species by no palatable ones in zones with high alpaca concentration highlighted the negative domestic camelid effect in the vegetation composition. Conversely, no vicuna affection on soil fertility and vegetation was observed. Therefore, it should be undertaken biodiversity protection actions bringing domestic camelid overexploitation under control in the Apolobamba Bolivian Andean.
Keywords: Bolivian high grasslands, camelid affection, plant communities, soil properties, vicuna

Introduction

The Apolobamba Integrated Management National Area is located in the Bolivian Andean. Its highlands provide a natural habitat for wild camelids such as vicuna (*Vicugna vicugna*) and domestic ones such as alpaca (*Lama pacos*). Aymara and quechua indigenous communities live in Apolobamba and their main economical activity is the domestic camelid raising. Vicuna is a near threatened species recognized by The World Conservation Union (IUCN, 2001). Ecosystems in the *puna* or high altitude grasslands in the Andean region are degraded as a consequence of anthropogenic activities such as the excessive cattle grazing (Rocha and Sáez, 2003). Because of the importance of the wild and domestic camelid raising for the Apolobamba´s inhabitant economy, it is fundamental to determine the natural resources condition and their availability to the camelid supporting.

However, the previous studies conducted in the Bolivian Andean grasslands showed limited information about soil, vegetation and camelid grazing affection (Seibert, 1993; Beck et al., 2002). In order to supply further information the objectives of this research were to: (i) evaluate the soil fertility (ii) identify and characterize the vegetation species and plant communities and (iii) analyze the effect of the camelid populations in soil properties, plant coverage and species palatability in the Apolobamba area. The information supplied by this research could improve the understanding of the properly Andean grassland management and the sustainable camelid exploitation.

Materials and methods

Study area

Apolobamba is an Integrated Management National Area located in the Northwest of La Paz, Bolivia, border with the Republic of Peru. The research was carried out in the puna of the Apolobamba area, the highest altitude ecological zone ranging from 4300 to 4900 m.a.s.l. and 1200 km^2 of extension (SERNAP, 2006). The study area is characterized by udic and frigid soil moisture and temperature regimes (USDA, 2010) with an annual average temperature of 4.5 ºC and total precipitation of 505 mm which is concentrated in five months, from November to March. The puna in Apolobamba is included at the North Puna eco-region (Navarro and Maldonado, 2002)

Soil sampling and analytical methods

Due to the vicuna management programme significance to the Apolobamba´s inhabitant development and the biodiversity protection, vicuna population density was the main reason to select the studied zones. Regarding to the last vicuna censuses carried out in Apolobamba (SERNAP, 2006), eight zones with dissimilar vicuna densities were selected. The selected zones were: zones 1 and 2 (low vicuna density, from 2.1 to 9.4 individuals km^{-2}), zones 3 and 4 (medium vicuna density, from 9.4 to 16.5 individuals km^{-2}), zones 5 and 6 (high vicuna density, from 16.5 to 23.2 individuals km^{-2}) and zones 7 and 8 (very high vicuna density, from 23.1 to 58.1 individuals km^{-2}). Zone 3 presented very high alpaca concentration while zone 1 presented high concentration. The other studied zones exhibited medium alpaca density.

In order to characterize the soils, three replicate plots of 5 x 5 m were chosen in each studied zone. Soil and vegetation samplings were carried out in the dry season from May to October. Since most of the roots were located in the surface layer, three replicate soil samples were randomly collected from 0 to 5 cm depth. The following soil analyses were carried out: Total Organic Carbon (TOC) was determined using a TOC Analyzer (Shimadzu 5000, Japan); total nitrogen (TN) was measured according to the Kjeldahl method (Duchafour, 1970); phosphorous was determined using the method of Watanable and Olsen (1965); cation exchange capacity (CEC) by Chapman (1965); pH in 1:1 (w/v) aqueous solution (Peech, 1965) and texture through pipette Robinson (FAO-ISRIC-ISSS, 2006).

Vegetation sampling and methodology

The same three soil sampling plots per zone were considered to carry out the vegetation sampling. Vegetation samples were collected and they were pressed and dried into an oven at 80 ºC during 24 hours. The botanical identification was carried out in the Herbario Nacional de Bolivia using its own botanical data base and the Missouri Botanical Garden´s one (W^3 Tropicos database). The phytosociological characterization of the plant communities was established following Braun-Blanquet (1964) methodology described by Seibert (1993). The plant coverage percentage was determined following to Huss et al. (1986) methodology, modified using a sample grid or transect of 50 x 50 cm and needles inserted with 90 º in each sampling plot.

Statistical analyses

The normality of the distribution of soil parameters and plant coverage was studied by Shapiro-Wilk´s Test through the residual analysis with Normal Probability Plot Test. One-way ANOVA followed by a post-hoc Tukey´s Test at $P<0.05$ was completed to identify significant differences between zones through the comparisons of all possible pairs of means. Pearson correlation analysis among soil properties, plant coverage, species richness and alpaca and vicuna density was studied (Statistix 9.0).

Results and discussion

Soil properties

Table 1 shows the physico-chemical soil properties studied. According to Vagen et al. (2006) classification in mountain soils, zones 1 and 3 presented TOC concentration close to the mean value for good soil fertility (55.72 g kg^{-1}) while zones 5 and 6 exhibited TOC contents under this mean value and similar to the average value (43.93 mg kg^{-1}). Oppositely, zones 4 and 7 presented the highest TOC contents which were much higher than the good fertility mean value. With respect to the TN contents, all the studied zones showed concentrations above the mean value for good fertility (3.90 g kg^{-1}) as the same as the CEC (11.76 $cmol_{(+)}$ kg^{-1}) (Vagen et al., 2006). Clay contents were similar to good fertility mean (14.32 %) (Vagen et al., 2006). Texture analyses showed sandy-loam and loam soils (FAO-ISRIC-ISSS, 2006). Zone 8 was extremely acid based on the Soil Survey Division Staff (1993) classification, while the other zones were very strongly acid and strongly acid.

Table 1: Physico-chemical soil properties, mean values and standard errors (n=9) from 0 to 5 cm.

Zones	TOC (g kg^{-1})	TN (g kg^{-1})	Available P (mg kg^{-1})	CEC (cmo+kg^{-1})	pH	Sand (%)	Clay (%)
1	51.5 ±6.4 cd	4.3 ±0.4bcd	266.4±45.6a	13.9±0.6 de	5.0±0.1 c	69.0±2a	11.1±0cd
2	58.9±5.2 bc	5.0±0.2 b	122.9±14.7c	20.9±0.4 ab	5.9±0.1 a	40.0±3d	18.0±2a
3	55.0±3.1 bc	4.6± 0.3 bc	165.4±9.4abc	16.3±0.8cd	5.6±0.2 a	60.6±3ab	16.0±1ab
4	91.7±5.6 a	7.2±0.4 a	209.1±7.0ab	23.5±1.5 a	5.1±0.1c	44.9±1cd	14.0±1b
5	37.3±2.0 d	3.3±0.2 d	156.6±12.1abc	14.2±1.1 de	5.4±0.1 b	63.4±4ab	11.1±2bcd
6	45.9±2.3 cd	3.9 ±0.1cd	150.3±17.3bc	12.3±0.6 e	4.9±0.1 c	66.0±1a	11.0±1d
7	72.7±3.8 ab	5.2±0.3 b	172.0±12.8abc	18.3±0.9 abc	4.9±0.0 bc	52.0±2bc	13.8±1b
8	61.1±5.9 bc	5.2±0.4 b	176.7±9.8abc	17.2±1.4 bcd	4.2±0.1 d	49.9±3bc	14.0±1b
	p(W):0.29	p(W):0.46	p(W):0.53	p(W):0.83	p(W):0.34	p(W):0.07	p(W):0.41
	p(B):0.16	p(B):0.30	p(B):0.08	p(B):0.06	p(B):0.06	p(B):0.06	p(B):0.09
	F:14.1***	F:17.0**	F:10.2**	F:14.3***	F:34.9***	F:18.4***	F:12.3***

TOC: Total Organic Carbon; TN: Total Nitrogen; Available P: Available Phosphorous; CEC: Cation Exchange Capacity

Different letter indicates significant differences among zones (p<0.05) according to Tukey´s Test

p (W): Shapiro - Wilk´s Test; p (B): Bartlett´s Test; (F): Statistic ANOVA Test

*, **, ***: significant differences among surface and surface at the level of probability 0.05, 0.01, 0.001, respectively; n.s.= not significant differences

Higher available P contents were registered in some studied zones, mainly in zone 1. Some researches established that the presence of high phosphorous concentration and the low clay contents could be associated to the phosphorous mobilization as a result of the erosive processes in grasslands (Quinton et al., 2001; Owens and Walling, 2002). According to these authors, erosive processes could take place in zone 1. However, in general, the soil parameters studied pointed out good soil fertility in the studied zones.

Botanical taxonomy

The botanical identification of the samples pointed out similar families, generas and species in the different studied zones (Table 2). The Poaceae family species were found in the entire of the studied zones with species such as *Aciachne acicularis*, *Festuca rigescens*, *Deyeuxia rigescens*, *Nasella brachyphylla*, *Deyeuxia minima* and *Stipa ichu*. The Caryophillaceae family was observed in all the zones and was represented mainly by *Pycnophyllum kobalantum*, although *Pycnophyllum molle* and *Paronychia andina* were only collected in some zones.

Different species of Asteraceae family were identified in different zones (*Senecio spinosus*, *Belloa sp*. and *Perezya pygmaea*). One Cyperaceae family species (*Scirpus rigidus*) was found while *Alchemilla pinnata* which belongs to Rosaceae family was observed in some zones (Table 2). Regarding to Plantagineaceae family, *Plantago sericea* was collected in three zones and *Azorella diapensioides* (Umbeliferae family) was also identified. Additionally, two Pteridophyta species were collected, *Selaginella peruviana* (Selaginellaceae) and *Parmelia sp*., (Parmeliaceae).

Since the most representative plant species found was *Pycnophyllum koballantum* (identified in the eight studied zones) and according to the Seibert´s (1993) phytosociological characterization in the puna of the Apolobamba area, the most plentiful plant community observed was *Pycnophyllum* grassland.

Table 2: Landscape characteristics and identified species in each studied zone.

Zone	Exposition	Altitude (m.a.s.l.)	Specie identified
1	North	4367	*Festuca rigescens, Deyeuxia rigescens, Nasella brachyphylla, Aciachne acicularis, Pycnophyllum kobalantum, Senecio spinosus, Plantago sericea,Selaginella peruviana, Parmelia sp.*
2	Northwest	4675	*Festuca rigescens, Aciachne acicularis, Pycnophyllum kobalantum, Paronychia andina, Belloa sp., Senecio spinosus, Scirpus rigidus*
3	West	4342	*Festuca rigescens, Deyeuxia rigescens, Deyeuxia minima, Aciachne acicularis, Pycnophyllum kobalantum, Perezya pygmaea, Alchemilla pinnata, Plantago sericea, Azorella diapensioides*
4	Southwest	4564	*Festuca rigescens, Deyeuxia rigescens, Aciachne acicularis, Pycnophyllum kobalantum, Paronychia andina, Scirpus rigidus, Alchemilla pinnata, Selaginella peruviana*
5	Northeast	4502	*Festuca rigescens, Deyeuxia rigescens, Stipa ichu, Aciachne acicularis, Pycnophyllum molle, Pycnophyllum kobalantum, Senecio spinosus, Belloa sp, Scirpus rigidus, Alchemilla pinnata, Selaginella peruviana*
6	North	4552	*Aciachne acicularis, Nasella brachyphylla, Pycnophyllum kobalantum, Belloa sp., Scirpus rigidus, Selaginella peruviana*
7	South	4560	*Deyeuxia rigescens, Stipa sp., , Aciachne sp., Pycnophyllum kobalantum, Paronychia andina, Scirpus rigidus, Alchemilla pinnata, Selaginella peruviana*
8	Southwest	4890	*Festuca rigescens, Pycnophyllum kobalantum, Paronychia andina, Scirpus rigidus, Plantago sericea, Parmelia sp., Selaginella peruviana*

Different species could indicate disturbance in zones 1 and 2. In this way, *Senecio spinosus* is a thorny species described by some authors as indicator of disturbance in the Apolobamba grasslands due to the camelid grazing over-exploitation as well as *Aciachne acicularis* (García and Beck, 2006). Species such as *Scirpus rigidus* or *Plantago sericea* observed in zones 1 and 2 could also indicate a relative disturbance degree (García and Beck, 2006). Therefore, we suggest that zones 1 and 2 presented disturbed *Pycnophyllum* grasslands.

Plant coverage and species palatability

Researchers such as Beck et al. (2002) assessed that the plant coverage (PC) in the altoandine unit of the Apolobamba area were between 5 and 30 % in dry soils. On the other hand, Genxu et al. (2007) established high PC when its percentage was above 50 % at the steppes in the Tibet Plateau. Following these researchers, the studied zones presented high PC (above 50 %) except for zones 3 and 6, with medium PC (from 50 % to 30 %) (Fig. 1).

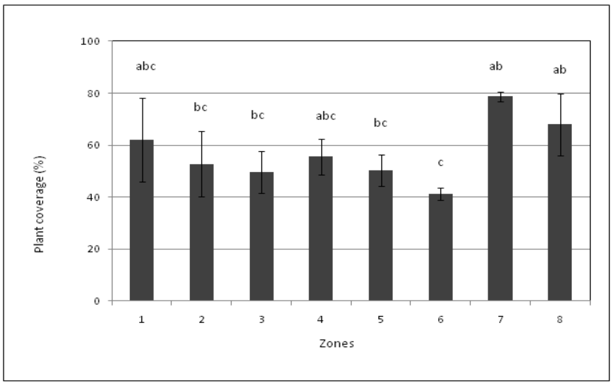

Figure 1. Plant coverage percentages. Mean value and standard error in each zone (n=3)

The correlation analyses pointed out positive correlation between soil properties and the PC, particularly with TOC, TN, CEC and exchangeable K^+ which suggests that soils with a higher fertility promote higher plant coverage.
With respect to those species more consumed by camelids or palatableness, a high number of palatable species were identified in all the studied zones. They were mainly grouped into the Poaceae family. Following to Borgnia et al. (2010), similar palatable genera for the vicuna were described in the Argentinean puna such as *Stipa*, *Festuca* or *Deyeuxia*. Other authors described that the *Stipa* and *Festuca* genera were highly consumed by the vicuna in Pampa Galeras, located in the Peruvian Andean steppes (Franklin, 1983). However, two unpalatable species described by García and Beck (2006) were observed in zones 1 and 2, *Senecio spinosus* and *Aciachne acicularis.* Also the unpalatable pteridophitas *Selaginella peruviana* and *Parmelia sp*., were identified in zone 1.

Effect of camelid population in soil properties and vegetation
Considering the relationships found between the vicuna and the alpaca densities and the soil parameters studied and taking into account the good fertility of the studied soils, we suggest that, in general, the soil properties and, therefore its fertility, was not affected by the camelid population. However, the high alpaca density, together with a high phosphorous concentration and low clay content reported in zone 1 could indicate that erosion process could be taking place associated with a frequent grazing of the alpaca population as Dorrough et al. (2006) established in other grasslands.
Zones with no disturbed *Pycnophyllum* grassland presented the highest vicuna density while the alpaca concentration was medium (zones 4, 5, 6, 7 and 8). Conversely, zones 1 and 2 presented disturbed vegetation with high alpaca concentration and low vicuna density. So, it could indicate the vicuna population preference for not disturbed *Pycnophyllum* grasslands. Additionally, we suggest the domestic camelid grazing influence in the original grasslands particularly in zones 1 and 2. The positive relationship between vicuna density and plant coverage indicates the vicuna preference for those grasslands with higher plant coverage. The high number of unpalatable species was observed in zone 1, with high alpaca and low vicuna densities, highlighting the substitution of palatable species by unpalatable ones. This point emphasizes the negative alpaca effect in the *Pycnophyllum* grasslands composition.

Conclusions
In general, the physico-chemical soil characterization exhibited the good soil fertility and no camelid grazing affection. However, erosive processes could be taken place particularly in one of the studied zones likely caused by the alpaca grazing. The botanical taxonomy pointed out that the most plentiful plant community was *Pycnophyllum* grassland although it was highly disturbed in two of the studied zones. The studied zones presented medium and high plant coverage which exhibited a positive relationship with the soil fertility. No vicuna affection in the soil fertility and the vegetation was observed. We suggest the vicuna preference for not disturbed *Pycnophyllum* grasslands with high plant coverage. The disturbed *Pycnophyllum* grassland and the substitution of palatable species by no palatable ones in zones with high alpaca concentration could indicate the negative domestic camelid effect in the original grassland composition. Therefore, it should be undertaken some protection actions to prevent the soil erosion, the changes in the grassland composition and the vicuna environment affection in the Bolivian Andean.

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Hydrocarbon Pollutants Biodegradation by Fungi

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Abstract

Oil spillage has a major impact on the ecosystem and organisms. One of the biological methods to clean up organic contaminants is Myco-remediation and fungus can be used to achieve this purpose. The most important role of fungi is decomposition of organic compounds from the simplest to most complex one in the soil. This microorganism decomposes light-weight hydrocarbons more rapidly than the heavy-weight hydrocarbons and they can also reduce heavy weight hydrocarbons to light weight hydrocarbons which are contained more harmful and toxic compounds such as asphaltene and resin in comparison to lightweight hydrocarbons. The high efficiency of fungi is due to having mycelium. Indeed, mycelium secrete acid and extracellular enzymes such as ligninolytic, lignin peroxidase, manganese peroxidase, which can break and convert Petroleum hydrocarbons as much as possible to non-toxic materials such as water and carbon dioxide and so far, different species of fungi depended on their efficiency are known to degrade petroleum in terrestrial and marine environments. In optimal conditions and Suitable population, native Fungus in comparison to non-native fungi will have more ability to remove oil contamination.
Key words: decomposition, fungi, hydrocarbon, mycelium, pollutant

Introduction

Spill oil pollutants have harmful effects on ecosystems and organisms. Therefore they are place in the group "life-anti compounds". Oil is a complex combination of chemical compounds that major part of it contains the hydrocarbons. This section are contains aromatic compounds, and aliphatic, asphaltene and resin.(Minai-Tehrani *et al*., 2006; Ebraimi *et al*., 2010). In natural environments, biodegradation of crude oil are produced material same to degradation of heavy oil. In such a case lack of saturated and aromatic hydrocarbons is a clear sign of crude oil biodegradation in ecosystem. Toxicity of hydrocarbons are increases by reducing number of carbon that bacteria and other microorganisms, are degradation large-chain hydrocarbons faster in the soil(Tabari *et al*., 2010). To remove petroleum contaminants are used from the physical, biological and chemical methods. Bioremediation technique, because of the possibility of soil reuse (Mancera-Lo´pez *et al*., 2008), and environmental sustainability is more considered than the mention two methods and also has superior economically (Ebraimi *et al*., 2010). Bioremediation is based on the capacity of microorganisms (including bacterial and fungal) to degrade organic pollutant compounds, such as hydrocarbons (Mancera-Lo´pez *et al*., 2008). Dating application bioremediation in environmental goes back to six hundred years BC which, the suggesting method of bioremediation began of the 1940s began (Ebraimi, 2010). In this regard, due to the widespread spread oil pollution and considering the adverse effects and secondary spill and transportation in the ecosystem, applied methods of cleaning up compatible with environmental is required for decontamination in each region (Ebraimi et al., 2010).

Mycoremediation

Mycoremediation is a form of bioremediation that application the fungal for clain up contaminated soil (Biiatt *et al*., 2002). Due to have myceliumu, fungi performed decomposition in environment very well. The mycelium break down lignin and cellulose that are two main building blocks of plant fiber (These are organic compounds composed of long chains of carbon and hydrogen, structurally similar to many organic pollutants) by secretes extracellular enzymes and acids. Mycelium easily degrades lower molecular weight hydrocarbons than heavier weight hydrocarbons (Gadd, G. 2001; Singh, H.

2006; Stamets, P. 2005). In year 1985, when the white-rot fungus Phanerochaete chrysosporium was dispousre, mycoremediation begin .these fungus are able to degrade a number of important environmental pollutants (Biiatt et al., 2002;) and because of their capable of degrading a wide range of PAHs are the most extensively studied species (Yucheng *et al*., 2008; Bogan *et al*., 1999; Collins *et al*., 1996; Pickard *et al*. 1999). Studies showed fungi such as Aspergillus sp., Trichocladium canadense, and Fusarium oxysporum to be degraded Low-molecular-weight PAHs and T. canadense, Aspergillus sp., Verticillium sp., and Achremonium sp. also has been degraded highmolecular-weight PAHs.(Haritash and Kaushik 2009). The factors that led to the fungi have been possible to the decomposition of petroleum hydrocarbons are including:

1- Fungi by using these compounds as carbon sources for growth perform oxidation reactions as a prelude to the detoxi fication and excretion of hydrocarbons (Prenafeta-Boldu *et al*., 2001; Cerniglia, White and Heich 1985).
2- This microorganisms Compared to most bacteria, have the ability to growin environments with low nutrient concentrations, low humidity and acidic pH (Mancera-Lo´pez *et al*., 2008). Some of the strains also can able to grow in condition low oxygen.(Haritash and Kaushik, 2009)
3- Fungi because of their ability to synthesize unspecific enzymes involved in cellulose and lignin decay that can degrade high molecular weight, complex and more recalcitrant toxic compounds, including aromatic structures, more interest. (Mancera-Lo´pez *et al*., 2008). Some of the fungi using intracellular cytochrome P450 enzymes for degrade PAHs (Yucheng *et al*., 2008).

So far, many studies have been on the role of fungi in the cleanup of oil pollution and many species have been identified. Leahy and Colwell reports Candida, Sporobolomyces, Aureobasidium, Rhodotorula and spp. are the most common marine isolates and Trichoderma and Mortierella spp. are the most common soil isolates. Aspergillus and Penicillium spp. have been frequently isolated from both environments that performed degradition of hydrocarbons. In study that conduct by Prenafeta-Boldu et al,.2001,five fungal strains including Cladophialophora, Exophiala , Leptodontium, Pseudeurotium ,zonatum were identified that are able to grow on toluene, also some of the strains grew on ethylbenzene and styrene.

Paper Braun-LuÈ llemann 1997, Observed 50% of the benzo[a]pyrene was removed by strains of Amanita excelsa, Leccinum versipelle, Suillus grevillei, S. luteus, and S. variegatus and 50% of phenanthrene also degraded by A. muscaria, Paxillus involutus, and S. grevillei.

The rate of removal of PAH increased when oxidation of these contaminants enhanced by filamentous fungi. That mycelial extension in soil enhanced contaminant degradation (Mancera-Lo´pez *et al*., 2008).

The number of indigenous bacteria reduce additional fungal to contaminant soil (Andersson *et al*. 2003; Yucheng *et al.*, 2008) or change the soil bacterial community (Yucheng *et al*., 2008; Corgie *et al*. 2006).

Discussion

Environmental conditions are effective on fungal activity for example cant active in a very salty environment. They are grows near-neutral pH but are more resistant to acidic PH than oil degrading bacteria (Tomas *et al*., 1998). Decomposition of petroleum hydrocarbons by fungi are because of their irregular structure of lignin, lignolytic that are secreted. oxygenase, dehydrogenase and lignolytic enzymes. The lignolytic system consists are secreted three Lignin peroxidase, manganese dependent peroxidase, phenoloxidases and H2O2-producing enzymes. Activity of the lignolytic fungi is extracellular and involved free radicals which first they are attack to the pollutant molecules and they are oxidized, soluble in water and also cause to these pollutants easier to be decomposed by bacteria (Haritash and Kaushik 2009).

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Determination of Optimal Conditions of Oil-eating Bacteria Activity on The Basis of Microbial Respiration Index

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Abstract
Extraction, refining and processing of petroleum products, is major part of industrial activities. On the other hand, spread of oil pollution in oil extraction and refineries area, is a serious threat to humans, ecosystems and environment. In recent decades, biological methods are considered as important management strategies because of their compatibility with the ecosystem. The purpose of this study was to compare microbial respiration of contaminated soil (as an activity Index of petroleum decomposition microorganisms) in different conditions. Hence, contaminated soil was brought from Ray's refinery vicinity and initial total hydrocarbon petroleum was measured (TPH). Then, different treatments were applied and soil microbial respiration rate was measured.Results from five periods showed that in the first, second and third periods after the establishment of treatments, most microbial respiration is related to sawdust treatments and then bacterial treatment over this time. But in the fourth period, in addition to increase in microbial respiration, bacterial treatment showed more correlation with the sawdust treatment. In the fifth period, bacterial treatment surpassed sawdust treatment while biological respiration rate was three times higher than the first one. Consequently, the establishment of bacterial community in soil containing sawdust (with the good aeration), with compatibility of oil-eating microorganisms showed that this treatment is the best treatment to use oil as a food source for the microorganisms over this time.
Keywords: Microbial respiration, Oil-eating bacteria, Optimization, Pollution

Introduction

Extraction, refining and processing of petroleum products, is major part of industrial activities, on the other hand, spread of oil pollution that they are among the most extensive environmental pollutants in oil extraction and refineries area is unavoidable (Ebrahimi, 2010). The accumulation of organic oily pollutants in environments is a serious threat to humans and ecosystems and since this compounds are containing small amount of nitrogen and phosphorus, raise ratio of carbon to nitrogen and carbon to phosphorus, that are not suitable for growth of microorganisms in these environments.(Abolhasani ; Ebrahimipour, 2008). Crude oil can be divided into four main sections, saturated hydrocarbons, aromatic hydrocarbons, resins and asphaltene (Coulon et al, 2005; Leahy, 1990) that number of carbon atoms of these compounds is 1 to 50. Light oil is composed of saturated and aromatic hydrocarbons and have low percentage of resin and asphalten but heavy oil that result of decomposition of crude oil in oxygen-free conditions In natural reservoirs, have amount less of saturated and aromatic hydrocarbons and it is more composed of polar compounds, resin and asphaltene. Saturated hydrocarbons are analysis first materials in the oil (Atlas, 1981, jabson 1972, Walker, 1976). But, resins and asphaltenes may not break down because of complexity in structure or are analysis in small amounts by cometabolism (Johnsen 2005; Shuttleworth 1995; Cerniglia1992; Cerniglia 1984). In natural environments, biodegradation of crude oil is created material similar to decomposition of heavy oil. In such a case lack of saturated and aromatic hydrocarbons associated with increased relative polar compounds is clear sign of biodegradation of crude oil. Generally, the decomposition of hydrocarbon (the largest part forming oil compounds), is the main process removal of oil pollution. However, aromatic and polar compounds with less percent have more stability and toxicity and require more time to decomposition. For cleanup of hydrocarbon contaminants is used of physical, chemical and biological methods (Ebrahimi et al,. 2010). In recent decades, cleanup oil pollution with biological methods, that used of the living organism for remove or detoxify environmental contaminants (Mohsrnzadeh et al,. 2010) are considered as important management strategies because of their compatibility with the ecosystem, speed up the decomposition process, the using simultaneous of physical and chemical methods, cost-effectiveness and analysis of hydrocarbon pollutants into nontoxic materials such as water and carbon dioxid (Ebrahimi et al,. 2010). This study was conducted in during five times on contaminated soils of Ray's refinery that located in suburban Tehran. This soil that passed long time of its contaminated, were analyzed with 38% of pollution in order to comparison of microbial respiration of contaminated soil (as an activity index of petroleum decomposition microorganisms) in the presence of different treatments including of organic and chemical fertilizers. Also was used to Bacillus subtilis, Bacillus megaterum and Pseudomonas putida bacterial for enhance the degradation of oil hydrocarbons.

Materials and Methods

Amount of oil-contaminated soil was collected from of the desired area. The soils were crushed and passed through a 2mm sieve after 2 days that they air-dried and 700 grams of soil was poured into each pot. Amount of total petroleum hydrocarbons measured was equivalent to 38%. Then different conditions were applied in each pot. In this experiment were two the dry and wet controls. The dry control have oil-contaminated soil without additives and was disturbed for aeration every 3 day and the wet control also was without additives too but with this difference that was added water to this control and was no aeration. To other pots, was added water and also was aerated (for the availability of oxygen needed of microorganisms) every 3 day. Inoculation of bacterial was performed the following;

Initially, Bacillus subtilis, Bacillus megaterum and Pseudomonas putida bacterial were prepared from Biology Department of Gorgan University. Then, it made sure; they have purified and were identified by staining (Gram staining). Each of the bacterial was cultured separately in liquid medium. After reaching to the certain time, 2/5 cc were taken of each culture medium containing bacterial and were sprayed to pots and for uniform distribution in the pots, they were blend with soil perfectly. Then amount and process degradation of the soil was measured and evaluated by using the microbial respiration index. Treatments were including:

1- dry control
2- wet control
3- A mixture of fertilizers, urea, triple superphosphate and potassium chloride with ratio of 20:5:1 in order to supply NPK
4- Animal manure with ratio of 5%
5- Sawdust with ratio of 10%
6- NPK fertilizer with animal manure
7- NPK fertilizer with sawdust
8- NPK fertilizer with sawdust and animal manure
9- Complex of bacterial

Results

The results are described as figure and fable during five periods. Table- 1 showed increasing microbial respiration rates over time in different treatments. According to table1, the highest rate of respiration can be seen in the complex bacterial (9) and sawdust treatment (5).

Table1 - comparison of oil reduction in different treatments at five periods.

Treatments	1	2	3	4	5	6	7	8	9
One period	0.041	0.048	0.022	0.047	0.055	0.029	0.017	0.028	0.048
Two period	0.058	0.051	0.025	0.057	0.066	0.034	0.035	0.037	0.066
Three period	0.059	0.052	0.042	0.065	0.079	0.037	0.042	0.051	0.067
Four period	0.06	0.063	0.046	0.102	0.124	0.05	0.045	0.054	0.124
Five period	0.112	0.105	0.049	0.122	0.15	0.057	0.058	0.088	0.15

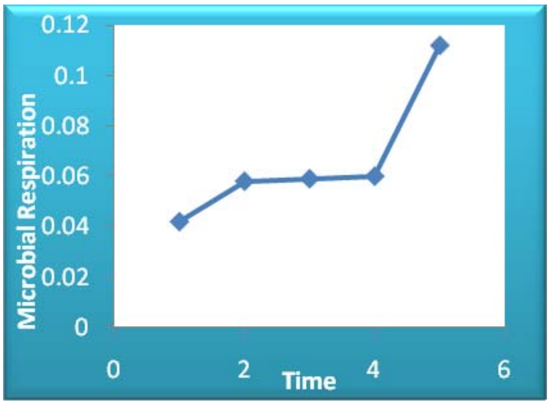

Figure-1- Microbial respiration in one treatment

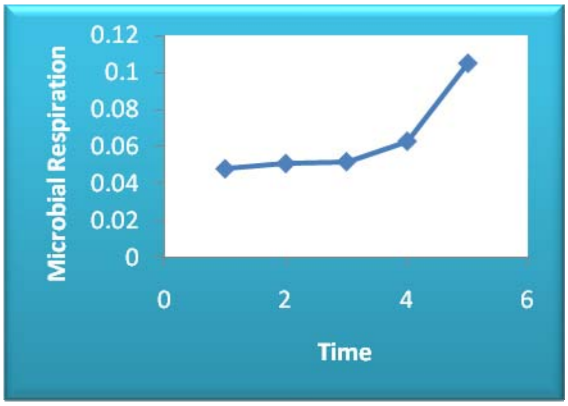

Figure-2- Microbial respiration in two treatment

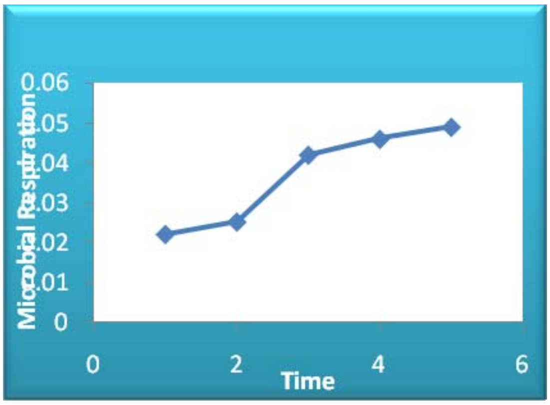

Figure-3-Microbial respiration in three treatment

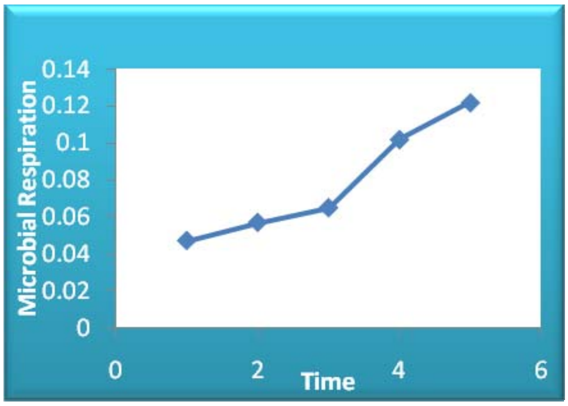

Figure-4- Microbial respiration in four treatment

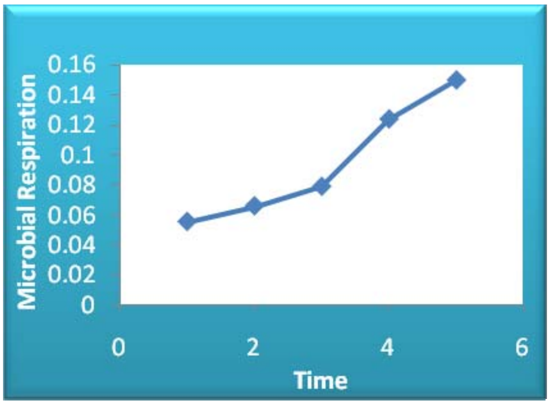

Figure-5- Microbial respiration in five treatment

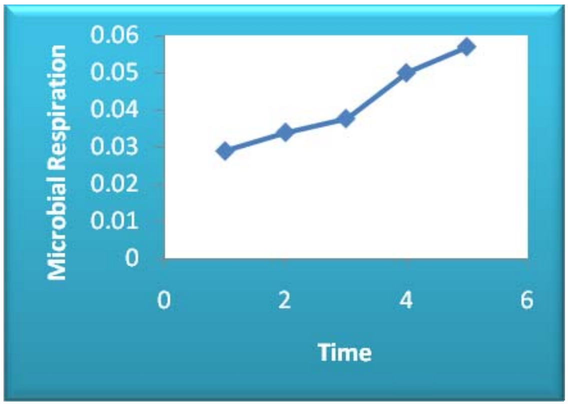

Figure-6- Microbial respiration in six treatment

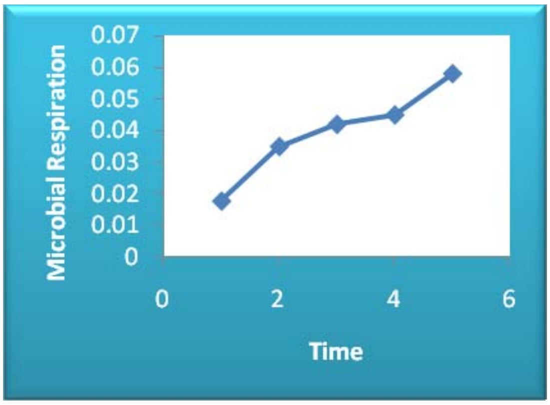

Figure-7- Microbial respiration in seven treatment

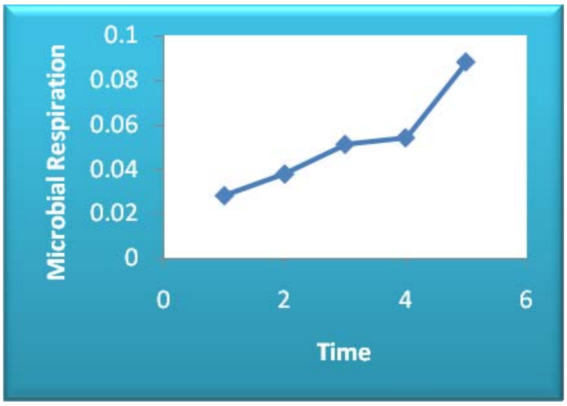

Figure-8- Microbial respiration in eight treatment

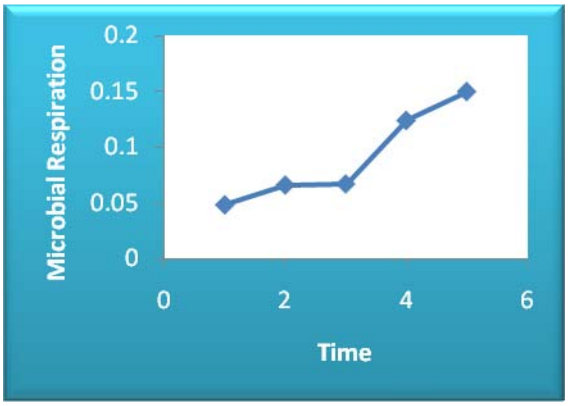

Figure-9- Microbial respiration in nine treatment

The processes of microbial respiration are ascending in all treatments during 5 periods. In fact over time respiration increased in all treatments. in early stages, respiration rate was higher in the sawdust treatment than other treatments but over time respiration increased in the bacterial treatment and in 4 and 5 period its respiration rates was same sawdust treatment. Microorganisms have been living in the soil that long time has passed after it infection (Ebrahimi et al., 2010). But due to the high percentage of pollution and lack of suitable conditions for growth, their activities are low. Therefore, are able to break the chains of petroleum hydrocarbons hardly in order to use the carbon in the oil. But activity of microorganisms is increased in sawdust treatment because of easy to use of the carbon present in the sawdust and microbial respiration also was increasing. This rise is indicative that microorganisms native of soil was stimulate and hydrocarbon chains break more easily by using the carbon in the sawdust and increased their ability. In Early stages of inoculation, respiration of the bacterial treatment is low. But respiration gradually increased with over time and compatibility bacterial with soil contaminated which indicate high strength of these bacteria in the degradation of petroleum hydrocarbons.

It is noteworthy, that native microorganisms of soil are activity with optimal growth conditions such as aeration and moisture needed which respiration increased will show this subject in control treatments (1 and 2). In research that by Labud (2007) was conducted in order to the impact of hydrocarbon pollutants on microbial population of the sandy and loam soil, to achieve this result that microbial respiration increased in crude oil contaminated soil. Also margesin (2000) was considered microbial soil respiration as an index for amount analysis of petroleum hydrocarbon contaminants in soil.

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Pollution of Natural Sources in Çanakkale Umurbey Plain

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Abstract
The Umurbey Plain is an important agricultural region of approximately 1700 ha area within the Lapseki district in Çanakkale province, Turkey. The main crops are fruits such as apple, peach, plum and cherry and, vegetables such as tomatoes and beans. Ground water wells are generally used for irrigation in this region. However, organic and heavy metal pollution from agricultural and industrial activities may have resulted in contamination of ground water. It is therefore important to assess the potential impacts that increasing agricultural and industrial activity may be having on this vital resource. In this study 47 water and 38 soil samples were analyzed from artesian wells to accurately determine the extent of ground water contamination. The pH values of water samples were between 7.24 and 8.18, and electrical conductivity (EC) values were between 617 µmhos cm^{-1} and 5390 µmhos cm^{-1}, and as much as 35000 µmhos cm^{-1} in some samples affected by proximity to the sea. The total heavy metal content of water samples were found to be within tolerable limits. However, the total heavy metal content of some of soil samples, particularly those close to the Umurbey Creek, were found to be at dangerous concentrations. This study found evidence for widespread organic and heavy metal pollution in the soils and ground water of the Umurbey Plain. While ground water levels are not currently at levels that would be harmful to public health, future expansion of agricultural and industrial activities co-incident with increased economic growth may result in future challenges. We therefore recommend continued monitoring of contamination levels in soils and ground water alongside programs to reduce pollutant discharge.
Keywords: Umurbey, water, soil, pollution, heavy metal

Introduction
In recent years, Umurbey town in Çanakkale province, has developed important export-oriented agricultural production, particularly of cherries and peaches. The main crops are fruits such as apple, peach, plum, cherry and vegetables such as tomatoes and bean. Cereal crops are cultivated in areas with high relief beyond the irrigation system of Lapseki. Although vegetable cultivation is not very common, tomato cultivation stands as a main vegetable production. Irrigation is largely provided from deep wells in the region. Umurbey Creek is active during the winter months but mostly dry during the summer season. However during the summer season irrigation water is provided by the wells drilled in the dry Umurbey Creek. In winter months the flow of Umurbey Creek rises, and occasionally overflows, damaging agricultural land in the Umurbey flood plain. In this study, soil and water samples were taken from from wells in different seasons in Umurbey Creek, the flood plain around the Creek and remote areas located away from the Creek.

Materials and Methods
In this study, soil and water samples were collected from the Umurbey Plain which covers approximatelly 1700 ha where intensive fruit cultivation occurs. The climate in this area is arid with relatively low humidity, and the soil has a greater soil water surplus in winter months and less water during summer season. According to the water balance of this region, evopotranspiration is greater than rainfall in the summer, and in winter rainfall exceeds evopotranspiration. Plant water requirements in spring are met by water stored in the soil over the winter season. However, in May many plants require supplementary irrigation (Yiğini, 2006). In this study 47 water, 38 soil samples were analysed. Thirteen water samples were taken from Umurbey Creek and others were taken from different of irrigation well depths (Figure 1). Soil samples were taken from different depths of the soil series indicated in Yigini (2006) in the Umurbey Creek area. The coordinates of sampling points were determined by the Global Positioning System (GPS). Parameters of the quality of irrigation water were analysed, and the total heavy metal contents of water and soil samples were also analysed. Soil samples were digested with Aqua regia to investigate total heavy metal contents using ICP-AES spectroscopy. The pH values of water samples were measured with an Orion 420

pH meter and Electrical Counductivity (EC) was determined using a LF 320 EC meter (Richards, 1954).

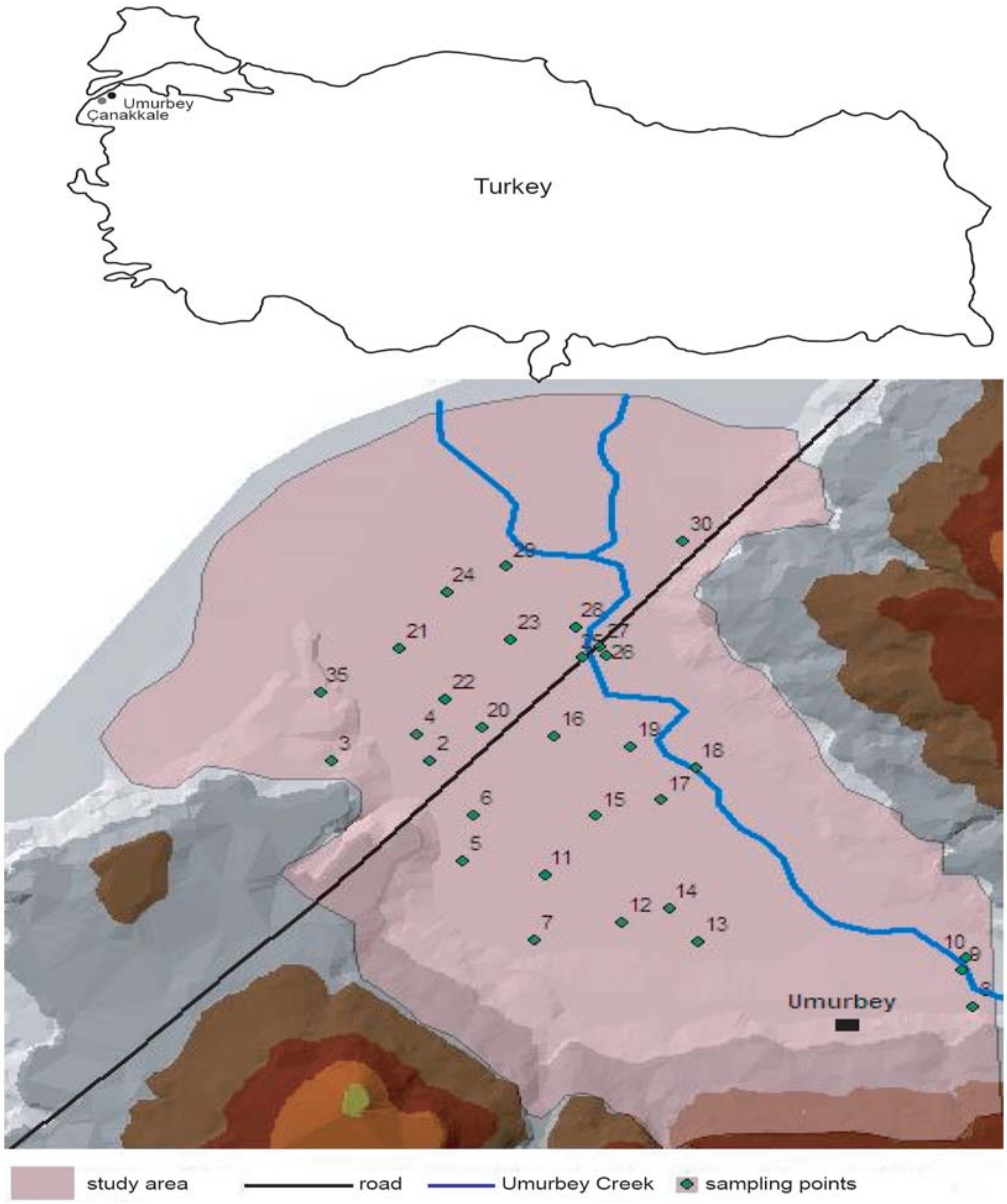

Figure 1. Points for water samples

NO_3-N and NH_4-N content were determined using the titration method of Mulvaney (1996). Ca^{+2}, Mg^{+2}, $CO_3^{=}$, HCO_3^{-}, CL^{-} were also analysed using the titration method (Soil Survey Staff, 1996), Boron was determined using the method of Keren, (1996) and the total heavy metals (Fe, Cu, Pb, and Zn) content was determined using ICP–AES spectroscopy. SO_4 content was determined gravimetrically (Soil Survey Staff, 1996; Merck,1982).

This survey of water samples in the Umurbey Plain involved approximately 30 wells used for irrigation in September 2004 and May 2005. Alongside these samples, some parameters of water samples from Umurbey Creek and irrigation canals were also investigated (Table 1). In these water samples the total heavy metal content (eg. Fe, Cu, Pb and Zn) was measured and the results are presented in Table 2.

Table.1. Chemical Analyses Results of Water Samples were taken in September 2004 (S) and May 2005 (M)

Sample Number	Season	pH	EC µS/cm	Ca+Mg mEq/l	CO_3 mEq/l	HCO_3 mEq/l	Cl mEq/l	NO_3 ppm	NH_4 ppm	Na ppm
2	S	7,29	1693	13,6	0	22,8	16	4,9	10,51	138,8
Çatalazmak	M	7,49	1636	23,6	0	5,2	9	17,51	10,5	76,33
3	S	6,82	8440	48	0	16,4	87	2,1	40,63	118,55
Çatalazmakucu	M	7,44	2770	33,6	0	4,6	25	66,5	19,26	144,62
4	S	7,87	1856	16,2	0	18,4	9	41,33	42,73	143,78
Çatalazmak Santral	M	7,52	1687	19	0	6,4	8	26,27	12,26	75,27
5	S	7,3	2400	19,6	3,2	23,4	13	21,02	33,62	230,37
Akçeşme Bridge	M	7,25	3290	31,2	0	8,2	19	21,02	10,51	189,62
6	S	7,29	2160	17,4	0	23,6	15	17,51	21,72	203,55
Petrol Station	M	7,08	2800	25	0	7,6	16	36,78	21,02	202,86
7	S	6,85	1647	16	3,2	13,6	8	9,81	9,81	133,08
Y.çayırı Glass House	M	7,36	1340	22	0	6,6	7	29,77	14,01	77,39
8	S	7,19	1256	9,8	0	11,4	9	8,41	2,8	94,42
Entrance of Gökköy Gçmk	M	7,06	1294	18,4	0	4,6	9	47,28	14,01	63,09
9	S	7,61	1159	8,6	0	9,6	9	10,51	1,4	98,89
Gökköy Gçmk Bridge	M	7,61	1420	15,6	0	4,4	1	24,51	17,51	44,4
10	S	7,2	1076	10,4	0,8	10,8	6	4,2	1,4	52,13
Gökköy Gçmk Creek	M	*	*	*	*	*	*	*	*	*
11	S	7,15	1526	14,8	0,8	24,6	9	8,41	1,4	98,89
Koşuyolu	M	7,25	1641	26,8	0	7,4	10	35,03	10,51	60,98
12	S	7,36	1316	13,6	0	18,2	8	2,1	0	89,82
Koşuyolu-UB Roundabout	M	7,2	1218	22	0	6,2	7	22,7	14,01	51,98
13	S	7,66	1470	16,2	0	19,4	10	7,01	2,1	62,42
Umurbey	M	7,23	1344	25	0	6	9	19,26	19,26	56,74
14	S	7,18	1580	12	0	18,4	10	14,01	3,5	32,07
Harmantarla	M	7,17	1510	25	0	6,2	3	33,27	15,76	56,21
15	S	7,38	1601	10	0	17,8	9	4,9	3,5	98,9
Behind of Petrol Stat.	M	7,35	1173	19,8	0	6,4	4	35	17,51	57,3
16	S	7,2	1580	10,4	0	17,2	10	4,9	2,1	103,46
Bekirağa Mvk. A. Öner	M	7,29	1293	20,2	0	6,4	7	21,01	14,01	60,98
17	S	7,37	1491	12,4	0,8	14,4	8	3,5	2,8	71,24
Profil Number 10	M	7,41	1452	28,2	0	6,4	10	21,02	21,02	58,86
18	S	7,21	1099	10	1,2	13,2	7	5,6	1,4	48,81
Profil Number 10	M	7,19	1072	18	0	4,4	8	21,02	10,51	49,86
19	S	7,51	1299	10	0,8	15,2	8	4,2	2,8	94,42
Körübaşı Environs	M	7,2	1478	19,2	0	6,8	9	12,258	15,76	63,09
20	S	7,41	1786	9,4	2	15,2	11	4,2	5,6	178,4
Kovukçınar	M	7,49	2210	29,8	0	6	8	14,01	19,26	66,27
21	S	7,1	15720	32	0	11	162	2,1		23866
Opposite of Çatalazmak	M	7,06	7310	67,2	0	6,4	58	21,015	12,6	261,09
22	S	7,41	2100	8,4	0	16	15	2,1	2,8	184,53
Opposite of Petrol Stat.	M	7,2	2690	23	0	6	13	22,76	21,01	155,21
23	S	7,08	2400	16,2	0	16,8	10	2,1	10,51	117,8
Kovukçınar	M	7,22	2220	39	0	4,8	7	14,01	14,01	66,8
24	S	7,23	2260	16,6	0	13	17	2,1	8,41	108,14
Kovukçınar through the sea	M	7,26	2040	39,2	0	6,8	10	24,51	12,26	67,8
25	S	6,81	9890	41,4	0	10,6	104	3,5	2,1	*
UB köprü deniz t.	M	7,81	7810	12,8	0	3,4	7	22,56	1,51	50,39
26	S	7,46	34300	24,2	0,8	13,2	378	3,5	3,5	*
UB Bridge	M	*	*	*	*	*	*	*	*	*
27	S	7,96	38600	24,4	1,2	9,8	422	4,2	14,01	*
UB Bridge	M	*	*	*	*	*	*	*	*	*
28	S	7,96	1496	16	0	16,2	24	3,5	3,5	329,02
UB Bridge 250 m.	M	7,15	3300	31,2	0	4,8	30	19,26	10,51	157,85
29	S	6,96	3360	19	0	10,6	38	3,5	2,1	127,88
Between UB Bridge - Sea	M	7,09	2390	33,4	0	5,8	16	12,26	15,76	70,5
30	S	7,61	3720	18,2	0	12,6	33	3,5	2,8	416,86
Kemiklialanaltı	M	7,26	2900	29,2	0	6,4	19	29,77	10,51	160,50
31	S	6,94	18940	53,6	0	26,4	206	2,1	2,1	*
Hotel	M	*	*	*	*	*	*	*	*	*
32	S	7,6	14500	35,2	0	21,8	151	2,1	2,1	*
Fener Plain	M	*	*	*	*	*	*	*	*	*
33	S	7,88	778	16,4	0	16,6	8	2,1	2,8	24,84
Tabaklar	M	*	*	*	*	*	*	*	*	*

34	S	7,47	758	16,2	0	23,4	7	4,2	1,4	22,32
H.Güven	M	*	*	*	*	*	*	*	*	*
35	S	6,97	1247	20,4	0	16	8	5,6	2,1	*
G.Burun	M	7,22	2140	27,8	0	5,8	10	29,77	14,01	72,62
K1	S	7.11	3690	10.6	0	14.2	32	2.80	2.80	398.46
Kemiklialan	M	-	-	-	-	-	-	-	-	-
K2	S	7.08	13600	11	-	11.6	9	81.26	3.50	57.08
Koruköyiçi	M	-	-	-	-	-	-	-	-	-
K3	S	7.32	550	8.4	0	10.4	6	2.10	3.50	34
Koruköy Dere	M	-	-	-	-	-	-	-	-	-
K4	S	7.56	511	11	0	6.4	4	2.10	2.80	43.25
Akçaalan D.geç.	M	-	-	-	-	-	-	-	-	-
K5	S	7.23	1066	12	0	14.2	6	4.20	2.80	67.23
Gökköy Gçmk	M	-	-	-	-	-	-	-	-	-
K6	S	7.08	1173	5.2	0	2.2	1.4	-	-	20257
UB- Bridge(23.12.2004)	M	-	-	-	-	-	-	-	-	-
K7	S	7.36	1293	4.2	0	2.2	1.2	-	-	71.55
UB-Bridge (30.12.2004)	M	-	-	-	-	-	-	-	-	-
K8	S	7.06	1452	1.4	0	2.4	1.9	-	-	64.56
UB-Bridge (16.02.2005)	M	-	-	-	-	-	-	-	-	-
K9	S	7.61	1478	4.4	0	2.4	1.2	-	-	42.55
UB-Bridge (20.02.2005)	M	-	-	-	-	-	-	-	-	-
K10	S	7.22	1218	12.2	0	2.0	1.1	-	-	69.62
UB- Bridge (02.03.2005)	M	-	-	-	-	-	-	-	-	-
K11	S	7.52	1344	5.6	0	2.8	1.0	-	-	72.56
UB-Bridge (14.04.2005)	M	-	-	-	-	-	-	-	-	-
K12	S	7.25	1510	4.0	0	4.2	1.3	-	-	180.63
UB-Bridge (25.05.2005)	M	-	-	-	-	-	-	-	-	-

* Non –determined.
UB: Umurbey

GUIDELINES FOR INTERPRETATIONS OF WATER QUALITY FOR IRRIGATION					
Potential Irrigation Problem		**Units**	**Degree of Restriction on Use**		
			None	**Slight to Moderate**	**Severe**
Salinity*(affects crop water availability)*					
	EC_w	dS/m	< 0.7	0.7 – 3.0	> 3.0
	(or)				
	TDS	mg/l	< 450	450 – 2000	> 2000

(FAO 1989, 1994)

As seen in Table 1, most of the water samples' Electrical Conductivity (EC) values are between 700–3000 µmhos cm^{-1} . Ayers, and Westcot (1985) indicated in the FAO Irrigation and Drainage Paper (FAO 1989, 1994), that according to irrigation water quality criteria, degree of restriction on use of these water EC limits are slight to moderate. Especially in the production of some fruits and vegeteables such as bean, tomato, pepper, apple and apricot., where their yield potential is reduced by 50% at EC values of 2000 and 3000 µmhos cm^{-1} in irrigation water (Ayers and Westcot, 1976). However some other water samples have EC values of around 30000 µmhos cm^{-1} EC due to the mixing of sea water in the Umurbey Creek during the summer season. Due to the effect of sea water, wells in the Catalazmak location also have high EC values; therefore these waters are not suitable for irrigation purposes. Irrigation waters in the Tabaklar location are extremely favorable (EC 758 µmhos cm^{-1}) for use in irrigation. Well waters in the sea side location of the Lapseki-Çanakkale main road are also unsuitable for irrigation due to their proximity to the sea. In terms of the Cl content of waters, after the EC measurements, it was found that waters taken near to Umurbey have acceptable levels of low and moderate Cl content. However in seaward areas, the Cl content was found to be above 10 mEq/L which is at the harmful level. NO_3 values in water samples, taken in May, were found higher then others. This is because of increased nitrogen fertilisation during the spring season. It was found that in most water samples (32 samples) , NO_3 –N concentrations were between 5-30 ppm, and therefore classed as mild to moderate. NO_3 –N concentrations in 26 of water samples were below 5 ppm which is considered to be innocuous for irrigation water, however 21 water samples exhibited values above 30 ppm, which is not acceptable irrigation water quality. The sample (K2) taken from wells in Korukoy has a high NO_3 content with 81.26 ppm, suggesting the possibility of localised problems in terms of public health. This high NO_3 content is probably due to contamination into the water wells from animals feeding and manure within the village. The impact of Na on the water wells is generally moderate. Approximately half of the water samples analyzed exhibit mild and moderate HCO_3 content. Boron content in the analysed waters is within acceptable limits, and below the upper limit for all water samples (Table 2). According to National Academy of Sciences (1972) and Pratt (1972), with the exception of a few water samples, all of the water sample heavy metal contents (Cu, Fe, Pb and Zn) are below tolerance limit values. Hence in terms of irrigation these heavy metal contents should not cause any problem (Table 2).

Table 2. Total Heavy Metal Contents of Water Samples

Sample Number	Season	Parameters (ppm)				
		Cu	Fe	Pb	Zn	B(mEq/L)
2	M	0.027	0.317	0.020	0.105	0.514
Çatalazmak	S	1.442	0.997	1.283	1.940	-
3	M	0.043	0.368	0.030	0.105	0.399
Çatalazmakucu	S	0.049	0.398	e	2.446	-
4	M	0.021	1.076	0.032	1.454	0.451
Çatalazmak Station	S	0.051	0.370	0.042	0.773	-
5	M	0.022	0.709	0.038	0.080	0.560
Akçeşme Bridge	S	0.028	0.491	e	0.385	-
6	M	0.027	1.921	0.031	0.235	0.476
Petrol Station	S	0.410	0.291	0.032	0.536	-
7	M	0.021	0.924	0.005	0.254	0.579
Y.çayırı Glass House	S	0.027	0.321	e	0.258	-
8	M	0.025	0.522	0.025	0.062	0.354
Entrance of Gökköy Gçmk	S	0.025	0.440	0.018	0.681	-
9	M	0.025	0.238	e	0.346	0.332
Gökköy Gçmk Bridge	S	0.049	0.238	e	0.127	-
10	M	-	-	-	-	-
Gökköy Gçmk Creek	S	0.025	0.186	0.066	0.230	-
11	M	0.027	1.054	0.050	0.344	0.397
Koşuyolu	S	0.058	0.779	e	0.114	-
12	M	0.023	0.538	0.015	0.142	0.455
Koşuyolu-UB Raundabout	S	0.042	0.400	0.107	1.162	-
13	M	0.020	0.390	0.029	0.958	0.382
Umurbey Alti	S	0.023	0.400	e	0.264	-
14	M	0.027	0.654	0.023	0.245	0.334
Harmantarla	S	0.032	0.982	e	3.234	-
15	M	0.027	0.853	0.015	0.551	0.507
Petrol Station	S	0.020	0.958	e	0.905	-
16	M	0.022	0.416	0.041	2.120	0.388
Bekirağa Mvk. A. Öner	S	0.039	0.974	e	0.594	-
17	M	0.028	1.443	0.029	3.499	0.529
10 nolu profil	S	0.045	0.133	0.104	0.419	-
18	M	0.029	0.500	0.023	0.831	0.362
Profil Number 10	S	0.051	0.268	0.014	0.652	-
19	M	0.025	0.362	0.029	0.066	0.334
Körübaşı Environs	S	0.029	0.252	e	0.188	-
20	M	0.024	0.809	0.023	0.095	0.292
Kovukçınar	S	0.042	0.148	e	0.502	-
21	M	0.035	0.492	0.055	0.199	0.581
Opposite of Çatalazmak	S	0.047	0.212	0.069	0.391	-
22	M	0.021	0.768	0.022	0.045	0.430
Opposite of Petrol Stat.	S	0.042	0.180	0.044	0.434	-
23	M	0.026	1.161	0.057	0.130	0.389
Kovukçınar	S	0.032	0.302	e	0.556	-
24	M	0.024	1.028	0.061	0.042	0.396
Kovukçınar through the sea	S	0.095	0.325	0.116	0.540	-
25	M	0.022	0.563	0.029	0.374	0.246

UB köprü deniz t.	S	0.045	0.235	0.050	0.669	-
28 UB Bridge 250 m.	M	0.087	0.817	0.077	0.124	0.494
	S	0.043	0.333	0.046	0.389	-
29 Between UB Bridge-Sea	M	0.071	5.796	0.049	0.344	0.284
	S	0.038	0.133	e	0.626	-
30 Kemiklialanaltı	M	0.025	1.260	0.008	0.981	0.463
	S	0.033	0.217	e	1.303	-
K1 Kemiklialan		0.040	0.280	e	1.660	-
K2 Koruköyiçi		0.040	0.270	0.040	0.680	-
K3 Koruköy Creek		0.030	0.430	e	0.720	-
K4 Akçaalan D.geç.		0.040	0.330	0.005	0.650	-
K5 Gökköy Gçmk		0.070	0.210	e	0.360	-
K6 UB-Bridge (23.12.2004)		0.134	1.708	0.033	1.280	-
K7 UB-Bridge (30.12.2004)		0.077	0.780	0.040	0.090	-
K8 UB-Bridge (16.02.2005)		0.130	1.670	0.060	0.090	-
K9 UB-Bridge (20.02.2005)		0.090	0.990	0.070	0.090	-
K10 UB-Bridge (02.03.2005)		0.160	1.780	0.060	0.180	-
K11 UB-Bridge (14.04.2005)		0.030	0.410	e	0.110	-
K12 UB-Bridge (25.05.2005)		0.090	1.280	0.100	0.270	-

The heavy metal contents of soil samples are below the hazardous limits set out by Basar et all. (2004), Fe; 5000- 50000 ppm and Scheffer ve Schachtschabel (1989) total limits of Zn; 300 ppm, Cu; 100ppm and Pb; 100 ppm, hence (Table 3). However, silty loam soil samples taken from Akçaalan, samples from Umurbey Creek, samples taken from around Umurbey bridge and also samples taken from 100 m before estuary of Umurbey Creek were determined to possess heavy metal contents that exceed these limit values. This may be due to mines operating in the entrance of Umurbey.

Table 3.Heavy Metal Contents of Soil Samples in Umurbey Plain (ppm)

Soil Samples		**Cu 324,755 nm**	**Fe 259,941 nm**	**Pb 220,354 nm**	**Zn 213,857 nm**
0 Umurbey	Çayı denize yakın	47,12	14897,88	441,22	1158,64
*1Gökköy G.	Köprü 0-20 cm	67.31	10454.00	772.35	4679.08
2 Gökköy G.	Köprü 20-40 cm	26,70	22924,96	37,58	141,95
3 Umurbey	Köprü 0-20 cm	60,99	23290,60	188,11	1010,73
4 Umurbey	Köprü 20-40 cm	34.00	13905,88	82,50	468,03
5 İki çay yat.	Arası 0-20 cm	45.67	23529.60	28.44	75.55
6 İki çay yat.	Arası 20-40 cm	100,40	22596,64	32,108	106,24
7 Akçaalan	Değirman Geç. 0-20	26,26	3459,88	578,96	762,50
8 Akçaalan	Değirmen Geç. 20-35	115,22	12662,00	2146,44	3153,48
9 Akçaalan	Değirmen Geç 35-55	198,23	11655,12	1195,11	-
10 Akçaalan	Değ Geç. Siltli Balçık	114,62	8765,36	2492,43	3274,20
11 Akçaalan	Değirmen G. Balçık	40,14	4531,12	1025,15	1339,06
12Koruköy	Koruköy Köprüsü	29,16	22585,84	90,80	141,812
13 Yılancık ç.	Çayırı Ap 0-30 cm	53,80	14378,56	16,74	49,27
14 Yılancık ç.	Çayırı A2 30-57 cm	126,44	22700,08	2004,04	3297,91
15 Kokarkuyu	Ap 0-18 cm	28,83	10259,60	15,04	33,45
16 Kokarkuyu	A2 18-44 cm	18,50	8362,04	11,63	27,96
17 Karaçayır	Ap 0-12 cm	28,22	17945,28	13,50	59,10
18 Karaçayır	Ad 12-44 cm	26,24	19518,20	16,43	60,47
19 Yemşen	Ap 0-32 cm	24,70	14201,80	13,37	47,55
20 Koşuyolu	Ap 0-34 cm	43,90	14199,28	14,54	45,62
21 Koşuyolu	A2 34-58 cm	89,33	15232,40	19,83	56,49
22Harmantarla	Ap 0-23 cm	67,65	23103,40	20,52	71,99
23Harmantarla	A2 23-57 cm	27,68	16153,84	14,92	48,50

24 Çaykenarı	Ap 0-20 cm	70,72	25893,32	28,38	94,10
25 Çaykenarı	A2 20-36 cm	37,66	16036,88	14,02	43,56
26 Geçemek	Ap 0-12 cm	98,10	22192,32	26,97	78,74
27 Geçemek	A2 12-35 cm	49,16	17345,12	21,74	55,58
28Kovukçınar	Ap 0-21 cm	40,28	19476,52	22,07	58,17
29Kovukçınar	A2 21-53 cm	50,64	24984,48	24,59	74,84
30Çıkrıkçıkuyu	Ap 0-25 cm	61,13	25682,76	26,94	78,10
31Çıkrıkçıkuyu	A2 25-60 cm	35,72	21502,84	20,43	60,36
32 Otel	Ap 0-22 cm	31,96	16125,28	28,54	73,98
33 Otel	A2 22-60 cm	38,70	22097,16	25,82	62,66
34 İşletme	0-20 cm	55,41	20768,36	23,70	72,62
35 Umurbey	Köprü 30.12.2004	134,03	10136,56	2100,20	3319,48
36 Ubey Köp.	Taşkın tarlası20.02.2005	52.38	8261.40	395.05	1314.60
37 Umurbey	Köprü altı 20.02.2005	47.57	7785.00	704.08	1714.82

*Samples with number of 1-34 were taken in August 2004.
nm: nanometer (wavelength of ICP spectroscopy)

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Effect of Gasoline on microbial respiration on silty clay and loamy sand soil

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Abstract
Petroleum hydrocarbons are one of the important environmental pollutants. It can move into soil body and change the soil properties and quality of ground water and then threaten the safety of human and other living organism. The aim of this study was to investigate the effect of gasoline on two type of soils (silty clay and loamy sand) microbial respiration with different properties. By this regard, two soils samples were chosen and polluted by gasoline in amount of half and one volume of soil porosity. Then, the soils were maintained under controlled conditions, after reaching to equilibrium, every month, microbial respiration was measured. The result showed, gasoline affect the soil microbial respiration very slightly. In fact, in both soils, gasoline cause little changes on soil respiration at both rate (0.5, 1 volume porosity of soil) in comparison to unpolluted soil.

Keywords: gasoline, petroleum hydrocarbon, pollution, respiration, soil

Introduction
Petroleum hydrocarbons are one of the important environmental pollutants. It can move into soil body and change the soil properties and quality of ground water and then threaten the safety of human and other living organism. Since the middle of the 19 the century, petroleum has been used for many decades, for illumination and, on a smaller scale, as lubricant. The invention of the internal combustion engine and its fast adoption in all transport forms enlarged the employment of this natural resource, thus increasing its demand, production, transport, stockpiling, and distribution, as well as the raw oil, and its by-products. All these activities involve pollution risks that can be minimized, but not totally eliminated, causing several problems for the environment (Pala et al 2006). Even though petroleum-degrading microorganisms are widely distributed in soil and water, they may not be present in sufficient numbers to achieve contaminant remediation (Joo et al., 2006). One of the best approaches to restoring contaminated soil is to make use of microorganisms able to degrade the toxic compounds in a bioremediation process. However, it is known that hydrocarbon biodegradation in soil can be limited by many factors, for example: microorganism type, nutrients, pH, temperature, moisture, oxygen, soil properties, and contaminant concentration (Ghazali et al., 2004; Walter et al., 2005; Semple et al., 2001; Bardi et al., 2000; Sabate´ et al., 2004). Iran is one the oil producing country in the world. So, soil pollution and water pollution by hydrocarbon product isn't new issues and demands a lot of attention to consider and solve this problem as serious one. Hence, the aim of this work was to determine the effect of gasoline on two type of soil (silty clay, loamy sand) microbial respiration.

Material and Methods
In this study, tow type of soils (loamy sand and silty clay) were chosen and then the mount of half and one volume of soil porosity, gasoline has applied and the soils were maintained under controlled conditions for several month. After reaching equilibrium, microbial respiration was measured every month. During this period of time, gasoline concentration in soil also was measured according to U.S. Environmental Protection Agency (EPA, 1992). Tow gram from polluted soil were weighed and added 10 ml dikoloromethan on soil. The mixture was shacked well and put it on the shaker. Then Surface solution which was separated from soil was put on the glassy tin that has weighed before. This process was repeated for three times and let the dickloromethan evaporate in the room temperature and then the rest of gasoline content was measured. This amount of gasoline showed the concentration of gasoline in the soil in the certain time during this period of time.

Results

Some properties of soils were determined and the results gathered in table 1 as below

Table 1: some Soil Properties

	Sample 1	Sample2
Texture	Loamy sand	Silty clay
Bulk density	1.57	1.26
Particle density	2.5	2.35
Porosity	37%	47%
Organic carbon	0.26%	1.22%
Organic matter	0.44%	2.10%
pH	7.47	7.73
EC(micro z/cm)	1300	1121

The results for soils respiration from gasoline polluted soils showed in figure 1 and 2 for loamy sand and silty clay soil. Both figure showed that gasoline has increased microbial respiration in silty clay and sandy soils but its effect on microbial respiration wasn't so significant especially in loamy sand soil. In fact, gasoline has affected the soil microbial respiration very slightly and in both soils, gasoline cause little changes on soil respiration at both rate (0.5, 1 volume porosity of soil) in comparison to unpolluted soil.
As you see in the figure 1 and 2, comparison of tow soils showed one volume of gasoline cause higher stimulant for soils respiration especially in silty clay soil in comparison to blank soils.

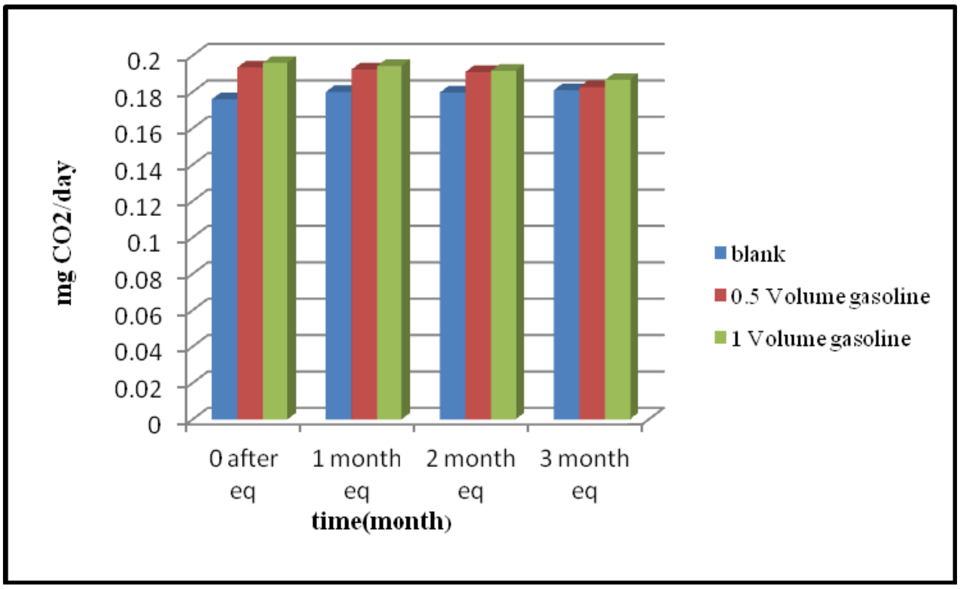

Figure 1- comparison of soil respiration for one and half volume of gasoline in the loamy sand soil

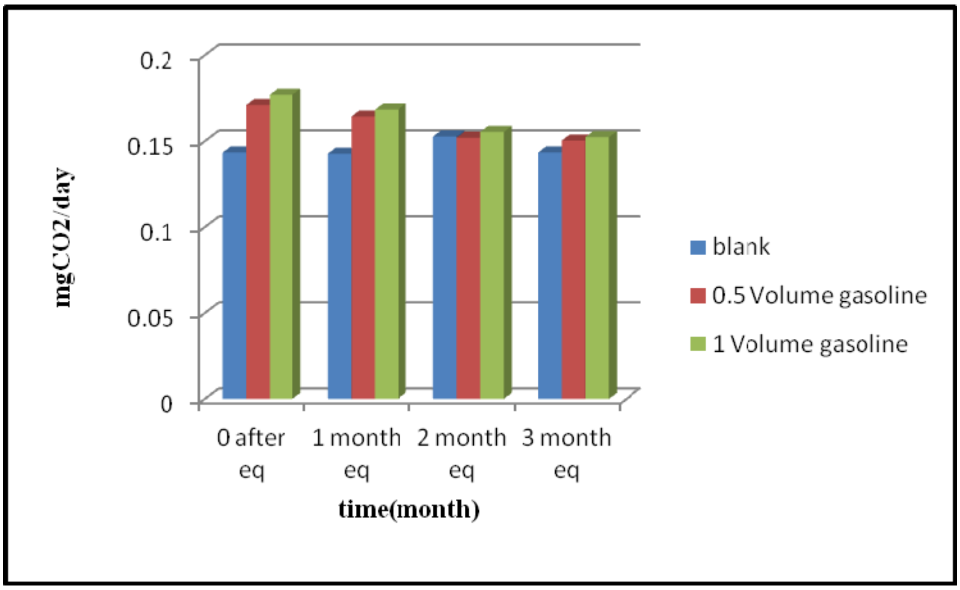

Figure 2- comparison of soil respiration for one and half volume of gasoline in silty clay soil

Figure- 3 showed gasoline concentration in soil during these months. As you see in the fig-3, gasoline concentration has decreased in the course of time and most reduction for gasoline concentration was related to silty clay soil with applied one volume of soil porosity of gasoline.

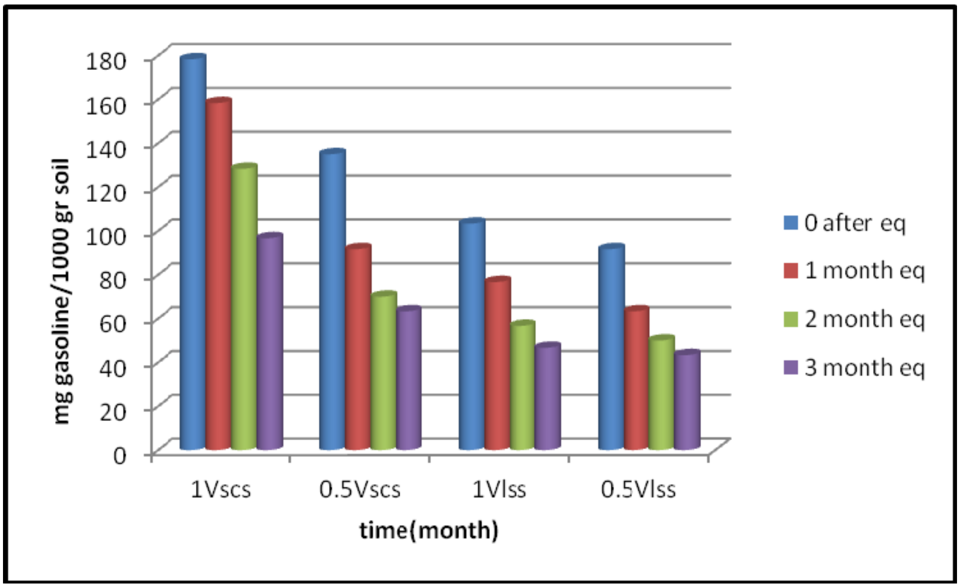

Figure 3- gasoline concentration for one and half volume of gasoline in soils

1V scs: 1 volume porosity of gasoline in the silty clay soil
0.5V scs: 0.5 volume porosity of gasoline in the silty clay soil
1V lss: 1 volume porosity of gasoline in the loamy sand soil
0.5V lss: 0.5 volume porosity of gasoline in the loamy sand soil

Discussion

The result showed there was a reduction in gasoline concentration in the course of time and had a slight increase in soil respiration, especially in one volume of soil porosity of gasoline in silty clay soil in comparison to blank soils and other polluted soils and this reduction in gasoline concentration and increase in soil respiration can be related to microorganism population and their activity beside other effective factors on soil remediation and microorganism activity.

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Effect of kerosene on two type soil respiration

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Abstract
Petroleum hydrocarbons are one of the most widespread pollutants in soil habitats due to the increased usage of petroleum products and Hydrocarbons have toxic effect on plants and living microorganisms and invertebrates. Hence, the aim of this work was to determine the effect of kerosene on two type of soil (silty clay, loamy sand) microbial respiration. For this, the mount of half and one volume of soil porosity, kerosene has applied and the soils were maintained under controlled conditions. After reaching equilibrium, microbial respiration was measured every month. The results showed kerosene increased microbial respiration in both soils. But kerosene caused higher soil respiration rate in clay soil at both rate (0.5, 1 volume porosity of soil) than sandy soil. On the other hand, higher amount of kerosene produce higher respiration rate in both soils and in the loamy sandy soil, the differences between soil respiration at both rate is more than silty clay one. Increase in Microbial activity can be because of microorganism population increase in soil which refers to usage of hydrocarbons as a carbon resource. It is probable most part of these bacteria's population is native oil eating bacteria.
Keywords: kerosene, petroleum hydrocarbon, pollution, respiration, soil

Introduction
Petroleum hydrocarbons are one of the most widespread pollutants in soil habitats due to the increasing usage of petroleum products and Hydrocarbons have toxic effect on plants and living microorganisms and invertebrates. In industrialized countries, contamination of soil by crude oil and petroleum products has become serious problem. The main sources of this contamination are: oil field installations, petroleum plants, liquid fuel distribution and storage devices, transportation equipment for petroleum products, airports and illegal drillings in pipe lines. The scale of the hazards imposed on the natural environment depends on the surface of the area contaminated by petroleum products, their chemical composition, and the depth at which pollutants occur (Wolicka et al., 2009). Crude oil and petroleum products contain many kinds of organic compounds, dominated by aliphatic and aromatic hydrocarbons (Fukui et al., 1999).
Oil products, including petrol not only modify physico-chemical (Tyczkowski., 1993) and biological properties of the soil (Borowiec et al., 1982; Lebkowska et al., 1995; Malachowaka-jutsz et al., 1997; Olanczuk-neymanL., 1994) but also contribute to limitations of the productive ability of arable crops. It is known that these compounds are able to affect the quality of surface and ground water and these products are potentially dangerous for animal and human health. Various microorganisms are able to use some fractions of crude oil as their sole carbon source and transform them into non-toxic compounds (Minai tehrani et al., 2006). Numerous microbes present in soil contaminated by oil hydrocarbons are able to grow, despite the high toxicity of these compounds. The ability to degrade and/or utilize oil hydrocarbons has been observed in numerous types of bacteria and fungi, and in yeast e.g. Candida, Saccharomyces (Bento and Gaylarde, 2001;Prenafeta-Boldu et al., 2002).
Hence, the aim of this work was to determine the effect of kerosene on two type of soil (silty clay, loamy sand) microbial respiration.

Methods and material
In this study, tow type of soils (loamy sand and silty clay) were chosen and then the mount of half and one volume of soil porosity, kerosene has applied and the soils were maintained under controlled conditions for three month. After reaching equilibrium, microbial respiration was measured every month. During this period of time, soil hydrocarbon content also was measured according to U.S. Environmental Protection Agency (EPA, 1992). Tow gram from polluted soil

were weighed and added 10 ml dikoloromethan on soil. The mixture was shacked well and put it on the shaker. Then Surface solution which was separated from soil was put on the glassy tin that has weighed before. This process has repeated for three times and let the dickloromethan evaporate in the room temperature and then the rest of oil content was measured. This amount of oil showed the concentration of oil in the soil in the certain time during this period of time.

Results

Some properties of soils were determined and the results gathered in table 1 as below

Table 1: Some Soil Properties

	Sample 1	Sample2
Texture	Loamy sand	Silty clay
Bulk density	1.57	1.26
Particle density	2.5	2.35
Porosity	37%	47%
Organic carbon	0.26%	1.22%
Organic matter	0.44%	2.10%
pH	7.47	7.73
EC(micro z/cm)	1300	112

The results for soils respiration showed in figure 1 and 2 for loamy sand and silty clay soil. Both figure showed that kerosene increased microbial respiration in silty clay and sandy soils. By increasing amount of pollutant (oil), soils respiration has increased more. But kerosene caused higher soil respiration rate in silty clay soil at both rate (0.5, 1 volume porosity of soil) than sandy soil in comparison to blank soils. On the other hand, higher amount of kerosene produce higher respiration rate in both soils and in the loamy sandy soil, the differences between soil respiration at both rate in comparison to blank soil is more in silty clay soil.

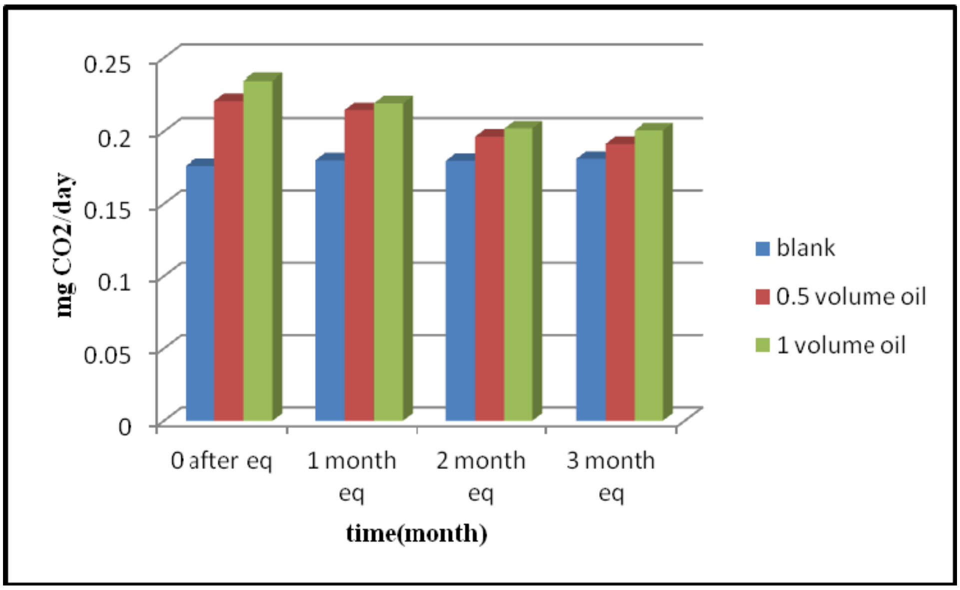

Figure 1- comparison of soil respiration for one and half volume of oil in the loamy sand soil

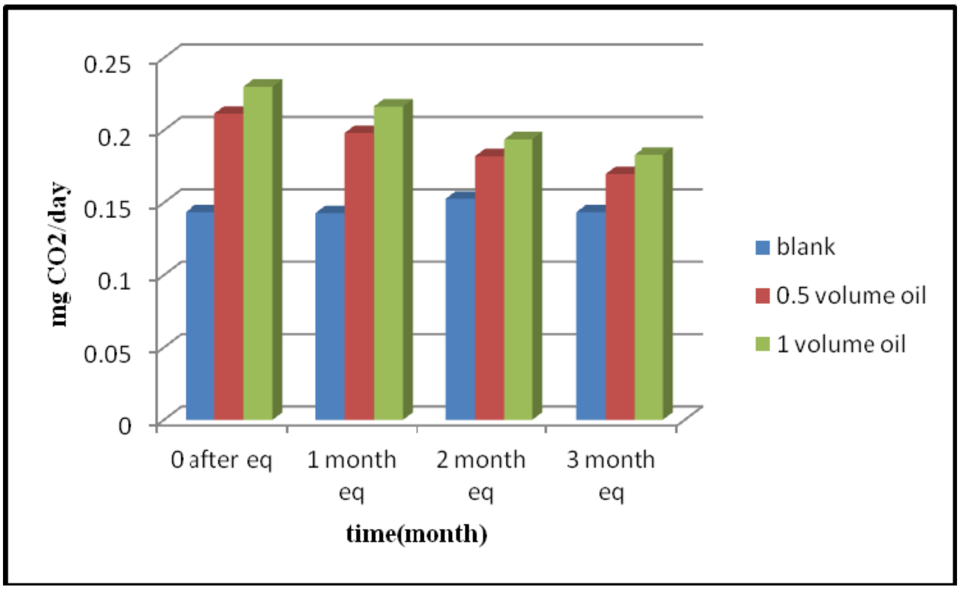

Figure 2- comparison of soil respiration for one and half volume of oil in silty clay soil

Figure- 3 showed oil concentration in soil during these months. As you see in the fig-3, oil concentration has decreased in the course of time and most reduction for oil content was related to silty clay soil with applied one volume soil porosity of oil.

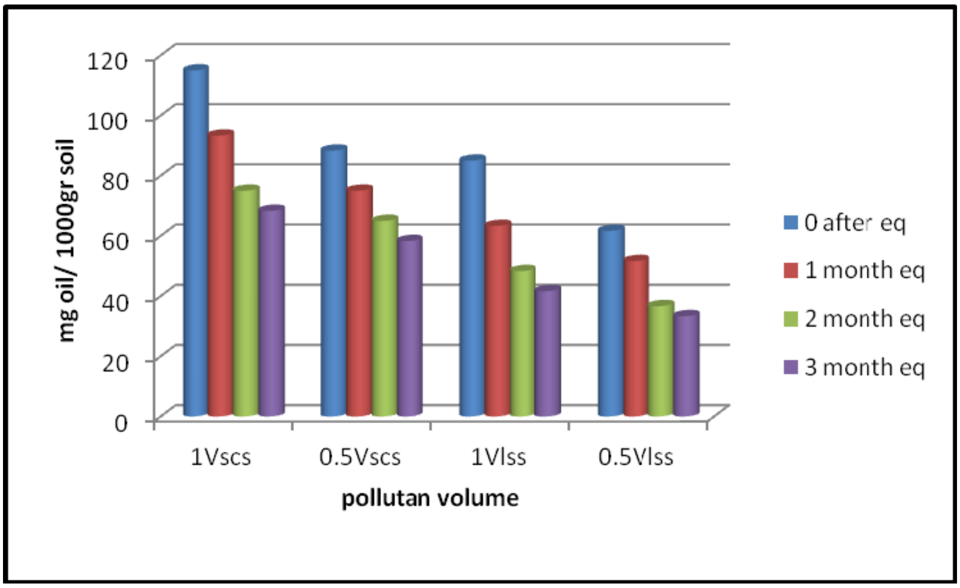

Figure 3- oil concentration for one and half volume of oil in soils

1V scs: 1 volume porosity of oil in the silty clay soil
0.5V scs: 0.5 volume porosity of oil in the silty clay soil
1V lss: 1 volume porosity of oil in the loamy sand soil
0.5V lss: 0.5 volume porosity of oil in the loamy sand soil

Discussion

An increase in Microbial activity can be because of microorganism population increase in soil which refers to usage of hydrocarbons as a carbon resource. It is probable most part of these bacteria's population is native oil eating bacteria. More volume of oil results in more soil respiration and greater reduction during the time by these bacterias.

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Comparison of Gasoline and Kerosene Transport in the Subsurface, a Laboratory Soil Column Study

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Abstract

Oil and fuels, are the most frequent type of pollutants of soil and water in the world. In this regard definition, the purpose of this research was comparison and study of the transportation of gasoline and kerosene in the loamy sand and silty clay soil textures, in the pilot scale and pulse condition. Hence some equipment including glassy cylinders with special outlet was designed. Then, bulk density and porosity of soil were determined and one-half and one volume porosity of soil, gasoline and kerosene was applied. Glassy cylinders were filled with soil homogeneously in the given depth and then soil was saturated by water. In this stage of procedure, half and one volume porosity of these pollutants have applied on saturated soil. Eventually, output volume of gasoline, kerosene and water per time was measured and Breakthrough curves were drawn. The results showed that in the sandy and clay soil, gasoline retention was more than kerosene. Kerosene flowed out of light and heavy soil by sharper gradient and less retardation. This difference in the pollutants output rate from sandy soil is more distinct than heavy one. The reason is that reactivity of gasoline with soils is more than kerosene which is probably because of structural form and its carbonic chain number and on the other hand, more intense surface absorption.
This study demonstrates on a small scale, that Comparison of transportation of gasoline and kerosene in the soil, are useful tools for prediction the possible leakage of pollutants in oil polluted soil.
Keywords: Breakthrough curve, Gasoline, kerosene, Soil column, Transportation

Introduction

Soil contamination is a growing environmental problem that involves the introduction of unwanted chemicals or organic compounds into the ground (Jacqueline et al., 2007). Petroleum hydrocarbon is one of the most widespread pollutants because of increasing growth of oil, gas, petro-chemical industrial and product process, transportation, transmission. Pollution by petroleum compounds has been a major concern since the mid 1960s in all over the world. Oils and petroleum are ever present pollutants in the modern environment. Oil due to solubility, volatility and bio-degradability of petroleum compounds has harmful effects on soil and water quality. On the other hand, hydrocarbon pollutant with several negative environmental effects cause changes on physical and chemical properties of soil, pollutes water reservoirs, farmland (Ebrahimi et al.,2009; Marins Pala et al., 2006; Mohan S et al.,2007). The oil pollution makes the soil conditions unsatisfactory for plants threaten their quality and human safety (DeJong, 1980). In Industrial countries, pollution by hydrocarbon product is considered as a serious problem and Iran is one the oil producing country in the world that a large amount of oil is extracted especially from southern part of Iran every year and refine in different part of Iran. So, release and leakage of hydrocarbon product during the process of extraction, transportation, refinery, result in soil and environment pollution that causes environmental problem. In order to solve this problem, satiable strategies should be chosen. Undoubtedly, these strategies are depended on comprehensive knowledge of current condition and facilities.
Then the aim of this study was to comparison and study of the transportation of gasoline and kerosene in the loamy sand and silty clay soil textures, in the pilot scale and pulse condition and soil porous media behavior in respect to transportation of these pollutant.

Materials and Methods

In this research two type of soils (loam sandy and silty clay) and tow type of samples, kerosene and gasoline were chosen and applied separately on the light and heavy soil in the pilot scale and pulse condition. Hence the equipment including glassy cylinders with the filter and special outlet in the bottom of these has been designed and the bulk density and porosity of soil has determined (Table 1). Then Glassy cylinders were filled with soil homogeneously in the given depth and then at first soils have been saturated by water. In this stage of procedure, the mount of half and one volume porosity of soil, gasoline and kerosene has applied separately on saturated soil. Eventually, output volume of gasoline, kerosene and water per time has measured and breakthrough curve were drawn.

Results

Table 1: Some Soil Properties

	Sample 1	Sample2
Texture	Loamy sand	Silty clay
Bulk density	1.57	1.26
Particle density	2.5	2.35
Porosity	37%	47%
Organic carbon	0.26%	1.22%
Organic matter	0.44%	2.10%
pH	7.47	7.73
EC(micro z/cm)	1300	1121

The result of gasoline and kerosene from loamy sand soil showed in figure-1 as below.

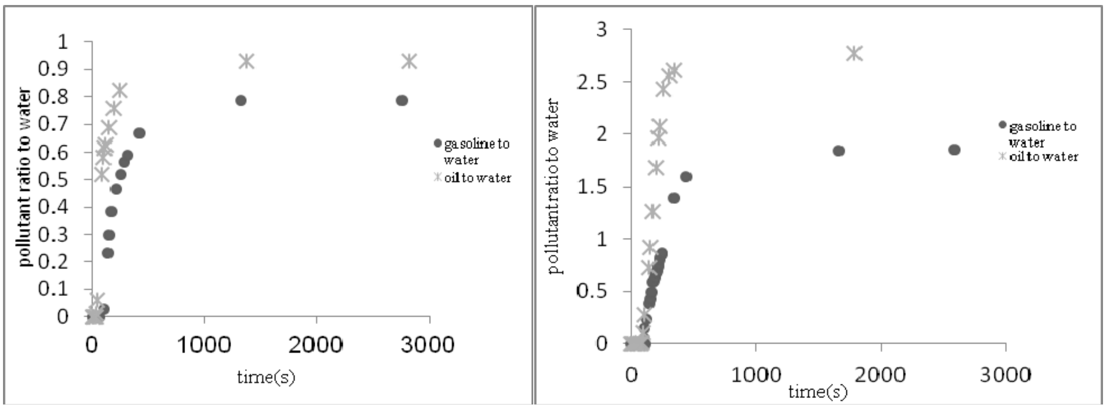

Figure 1- output volume of gasoline and kerosene to water from loamy sand for and one volume of -half (left) porosity of soil(right)

The result from gasoline and kerosene breakthrough curve in light saturated soil column showed that gasoline retention was more than kerosene. Kerosene flowed out of light soil by sharper gradient and less retardation. In the same time, the comparison of kerosene and gasoline transportation showed that the output rate of kerosene is more than gasoline especially in one volume application of pollutant. The reason is that reactivity of gasoline with light soil is more than kerosene which is probably because of structural form and its carbonic chain number and on the other hand, more intense surface absorption.

The result of kerosene and gasoline from silty clay showed in figure-2 as below.

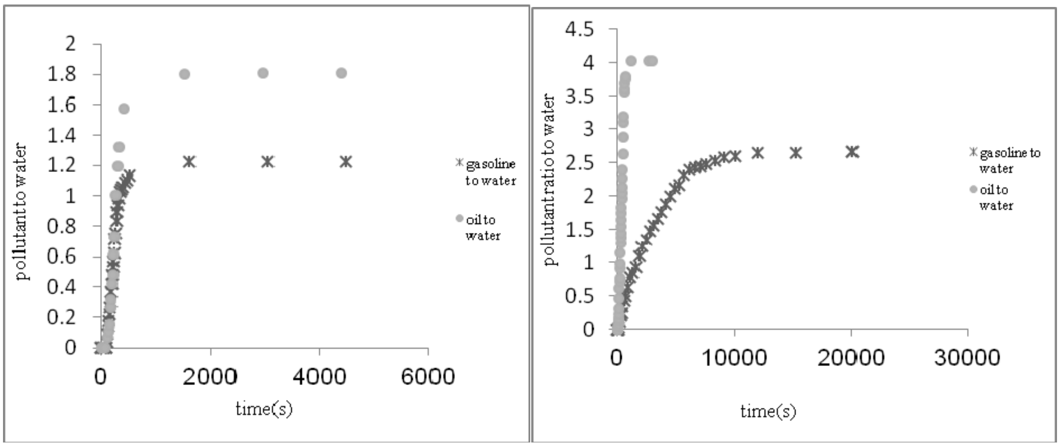

Figure2- output volume of kerosene and gasoline to water from silty clay soils for half (left) and one volume of porosity of soil(right)

The results showed that in the silty clay soil, gasoline retention was more than kerosene and Kerosene flowed out of silty clay soil by sharper gradient and less retardation and higher velocity. On the other, in the same time, kerosene to water output ratio was more than gasoline to water ratio and both graphs showed the same procedure in half and one volume of pollutant.

Discussion

This experiment showed in the both soils, movement of gasoline was slower than oil and its retardation in the soils was more than kerosene. The reason is that reactivity of gasoline with light and heavy soils is more than kerosene which is probably because of structural form and its carbonic chain number and on the other hand, more intense surface absorption.

This study demonstrates on a small scale, that Comparison of transportation of gasoline and kerosene in the soil, are useful tools for prediction the possible leakage of pollutants in oil polluted soil.

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Nitrate movement in irrigated fields
The case of Kopais Plain. Greece.

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Abstract
The intense agricultural activities, especially the utilization of high doses of fertilizers significantly change the natural ecosystem and may lead to a rapid deterioration of the quality of water resources. Intensive crop production is associated to high risk of nitrate leaching. The present study aims to investigate the movement of fertilizers, especially of nitrates, in soil and its effect on surface water quality in Kopais plain. In this area, with the drainage/irrigation system, a shallow water table can be observed below the soil surface at the irrigation period. In irrigated fields with the main cultivated crops of the region, the concentrations of nutrients in soil solution at different depths were determined. The amounts of precipitation and applied irrigation water were recorded. The depths of water table beneath the fields were recorded and its concentration in nutrients was measured. Water quality in the adjacent ditch was also determined for the same periods. From the results obtained, it was obvious that even when the soil solution and the water beneath the fields presented high values of nutrients concentration, the concentration of nutrients in the ditch water was substantially lower. This may indicate that although agricultural practices impose a pressure, the quality of surface water was not seriously affected.
Keywords: soil solution, fertilizer, nitrate movement

Introduction
Human activities have altered the global biogeochemical cycle of N, the rate of nitrogen input into the terrestrial N cycle has approximately doubled and expected to increase further. The global production of agricultural fertilizers increased from less than 10 million metric tonnes of N in 1950 to 80 million metric tonnes in 1990 (Galloway et al., 1995; Vitousek et al., 1997). Nitrogen is released to the environment from point and non point sources. Agriculture is the largest source of N, other sources being septic tanks, livestock wastes, municipal landfills. One of the major environmental problems is nitrate losses from agricultural areas to surface and groundwaters. The excessive input of phosporus and nitrogen can lead to highly undesirable changes causing eutrophication in freshwater and marine ecosystems (Smith, 1999). Nitrate ions are highly soluble in water and since they are not adsorbed by clay complex and humus are mobile and leach to deeper soil layer with percolating water (Addiscott, 1991; Jury and Nielsen, 1986) contaminating surface and groundwater. Drinking water limit for nitrate concentration is set at 50 mg/l (E.C., 1998; WHO, 2006) Nitrate is the end product of the reactions converting other forms of nitrogen in soil. It can be removed from the soil either by plant uptake or by denitrification. Denitrification is the biological transformation of NO_3^- to gaseous N. Anaerobic conditions and a carbon source for the denitrifying bacteria are the prerequisites for denitrification which can occurs in very wet soils or inside soil aggregates at high moisture content (Jury and Nielsen, 1986). Nitrate losses from leaching are largely a function of soil characteristics, environmental conditions, land use, type, timing and amount of nitrogenous fertilizers and agricultural management practices (Grant et al., 1986; Russelle and Hargrove, 1986). It is a crusial issue to manage the agricultural system in such a way as to increase the fertilizer efficiency by maximising plant uptake and minimising losses to waters.

Kopais plain is an intensively cultivated area with significant water resources. Irrigation water is provided by the surface waters, Kifisos and Melas rivers, through the system of canals and ditches that drain the region. The quality of Kifisos and Melas rivers can be directly affected by the quality of shallow groundwater and the leaching of nitrates from the fields. Also the basin's surface outflows account for most of the inflow into Lake Yliki, one of the three main reservoirs of Athens for drinking water. This study aimed to examine the leaching potential of nitrates and evaluate the effect of agricultural activities and especially of nitrogenous fertilisers to the quality of surface waters. For 3 years the concentrations of nitrates in soil solution, in the shallow groundwater and in the adjacent ditch was determined in two sites under the common agricultural practices of the region to assess the actual impact of applied fertilisers.

Materials and methods

The Kopais plain, an area of 240 km^2, is located in Central Greece, at Eastern Sterea Ellada region, belongs to Boeotia Prefecture and it is one of the most intensively cultivated areas of the country. Kopais Plain has been created by the drainage of Kopais Lake at the end of 19th century. The sub-basin of Kopais plain is situated at the end part of the downstream basin of Boeoticos Kifisos. The network of canals (Inner Canal, Central Canal and Collector Canal) and ditches draining Kopais plain are presented in Figure 1. Boeoticos Kifissos river flows through Kopais plain in the artificial riverbed created during the drainage of the lake and through Karditsa tunnel flows into Lake Yliki. During its flow receives the waters of several smaller rivers, the most important of which are Herkynas, Lofis and Falaros. Melas river, fed mainly by Chariton springs, the most important springs of the region, and from Polygyra springs also, partly canalised flows along the north side of Kopais and discharges at the beginning of the Collector canal. The basin of Melas river has an area of 316 km^2. Other significant springs are Herkynas springs, Mavroneri springs and Lofi springs (Pagounis M. et al, 1986).

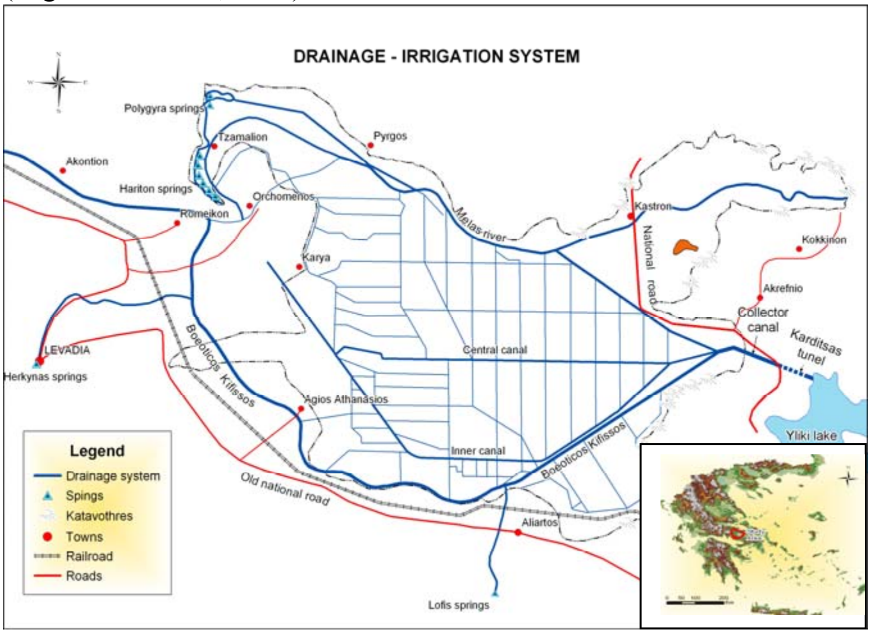

Fig. 1: Drainage system of Kopais Plain

The main crops cultivated in Kopais plain includes cotton (8500 ha), cereals (4370 ha), alfalfa (3800 ha), corn (3000 ha), vegetables (2000 ha) especially tomatoes (1400 ha). The irrigated land is 16000 ha. (Organisation of Kopais) Irrigation water derives from Kifisos, Melas and the main springs of the region. The network of open canals/ditches is used to provide also irrigation water to the fields. Water level in the open canals/ditches is controlled by mechanical gates. The same network of open canals/ditches is used for drainage. In the past subirrigation method was used but now irrigation water is applied by sprinklers or guns. In spring the runoff of Kifisos and Melas to the lake Yliki is almost eliminated and during approximately two months, in the summer, water from Yliki is transferred to the canals/ditches by pumping to maintain the appropriate water level in the ditches.

In two experimental sites (A) and (B) representative of the pedological and hydrological conditions of the region the common crop rotation and actual farming practices were followed. The main physical and chemical characteristics of the soil in the experimental sites were determined. In Table 1 the main characteristics of soil at Site (A) and (D) were presented. Experimental fields are rectangular in shape (200 m x 500 m) with a size of 10 ha approximately. The cultivated crops were in site (A) wheat (8/12/92-3/7/93), corn (14/4/94- 28/8/94) and corn (19/4/1995- 26/8/1995) and in site (D) cotton (26/4/1993- 14/10/1993), wheat (9/12/1993-27/6/1994), corn (9/7/1994-

3/11/1994) and cotton (22/4/95- 28/11/95) over the monitoring period. The amount, type and timing of fertilization were also recorded. Irrigation was applied at the experimental sites by guns. Rainfall and irrigation heights were recorded in each field by raingauges.

In the centre of each field a pit was dug and ceramic suction cups were installed at different depths at each site to extract the soil solution. Suction cups were installed at different layers, at site (A) at depths of 25, 36, 48, 60, 74, 90, 105 and 128 cm and at site (D) at depths of 23, 44, 56, 64, 88, 95 and 125 cm. Groundwater level fluctuation during the monitoring period was measured periodically and water samples were taken through a tube installed in the centre of each field. EC and nitrate concentration of soil solution and of groundwater samples were measured. Sampling positions of groundwater in site (A), and (D) were in a distance of approximately 100 m, and 700 m respectively away from the inner ditch. The sampling position in site (D) is approximately in the middle of the distance between the inner ditch and Kifisos river. Water samples from the inner ditch were taken periodically and determinations of EC and NO_3^- were made. Nitrate concentration of water samples were measured by the cadmium reduction method (APHA, 1989) in a double beam spectrophotometer. Water samples were also taken at the outlet of surface waters, after Karditsa tunnel and determinations of EC and NO_3^- were made.

Table 1 Physical and chemical characteristics of soil in the experimental fields.

Soil layer	Depth (cm)	Sand	Silt	Clay	$CaCO_3$	Organic matter	Bulk density	Ks	Porosity
		%	%	%	(%)	(%)	(gr/cm^3)	(cm/h)	% v/v
SITE A									
1	0-25	21.2	28.6	50.2	65.5	4.422	1.080	2.25	53.21
2	25-34	19.6	27.8	52.6	67.6	4.250	1.180	0.63	50.87
3	34-46	35.0	13.0	53.0	72.2	3.350	1.120	0.83	52.20
4	46-70	17.4	14.2	68.4	77.9	1.072	1.130	9.13	52.02
5	70-81	15.6	14.0	70.4	76.6	1.407	1.140	10.04	54.75
6	81-107	15.1	17.8	67.1	81.9	1.310	1.110	1.62	55.09
7	107-144	20.9	24.6	54.5	63.0	0.536	1.353	8.71	48.56
8	144-151	28.0	24.0	48.0	53.4	0.938	1.491		
9	151-160	36.7	18.2	45.1	59.5	5.590	1.082	9.75	55.35
10	160-230	17.7	36.8	45.6	44.3	0.603			
11	230-	14.3	34.0	51.7	34.4	0.871			
SITE D									
1	0-20	47.5	16.5	36.0	30.4	8.040	0.933	39.5	56.87
2	20-35	28.6	18.1	53.3	30.8	7.705	0.933		58.09
3	35-43	30.5	6.9	62.6	73.6	2.546	0.890	8.83	55.41
4	43-55	23.4	9.9	66.7	60.8	3.551	1.010	67.45	55.65
5	55-63	91.9	3.3	4.7	8.8	0.402	1.447	68.83	38.36
6α	63-90	92.8	4.0	3.2	7.0	1.675	1.310	65.17	39.89
6β	90-115	94.3	2.5	3.2	7.2	0.469	1.300	156	41.96
7	115-121	49.7	34.0	27.4	22.0	1.608			
8	121-	20.4	40.6	39.0	37.2	0.536	1.383	0.08	47.01

Results and discussion

The rainfall and irrigation heights, the cultivated crops and the depth of water table from the soil surface at each site during the monitoring period are presented in Fig. 2. The depth of groundwater table was varying during the year depending on the amount of rain, the depth of water table in the neighbouring irrigation/drainage ditch and the distance of each site from it, the applied irrigation heights and the crop evapotranspiration. Fluctuation of water table during the year in site D was smaller than in site (A). Water table in autumn obtained its deepest level. In the first year (1993) due to the very dry conditions prevailing during the previous year and the significant shortage of available water the applied irrigation heights were significantly small.

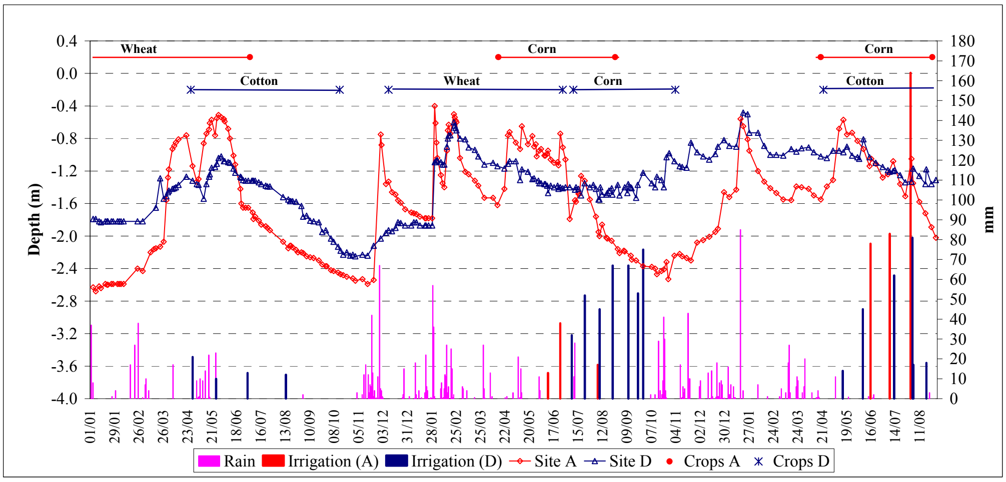

Fig. 2: Rainfall, irrigation heights, cultivated crops and water table depths at sites (A) and (D).

The mean, minimum and maximum values of nitrate concentration of soil solutions extracted by ceramic suction cups at different depths for the period 1993-1995 are presented in Table 2. Samples were taken whenever soil moisture is high enough to allow the extraction of soil solution.

Table 2: NO_3^- concentration of soil solution at experimental sites (A) and (D).

Site A				**Site D**			
Depth (cm)	NO_3^- (mg/l)			Depth (cm)	NO_3^- (mg/l)		
	Mean	Min	Max		Mean	Min	Max
25	184.27	2.06	638.20	23	295.51	76.30	541.60
36	244.94	6.05	671.75	44	325.58	54.02	819.50
48	186.23	6.05	612.75	56	344.05	37.30	601.00
60	149.69	20.43	455.50	64	369.34	48.23	677.60
74	86.31	16.34	164.10	88	422.73	228.25	684.50
90	84.76	7.86	195.23	95	319.03	79.96	612.00
105	72.17	14.92	147.75	125	243.08	85.48	484.90
128	65.40	6.44	97.13				

Nitrate concentration of soil solutions in Site A fluctuated between 2.06 mg/l to 671.75 mg/l. Higher values of nitrate concentration were observed in the upper soil layers. In Site D, nitrate concentration of soil solutions presented higher levels than in site A varying from 37.3 mg/l to 819.5 mg/l with higher mean values in all depths. Nitrate concentration of soil solutions at Site D for all depths are shown in Figure 3.

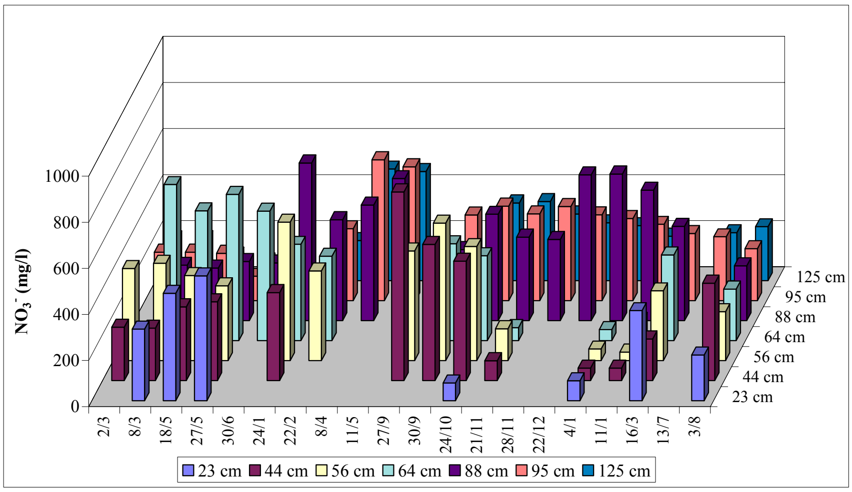

Fig. 3: Nitrate concentration of soil solutions at site D.

The maximum concentrations of nitrate occurred after the application of fertilizers. During the period of rains nitrate concentration was reduced in the upper soil layers, especially in the third year, where there is no cultivation during winter.

In Figure 4(a) nitrate concentration of groundwater at sites A and D and of water samples taken from the Inner ditch and from the outlet of the basin at the end of Karditsa tunnel are shown. Nitrate concentration of groundwater differs also between sites and it is in accordance with the nitrate levels of the corresponding soil solutions. So, in groundwater at site (A) nitrate concentration values varied between 3.31 mg/l to 146.98 mg/l with a mean value of 48.25 mg/l, whereas in site (D) groundwater presented much higher values of nitrate concentration varying between 4.03 mg/l to 407.1 mg/l with a mean value of 161.89 mg/l. This can be attributed to the amount of nitrates that was leaching from the upper layers but also to the dilution effect from the water of the adjacent ditch which had much lower values than groundwater. So, groundwater in Site A which is located next to the Inner ditch, seems to be strongly affected by the horizontal movement of water from the ditch causing a significant dilution. Groundwater in Site D was less influenced since it was situated in a distance of 700 m away from the inner ditch and at the same dinstance from Kifisos.

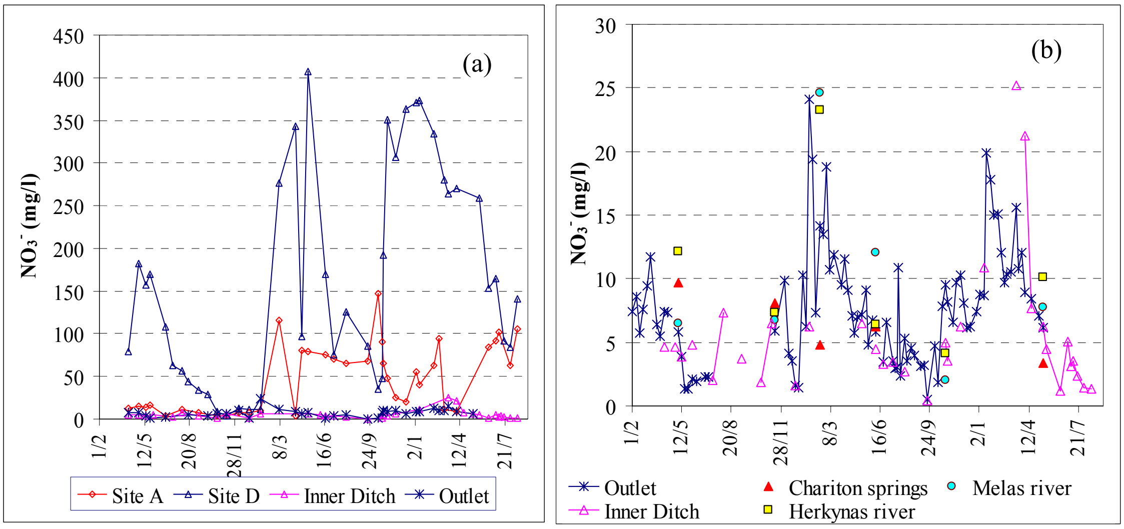

Fig. 4. Nitrate concentration of groundwater at experimental sites and nitrate concentration of water at the Inner Canal (ditch) and at the outlet (at Karditsa tunnel).

From Figure 4a and 4b it is obvious that despite the high values of nitrate concentration in the groundwater, the nitrate concentration of water in the inner ditch did not exceed 25 mg/l. For the monitoring period nitrate concentration of water in the inner ditch had a mean value of 5,23 mg/l. The nitrate concentration of water at the outlet of the collector Canal at Karditsa tunnel presented also low values not exceeding for the monitoring period the value of 25 mg/l. Similar low values of nitrate concentration were found for Melas and Herkynas rivers.

Nitrate movement in the soil and nitrate content of groundwater varied between fields due to soil characteristics, to cultivated crops and applied fertilisation, to water table depth and to the dilution effect of the water from the ditches. Although nitrate leaching to groundwater seems to happen during the rainy period, the rising of water level in the ditches during spring seems to reduce the movement of nitrate from the groundwater to the surface water. The maintenance of a shallow water table allows better utilization of nitrates from the crops and also can enhance denitrification process, especially in soils, such as in Kopais plain, rich in organic matter and with clay layers that can reduce the movement of water and nitrate. Controlled drainage and water table management is a strategy of optimising water use, reducing unecessary drainage and control nitrogen losses from agricultural fields. Water table management can reduce NO_3-N in drainage water by over 60%. (Madramootoo et al.,1993; Bengtson et al., 1984; Gilliam and Skaggs, 1986 and Evans et al., 1991, 1992).

Overall, although the nitrate concentration of soil solutions and groundwater in Kopais plain presented high values, due to the intensification of agricultural production and the high rate of nitrogenous fertilisers applied, the quality of surface waters do not appear to be seriously affected and the concentration of nitrate in surface waters had values lower than 25 mg/l.

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Adsorption process of diesel on unsaturated condition affected by texture and soil moisture

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Abstract
Petroleum pollutants, due to solubility, volatilization and biodegradability, penetrate into the subsurface environment quickly and even little amount of these pollutants can contaminate a wide area of soil and water resources. Adsorption of pollutants by soil particles is the most important mechanism affecting petroleum hydrocarbon transportation in unsaturated and porous soil environments. In this regard, the aim of this study was to determine petroleum hydrocarbons fate in soil, with a focus on their adsorption by 3 soils of different textures namely loam, sand and clay. Therefore, a bath system was designed in a batch experiment, including water and diesel mixtures with four ratios 4:1, 3:2, 2:3, 1:4, only water and only diesel all in three replicates. Soils were extracted by centrifugation and the quantity of adsorbed diesel was determined. Results showed that with diesel alone, clay soil had the most adsorption and sandy soil had the lowest. The adsorption of pollutants with all soils showed significant reduction by adding water to the bath system which suggests a greater affinity for water molecules relative to diesel by soil surface.
Keywords: Adsorption, Diesel, Soil.

Introduction

Contamination of soil and groundwater by organic and toxic pollutants is a wide spread environmental problem. Oil products are one of the most important contaminants in ecosystems. Besides the ability to provide food and other life requirements, soil is also capable of refining (purifying) the water. It owes these characteristics to its physical (providing pores for percolating fluids), chemical (adsorption and emission) and biological (decomposition of organic materials) properties (Mirbagheri et al., 2008).

In Iran as a country which enjoys a great number of Petroleum resources, extensive pollutions have already occurred due to exploitation, refining and transportation and unfortunately not yet considered seriously. Although oil products are beneficial for the welfare and comfort of modern human society, they may cause various diseases including cancer. Several studies have already proved carcinogenic and mutagenic properties of hydrocarbon compounds by entering the cycle of human food. Among the wide variety of petroleum compounds, polycyclic aromatic compounds are of greater importance (Akbari, 2004).

The complexities in understanding chemical and mechanical reactions of soil and contaminants in the presence of air and water phases necessitates studying pollutants in saturated and unsaturated soil environments to identify the dominant processes for the transmission of different pollutants which are then used to design physical models for their transfer in soil (Ebrahimi et al., 2009).

Diesel a mixture of different hydrocarbons including paraffin, naphthalene, aromatics, cycloaromatics and multiple ring aromatics is heavy cutting oil. This substance is a liquid at room temperature and has straw yellow color and sharp smell (Shih et al., 2004).

Diesel is used as a raw material for many oil products in refineries. Diesel fuels a variety of domestic and industrial burners to provide heating and lighting and also is used with internal combustion diesel engines. Diesel Spillages from the underground storage tanks in refineries, warehouses and gas stations and random splurges of oil products in distribution centers are entered into the soil environment due to their specific physical and chemical properties and may lead to environmental pollution. According to epidemiological and clinical studies diesel is a harmful substance to human health with carcinogenesis risks and is classified harmful according to resources recycling and protection act (EPA, 2006).

Adsorption phenomena are used in the purification processes and separation which require solid porous media. Porous solids with extensive surface area per unit volume assume large adsorption capacity. Diesel adsorption rates in soils are of particular importance from environmental point of view. Soil organic and mineral matter, air, water and microorganisms such as bacteria and fungi influence soil physical and chemical properties. Adsorption of diesel in the soil disturbs the ecological balance between the soil and living organism and limits their population. Also the Knowledge of diesel adsorption processes in soils play an important role in making appropriate decisions in taking right approach for cleaning soils contaminated with petroleum products in the event of storage tank leakages or accidents leading to overturning tanker trucks (Geological Survey, 2001).

Due to the importance of this issue, it seems essential to investigate the changes in properties of soils by oil contamination. In recent decades, soil properties contaminated by oil have been studied and several methods have been proposed for refining oil. Likewise effects of pollution on plants in contaminated soils have already been investigated. Some encouraging results have been achieved as a consequence of these efforts. However, the characteristics of the initial adsorption of soil, which is the source of further contamination in soils, are not taken into account (studied) sufficiently. The behavior of unsaturated transport at the early stages of miscible displacement of hydrocarbon pollutants also needs more investigation (Ebrahimi et al., 2009). This study evaluates diesel adsorption in soils of different texture with a batch experiment.

Materials and Methods

In order to examine the effect of different soil textures on the quantity of diesel adsorption, three soil textures namely silty loam, loamy sand and silty clay were obtained from three different locations in Golestan Province. Soil samples air dried and passed through 2 mm sieves.

Adsorption of treatments and extraction

At this step, a system was designed in a batch experiment with 6 treatments in 3 replicates, including water and diesel mixtures with four ratios 4:1, 3:2, 2:3, 1:4, water only and also diesel only. Ten ml solutions of the above treatments was added to 4 gr soil in a centrifuge tube and shacked 24 hours. Thus considerable time was provided for solid-solution interactions and maximum pollutant adsorption onto the soil surface. Soil adsorption at equilibrium was determined in soil extracts after 10 min centrifugation at 3000 rpm.

Results and Discussion

This project investigated the adsorption of water and diesel and their mixtures of various ratios with three soils of different textures. Results showed that with diesel alone, clay soil had the most adsorption and sandy soil had the lowest (Fig.1).

The adsorption of pollutants with all soils showed significant reduction by adding water to the bath system which suggests a greater affinity for water molecules relative to diesel by soil surface. Increasing water to soil ratio reduced soil adsorption slightly however with all soils (Fig.1). These results also suggest diesel is prone to deep percolation and ground water contamination anywhere rainfall exceeds evapotranspiration.

Knowing the behavior of diesel adsorption by soil surface after leakage and seepage and remobilization potential is crucial to predicting risks to environment. The flow of pollution sources and the extent of the contaminated soils are predicted. Knowledge of this behavior is also essential for pollution risk assessment when pollutants are spilt as pulses.

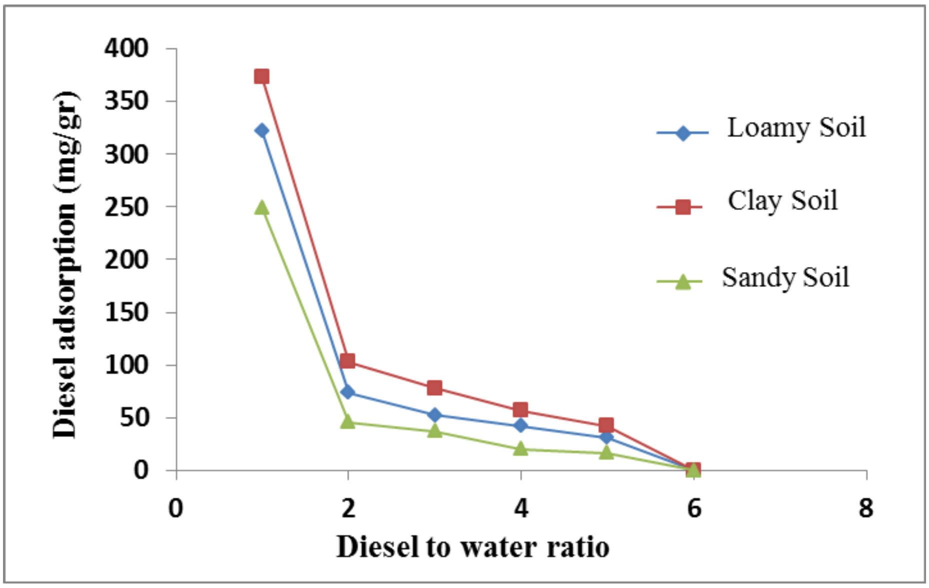

Figure 1. Adsorption of water and diesel and their mixtures of 4:1, 3:2, 2:3, 1:4 ratios with three soils of different textures

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Adsorption of diesel pollutant in different unsaturated soils

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Abstract

One of the most important processes affecting the fate of hydrocarbon contaminants in soil is absorbing by soil particles. It plays a key role in the movement of hydrocarbons in soil, and imposes effects on other processes, especially biological and chemical aspects in soil. The aim of this paper was to investigate the fate of petroleum hydrocarbons in soil by studying how the adsorption of hydrocarbon pollutants would be done by three types of loamy soil, sand and clay respectively. Therefore, a system consisting of nine different pollution compositions of diesel and water pollutants, such as diesel-only, water-only, and seven different combinations of water and pollutants were applied. The importance of this decision was due to the existence of agricultural soils unsaturated system. By adding the pollutant combination to the soil and spending the required balancing time period, the extraction was done by dichloromethane solvent and the amounts of diesel absorbed by the soil was obtained. The results of adsorption isotherms showed that increasing the concentration of pollutants, was caused exponential increasing in diesel adsorbed in all pollutant-water treatments. However, the slope of the curve slightly declined at higher concentrations so that due to lack of adequate absorption level for the adsorption process at higher concentrations, increasing in the pollution did not lead to increasing the rate of adsorption. Adsorption curves fitted by Langmuir and Freundlich models showed that both of these models had a good fitting with our data. But Langmuir model in explain the hydrocarbon contaminants adsorption in soil was more capable.
Keywords: Adsorption, Diesel, Freundlich, Isotherm, Longmuir, unsaturated soil.

Introduction

One of the major environmental problems today is hydrocarbon contamination resulting from the activities related to the petrochemical industry. Release of hydrocarbons into the environment whether accidentally or due to human activities is a main cause of water and soil pollution. Oil hydrocarbons in soil can be found as vapor phases, masses of liquid hydrocarbon, phases which are adsorbed from the soil surface, or solution phases in the water.

Among the various mechanisms, such as broadcasting (propagation), absorption, evaporation, as well as biological and non-biological decompositions, which are effective in reduction in mass, toxicity, volume, concentration, and mobility of oil pollutants in vados zone, the adsorption of oil by soil particles is one of the most important processes determining the destination of the oil in the soil (Mirbagheri *et al*., 2008)

This process plays a vital role in the movement of carbohydrates in the soil, and also has a noticeable influence on other biological and chemical processes. Due to the importance of this issue, it seems essential to investigate the changes in properties of soil causing by the leakage of oil contamination. In recent decades, the characteristics of soil contaminated by oil have been studied and several methods for oil refining have been proposed. Likewise, much research has been done on the effect of pollution on plants in the contaminated area as well as other aspects after the occurrence of the pollution. And some favorable results have been achieved as a consequence of these great efforts. However, the characteristics of the initial absorption of soil, which would be considered as the source of further contamination in the soil, are not taken into account (studied)

sufficiently. Moreover, regarding and insisting on the soil properties, the behavior of the unsaturated soil in the early stages of the propagation of hydrocarbon pollutions (Ebrahimi *et al.*, 2010).
In this regard, this study is performed in order to determine and evaluate the oil adsorption in unsaturated conditions being affected by the soil texture and moisture.

Materials and Methods
In order to examine the circumstances of the adsorption of oil and the effect of different soil textures on the quantity of such adsorption, three types of soil textures, namely, silty loam, loamy sand and silty clay was obtained from three different area of Golestan Province. These soil samples were passed through 2mm sieves after getting dried by the air contact.
At this step, a system consists of 9 different diesel pollutants and water was considered. The considered treatments include three replications of only diesel treatment, only water treatment, and seven different treatments combined of diesel pollution and water. The importance of this choice is due to the unsaturated agricultural soils system. Therefore, 4gr soil was poured in a centrifuge tube and it was saturated with any of these treatments of water and diesel, and then it was shaken for 24 hours. Thus, a considerable possibility was provided for contacting and obtaining maximum adsorption of pollutants into the soil surface. After establishing the final equilibrium state, centrifuged was done and the amount of pollutants in soil extracts was estimated.
Gasoline extracted from soil and the supernatant extract was according to approach Marcos-Rvsha with little change. In this stage, on 4 gr of soil contaminated was added about 20 ml of dichloromethane. The solvent dichloromethane, Transfer capability of pollutants by adsorption on the soil surface into their, Therefore, diesel of contaminants are separated from soil surface and have been transitioned into the solvent. For this purpose, Mixed the solvent, pollutants and soil was shaking strongly for 30 minutes with mechanical Shaker. The desired complex with 3000 rpm was centrifuged for 10 minutes, finally the soil to be deposited. In the next step, Supernatant containing the contaminants was separated from the soil and in the glass container was weighed before it was poured. The above step was repeated 2 times. Then the solvent was allowed for 24 hours in the room temperature evaporates for dichloromethane and remainder was weighed again. The weight of diesel contaminants in soil revealed.

Results
In this project, adsorption of diesel took place in three type soils by different texture and concentrations of diesel. The adsorption experimental results showed that in only contaminants treatment, clay soils have the highest and sandy soil have the lowest adsorption. By adding the water to the pollutants system, the adsorption of pollutants showed significant reductions that this rate with increases water became higher and the slope the curve in sandy more than loamy and in the clay soil was lowest. Adsorption equilibrium isotherm indicated that in all treatments pollution-water, adsorption of diesel contaminant with increasing concentration, to exponentially increase. However at higher concentrations, the slope of the curve was slightly down. So, at higher concentrations of contaminant due to lack of sufficient absorbent surface to continue the process of adsorption, with increasing contaminant, increasing the rate adsorption did not created. Adsorption curves fitted by Langmuir and Freundlich models showed that both of these models had a good fitting with our data. But Langmuir model in explain the hydrocarbon contaminants adsorption in soil was more capable. Freundlich and Langmuir curve fitted a good with R^2 rate about 98%.

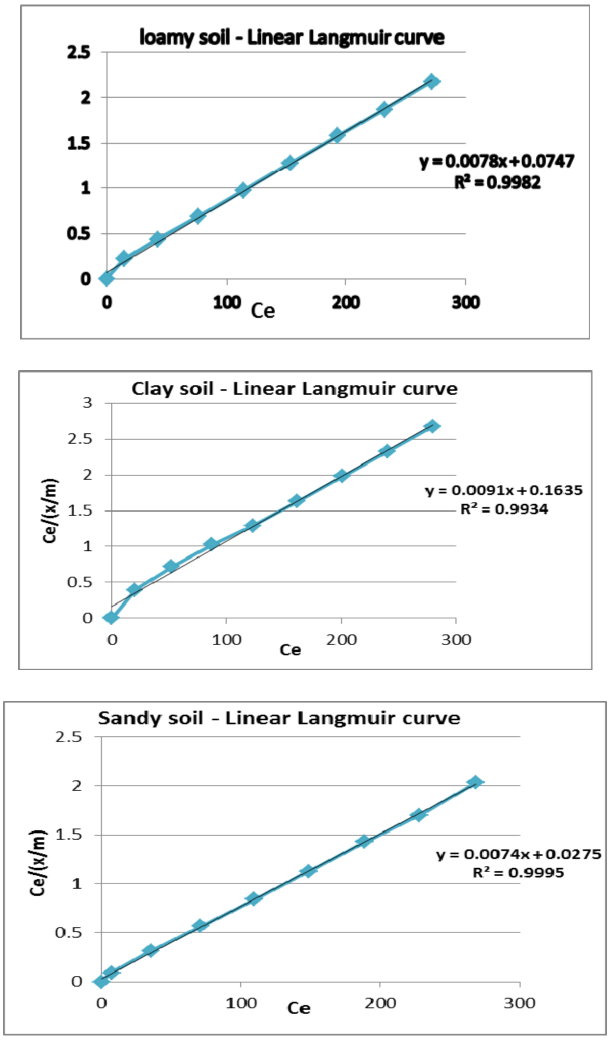

Figure 1- Langmuir adsorption curves of diesel pollutants in the three soil textures

Discussion

Due to specific characteristics diesel, with its leakage and seepage into the environment, Scrutiny its behavior, especially study adsorption diesel, that it is as a new source of the pollution, in different soils especially in shock conditions, it seems to be essential. Thus the flow of new pollution sources designated, in addition, rate contaminated soil is also determined.

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Contribution of nitrate loads in irrigation return flows to the nitrate budget in an irrigated area

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Extended Abstract

Nitrate (NO_3) concentration in irrigation return flows (IRFs) is the indication of N loss from soil, thus fertilizers, and also level of nitrate pollution in water sources. However, measurement of NO_3 concentration itself is not the only parameter to use as pollution and loss criteria, these values should be converted to the NO_3 load for better assessment of the pollution level and its environmental and economical impacts.

Irrigation return flows have been recognized as the major diffuse or non-point pollution contributor of surface and groundwater bodies (Aragües and Tanji, 2003). Water quality of IRFs is predominantly affected by salt and nitrate concentrations. Nitrate concentrations up to 250 mg L^{-1} (European Union, 1998) were recorded for shallow aquifers, while the concentration rarely reach 50 mg L^{-1} for surface waters of the irrigated areas. High NO_3 concentrations have been measured in IRFs, especially during N fertilization to small grains. Seasonal patterns of N fluxes influenced by fertilization, irrigation scheduling and rainfall distribution have been studied. Isidoro et al. (2006) found that 75% of the total NO_3-N load was exported during the irrigation season in an irrigation district in Spain. Optimizing N fertilization, irrigation scheduling and irrigation efficiency could reduce N export to drainage waters (Cavero et al., 2003; Isidoro et al., 2006) and groundwaters (Thorburn et al., 2003).

Therefore, the research was conducted in Akarsu Irrigation District (9,495 ha) of Lower Seyhan Plain in southern Turkey 2007 to 2009 for three years. The soils of the *Akarsu Irrigation District* are mainly alluvial, deep and high in clay and calcium carbonate, and deep cracks are common during the dry summer season. According to Soil Taxonomy classification (Dinc et al. 1995), the soil moisture regime is *xeric*, and the soil temperature regime is *thermic*. Maize as a first and second crop was the major crop, followed by citrus orchards and wheat in the research area during these three years.

Samples of IRFs were taken from three drainage gauging stations, as two inflows and one outflow, in each month during the each hydrological year (HY) (from Oct.1st to Sept. 30th). Amounts of drainage water (mm), NO_3 concentrations and NO_3 loads of the samples in these stations were either measured or analyzed from October to September. Drainage waters were analyzed for NO_3 concentrations based on the Standard Methods (1998). NO_3 and NO_3-N loads in drainage waters were obtained by multiplying the daily NO_3 concentrations and daily discharge rates. Nitrate loads (L_{NO3} *(kg* ha^{-1} day^{-1} based on irrigated area*)* in IRFs in a given day, *j,* were derived by employing the following equation (Eq. 1).

$$L_{NO3},_{j} = 0.001 \left[\frac{Q_{oj}\, C_{oj} - Q_{inj}\, C_{inj}}{A} \right], \quad j = 1, 365 \tag{1}$$

where Q_{oj}, Q_{inj}, A stand for drainage out- and inflows (m^3 day^{-1} in any day of the HY), irrigated area (ha), respectively. C is the respective mean NO_3 concentration (mg L^{-1}) of the daily mean drainage discharge rate measured in a given day *j*. Nitrate loads in irrigation water can be calculated as the same way by changing the places of the terms in the parenthesis in Eq. 1. Total NO_3 load for HY can be obtained by adding the daily loads as in Eq. 2:

$$L_{NO3,s} = \sum_{j=1}^{t} L_{NO3,j} \quad (2)$$

where L_{NO3} is the total NO_3 load in either irrigation or IRFs for a period of *t-day* long (*t*=365 days for the whole hydrological year).

Amounts of drainage waters were higher in rainy and especially in irrigation season (April to August) because of the intensive irrigation to many summer crops of the study area, and measured as total of 788, 779 and 953 mm in these three years. Nitrate concentration range was between 7.9 to 47.3 mg L^{-1} across the months and the years. Even though some of these concentration values are far beyond the critical pollution level, the total yearly NO_3 loads/mass in drainage waters were average of 176, 130 and 245 kg NO_3 ha^{-1} in 2007, 08 and 09, respectively. These values were converted to NO_3-N (kg NO_3-N ha^{-1}), and partitioned for the hydrological year (HY), irrigation season (IS) and non irrigation season (NIS) (Table 1). About 43 to 52 % of the N load in IRFs was outflowed during the NIS. Therefore, almost %50 of the loss happens during the IS.

Table 1. Nitrate-N load partitioning in hydrological year (HY), irrigation season (IS) and non irrigation season (NIS), and % load of NIS in total.

Year	HY	IS	NIS	% of NIS
	kg NO_3-N ha^{-1}			
2007	39.7	20.9	18.8	47.4
2008	29.3	16.9	12.5	42.6
2009	55.3	26.7	28.6	51.5

Nitrate loads in IRFs were peaked in winter based on boosted fertilization of winter crops such as wheat, and in summer with heavy fertilization and irrigation, i.e. corn receives 340 and 330 kg N ha^{-1} as in excessive amounts. Type of irrigation is also important factor on magnitude of the load. Since it is difficult to control precipitation, management of irrigation and also fertilizer practices are very critical to lessen the NO_3 export to IRFs.

In the related European projects (Qualiwater and Medsalin), nitrogen budget, including the IRFs as one of the budget parameters, was calculated in the District. Nitrogen loss to the IRF was the second important budget output after the plant N uptake (Karnez, 2010). Therefore, considering load values could be better indication in assessment of NO_3 loss, water pollution and total nitrogen budget of the irrigation district.

Keywords: irrigation return flow (IRF), nitrate pollution, concentration, load, irrigation district

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Effects of autumn catch crops and tillage regime on soil nitrate leaching from grazed annual green manures

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Abstract

Benefits of green manures (GM) can be further supplemented by livestock grazing. However, readily available NO_3^- from grazed GM can potentially be lost from the system by leaching from the root zone. The objective of this study was to determine how two management factors; type of catch crop and tillage; affected NO_3^- leaching and N uptake in a subsequent crop. A three year experiment was established in Manitoba, Canada in 2010. In the first year, upon grazing (termination) of mixture of pea/oat/soybean green GM, catch crops (barley and oil seed radish) were seeded directly (reduced-till) or after cultivation (tilled). Control plots had no catch crops. Second and third year tests crops will be wheat (harvested in 2011) and fall rye (for summer 2012). Measurements included aboveground biomass and N content of catch crops and wheat, as well as wheat grain. Soil samples were taken to 120 cm, before grazing, after grazing, upon termination of catch crops, before seeding wheat and after wheat harvest. In 2010 the oil seed radish catch crop biomass contained significantly more N than barley; 85 kg ha^{-1} and 73 kg ha^{-1} respectively. Higher soil profile N in control (117 kg ha^{-1}) vs catch crop treatments (average 59 kg ha^{-1}) suggests reduced N leaching with catch crops. Significantly lower wheat yield in the catch crop vs control plot suggests that catch crops restricted N availability to the following crop. No significant tillage effects were observed. Catch crops reduced leaching of NO_3^- regardless of tillage system, but also reduced wheat yield.

Introduction

Organic farmers in Northern Great Plains of North America include green manures into rotation to enhance soil fertility. Although sustainable in the long run, this practice lacks immediate economic benefit. Livestock integration has been suggested as a means of adding value; by utilizing a portion of N and C stocks of green manure biomass for live-weight gain. In grazed systems some of the nutrients (15-20%) consumed by the livestock are used for maintenance and productivity, but most of the nutrients are returned to soil in urine and faeces (Whitehead, 2000). The plant availability of N in faeces and urine is very high and if not taken up by plants may be susceptible to losses through leaching, volatilization and denitrification (Williams and Haynes, 1994). This phenomenon is well documented in pastures with living biomass and roots, however, there is limited data on annual grazed green manure systems. Deposited faeces and urine unto a soil devoid of living biomass may be more liable to losses than a ground with living biomass.

Unlike green manures, which are seeded with the purpose of increasing soil fertility, catch crops are mainly grown to capture excess nutrients, particularly N. In the temperate regions of the world, fast growing, cold tolerant, and high N demanding plants such as barley (*Hordeum vulgare*) and oil seed radish (*Raphanus sativus*) might be suitable. Beside effective capture of soil N, catch crops, ideally, release this N to the succeeding cash crops when the cash crop N demand is high. However, mineralization rates are often dictated by both environmental conditions and residue quality (Griffin, 2008; Andersen and Jensen, 2001). For instance, in the Canadian Prairies, the mineralization from plant residue and soil organic matter is low; less than 30–40% in the first year (Janzen and Schaalje, 1992). Therefore, low lignin, low C:N ratio crops are preferable for fast decomposition and mineralization.

Another point to consider in management of grazed green manures is the tillage. Tillage immediately after grazing can increase the N mineralization rates, may distribute the urine and dung patches more evenly and provide a better seed bed. On the other hand, residue left on soil after grazing can offer protection against erosion and may provide weed control.

This paper reports from first two years of grazing green manures experiment with catch crops. Our objective was to investigate the N capture and release patterns of catch crops that are seeded directly or conventionally following grazing green manures under. We hypothesized that catch crops will significantly reduce N leaching from grazed green manure plots compared to no catch crop plots.

Materials and Methods

The trial was conducted at University of Manitoba Ian Morrison Research Station located at Carman, Manitoba, Canada (49^o 49' N, 98^o 00' W) on a Hochfeld fine sandy loam soil (coarse, loamy, mixed Udic Haplocryoll). The green manure mixture of forage pea (*Pisum savitum* cv. 40-10), soybean (*Gylicine max* cv. Prudence) and oat (*Avena sativa* cv. legget) was seeded in May 2010 and grazed in first week of August 2010. Every plot was grazed by 3 ewes and 2 lambs for 24 hours. The plot size was 2m in width and 9m long and was surrounded by metal fence for precision and protection.

Upon termination by grazing, barley (*Hordeum vulgare* cv. cowboy) and oil seed radish (*Raphanus sativus* L.) were seeded either directly or after cultivation of the grazed plots. The experimental design was randomized complete block design in split plot arrangement with four replicates. The main plots were seeding type (i.e. direct-seeded or cultivated) and the subplots were the species (i.e. barley, oil seed radish and control plot with no catch crop). Differences were considered significant at $P < 0.05$. Means were separated using a Fisher protected LSD test.

Soil samples were taken using Dutch auger at four 30cm increments up to 120cm depth and analyzed for NO_3^- using KCl extraction method. Biomass of catch crops as well as wheat was collected from 2x 0.4 m^2 areas within each plot. Samples were sorted and dried for 2 days at 60 ^oC and weighted for dry matter content. Dry biomass samples were ground with Wiley Mill No.1 (Arthur H.Thomas Co., Philadelphia, PA). The ground samples were subsampled and analyzed for N concentration by combustion analysis using a LECO FP-528 (LECO, St. Joseph, MI).

Results and discussion

Both barley and oil seed radish (OSR) established well and produced 4615 kg ha^{-1} and 3797 kg ha^{-1} of dry matter respectively. In 2010 the OSR catch crop biomass contained significantly more N than barley; 85 kg ha^{-1} and 73 kg ha^{-1} respectively. Wheat maturity biomass N content was lowest in barley plots (36 kg ha^{-1}) but similar in OSR and control plots, indicating the faster mineralization of OSR compared to barley (Table 1). However, wheat yields were significantly lower in both catch crop plots compared to control plots. The yield potential of wheat is set in earlier stages of development, therefore, low availability of N from OSR plots in earlier stages may have resulted in low yields in spite of higher availability of N in the later stages (Borràs et al., 2004).

At all soil profiles, control plots had significantly more NO_3^- than barley and OSR catch crop plots, when they were terminated in 2010 (Fig 1). Also in 2010, pre-seeding soil NO_3^- levels were 45.9 kg ha^{-1} (data not shown) and, by the time catch crops were terminated soil NO_3^- was 117.4 , 64.3 and 55.3 kg $ha^{-1}$for control, OSR and barley respectively (0-120cm). Clearly, N mineralization from urine and faeces, as well as, from soil organic matter more than doubled the amount of soil NO_3^- , from 45.9 kg ha^{-1} to 117.4 kg $ha^{-1}$for control. However, barley and OSR were able to capture half of that available NO_3^-. After the harvest of wheat in 2011, control plots had significantly less NO_3^- than barley and OSR plots at 0-30cm, but the NO_3^- levels at other depths were similar (fig 1). Total NO_3^- (0-120cm) were similar in all plots, suggesting that available NO_3^- in control plots were either taken up by wheat or lost.

There were no significant differences between the direct seeded and conventionally seeded plots in terms of biomass production (Table 1). Even though the conditions for seed germination were optimal in 2010, the performance of direct seeded plots is noteworthy, as it implies reduced soil disturbance and energy savings. There was no significant catch crop by tillage interaction as both barley and OSR performed similarly well under both tillage treatments.

Table 1: Effect of management (DS: direct seeding and CS: conventional seeding) and catch crop (barley and oil seed radish (OSR) on the catch crop biomass production, catch crop biomass N uptake, wheat anthesis N uptake and wheat yield. Differences were considered significant at *P* < 0.05.

Crop	Management	2010 catch crop biomass	2010 catch crop biomass N	2011 Wheat anthesis N	2011 wheat yield
		---------------------------------------kg ha^{-1}----------------------------------- -------			
Control	DS	n/a	n/a	41	1733
	CS	n/a	n/a	55	1874
Barley	DS	4184	70	37	1281
	CS	5047	77	36	1221
OSR	DS	3748	91	42	1354
	CS	3846	80	40	1559
	ANOVA				
	Source of Variation				
	Crop	<.0001	0.045	0.020	0.002
	management	0.393	0.845	0.366	0.408
	Crop x mng	0.365	0.109	0.105	0.839

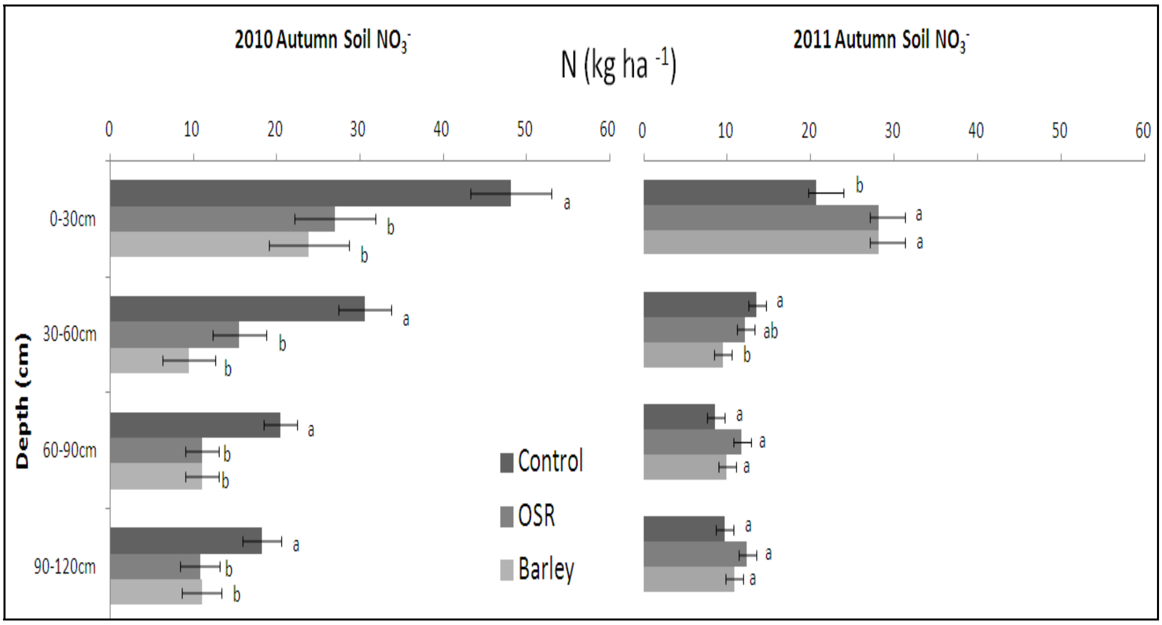

Figure 1: Soil NO_3^- in 2010 autumn (catch crop year) and 2011autumn (wheat year). Means within the same column followed by the same letter are not significantly different according to Fisher's LSD test (*P* < 0.05).

Conclusions

Overall, catch crops captured most of the available NO_3^- , thus prevented the leaching of NO_3^- beyond root zone (i.e. beyond 120cm). However, the N release to wheat in the second year was inhibited compared to no-catch crop plots. There were no significant tillage effects in terms of biomass production, soil NO_3^- concentrations or wheat N uptake, thus direct seeding may be recommended as a means or reducing soil disturbance and saving energy. Catch crops reduced leaching of NO_3^- regardless of tillage system, but also reduced wheat yield. This study will continue into third year with fall rye test crop to assess N mineralization from catch crops.

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Survey of Some Soil Borne Plant Pathogenic Fungi Contaminating Different Plants Soil

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Abstract

Isolation of pathogenic fungi contaminating different soil .Isolation of pathogenic fungi contaminating the soil planted with onion, garlic, potato, sugar beet and soybean .pathogen city tests indicated that *Sclerotium cepivorium* isolated from onion soil caused damping off onion. *Fusarium subglutinans* isolated from garlic soil caused damping of garlic *Fusarium proliferatum*, *Fusarium solani and Rhizoctonia solani*.isolated from potato soil caused damping off potato *Fusarium lateritium*, *Fusarium xylarioides* and *Fusarium camptocearas* isolated from soil caused damping off sugarbeet . *Fusarium proliferatum and Fusarium oxysporum* isolated from wheat soil causing damping off wheat. *Fusarium solani* and *Fusarium moniliforme* isolated from soybean caused damping off soybean. *In vitro* and under greenhouse conditions Addition of1%of leaves powder of sweet orange reduce %growth mycelia and damping off caused by all tested soil borne pathogens compared with untreated.

Keywords: Soil Pathogenic Fungi leaves orange

Introduction

The soil contain mined with soil borne plant pathogens causes many important diseases for several important plants for human bean nutrition such as Onion, garlic, potato, sugar beet, soybean and wheat. The fungi

Sclerotium cepivorum is widely distributed plant pathogenic fungus in the Egyptian agricultural soil. is the causal agent of the white rot of onion (Embaby *et al* .,2006) *Fusarium subglutinase* reduced emergenceof garlica (**Crowe,**2006). Potato plants are attacked by *Rhizoctonia solani, Fusarium proliferatum* and *Fusarium solani* causing crown rot, root and seeding damping off (Brewer2005; Uddin2010; Almasia 2008; El-Kot2008; Uddin2011). Sugarbeet an important crop for sugar can be attacked by *Fusarium lateritium, Fusarium xylarioides,* and *Fusarium campasitras,* causing damping off sugar beet (Hanson and. Hill 2004; El-Kazzaz *et al*., 2008). *Fusarium solani* and *Fusarium moniliforme* cause. Damping off, root rot of soybean plants (El-Kazzaz 2008) wheat infected with *Fusarium proliferatum* and *Fusarium oxysporum* Causing damping off (El-Kazzaz2008)

The aim of this study evaluated the qualitative pathogenic fungi and quantity contemning the soil and causes sever disease incidence in different field plants, Pathogen city tests of the fungi contained the soil Reduce damping off caused by all tested soil borne pathogens by using leaves powder of sweet orange

Materials and Methods

Isolation and identification of the causal pathogens:

Soil was collected from nine plots in each of two fields cultivated with onion garlic sugar beet wheat and soybean in Drank Manfulat Assiut El-Kussiah ,Elbader and Assiut Governate. Plots were randomly selected from a test area. Soil samples were collected from the center two rows of each plot to a depth of 15cm using a 2.5-cm-diameter soil probe. Counting fungi by dilution plate method as described by (Nitta 1992) .Identification of the isolated fungi was carried out by using morphological characteristics of mycelia, spores according to Domsch *et al.,* (1980) Singh (1994) (;Moubasher 1993; leslie and Summerell 2006). Then confirmed by Assiut University Mycological center (AUMC).

Pathogenicity tests:

Pathogen city tests of isolated fungi isolated were determined on the same host isolated from under greenhouse condition. Sterilized pots (30cm in diameter) containing sterilized (1 clay: 1sand) (v/v) were seeded. Inoculums of *S.cepivorum* were prepared by growing on autoclaved sand-corn meal medium (20k coarse sand ,1k ground maize and liter water) in 500 ml milk bottle and incubated at 20 °C for 30 days Petri plates sclerotia were mixed with autoclaved clay soil at the rate of 5sclerotia /g soil. Inoculate of the another tested fungi were prepared by growing each isolate in 500ml. Bottles containing barley grains medium (100g barley grains + 50ml water) and incubated at 28°C for 15 days .Soil infestation was carried out 7 days before sowing and by adding the fungal inoculums separately to each pot at the rate of 5% of soil weight, Pots containing non –infested soil mixed with 5% of sterilized barely medium were used as control. Each pot was sown with ten seeds individually for Gize6 onion , sads1wheat oskarpoly sugar beet and Clark soybean individual 10 cloves of Balady garlic and ,10 sprout for Diamond potato. Four pots were used for each isolate as replicates .Pots were examined daily and the percentages of pre- and post-emergence seedling damping- off was recorded after 3 and 6 weeks from sowing respectively..

Effect of the leaves powder of sweet orange (*Citrus sinensis*)on the tested pathogenic soil born fungi *in vitro*

The leaves powder of sweet orange (*Citrus sinensis*) were air dried on the bench for 10 days on the laboratory and then grounded into uniform powder using blander .one gram of The leaves powder of sweet orange was dissolved in 10 ml of PDA broth medium ,sterilized by filtration through Seitz filter, then 10 ml was added to Flask (250 ml)contained 90 ml 0f autoclaved PDA medium were melted, cooled to about 52C mixed to give final concentration 1%.Flask,mixed and poured in sterilized Petri dishes(20 ml/ plate) Plates were inoculated with equal disks(4 mm in diameter) of al tested soil born pathogen .Three replicates were used for each treatment .Control treatment was carried out by cluttering the tested fungi on PDA medium without addition of The leaves powder of sweet orange The inoculated plates were incubated at 28C until the fungal growth covered the plate surface of the control treatment diameter of fungal growth in all treatment was determined and the percentage of mycelia growth reduction was calculated using the formula

$$\% \text{ of growth reduction} = \frac{\text{growth in control} - \text{growth in treatment} \times 100}{\text{Growth in control}}$$

Effect of the leaves powder of sweet orange on the tested pathogenic soil born fungi under greenhouse conditions during growing seasons2010 and 2011

The same method for seeded, infestation and inoculums preparation at the rate of 5%of soil weight as described above except use soil (1clay:1sand) treatment with **the tested pathogenic soil born fungi individually and** leaves **powder** of sweet orange1% weight of soil in pot 30Cm and control soil (1clay:1sand) without treatment with the leaves powder of sweet orange but with the tested **pathogenic soil born fungi individually** percentages of pre- and post- emergence seedling damping- off was recorded after 3 and 6 weeks from sowing respectively.

Statistical analysis:

Data were subjected to statistical analysis and means were compared using L.S.D. test (Gomez and Gomez 1984).

Results and Discussion

Isolation, Identification and population of the causal Fungi from different localities

The isolated fungi are presented in Table (1) reveal that tested isolates were identified *Scelerotium cepivorum* the total count17 x10^4 were isolated from soil planted with onion *,Fusarium subglutinase* the total count10x10^4 were isolated from soil planted with *garlic ,Fusarium subglutinase*. *Rhizoctionia solani* the total count12 x10^4 *Fusarium proliferatum* the total count10 x10^4 *Fusarium solani* the total count12 x10^4 were isolated from soil planted with potato. *Fusarium lateritium, Fusarium xylarioides, and Fusarium campasitras* the total count10 x10^4 were isolated from soil planted with sugar beet. *Fusarium solani* total count14 x10^5 *Fusarium moniliforme* total count16 x10^4 were isolated from soil planted with Soybean. *Fusarium proliferatum* total count14 x10^4 *Fusarium solani* total count16 x10^4 were isolated from soil planted with wheat as reported by Hossein and Wadud(1976) they found that fungal population was differed in the soil cultivated with different crops such as potato ,tomato bean cabbage and papaya.

Table 1. Source of pathogenic fungi and population

Fungi	Plant soil	Location	Total Count(C.F.Ux10^4/gsoil)
Scelerotium cepivorum	Onion	Darnk	17
Fusarium subglutinase	Garlic	Assiut	10
Rhizoctionia solani	Potato	Manfulat	12
Fusarium proliferatum	Potato	Assiut	10
Fusarium solani	Potato	Assiut	12
Fusarium lateritium,,	sugar beet	Assiut	10
Fusarium xylarioides	sugar beet	Assiut	10
Fusarium campasitras	sugar beet	Assiut	10
Fusarium solani	Soybean	Assiut	14
Fusarium moniliforme	Soybean	ElKussiah	16
Fusarium proliferatum	Wheat	Elbader	14
Fusarium oxysporum	Wheat	Manfult	16

Pathogencity tests:

Isolated fungi were tested under greenhouse condition for their pathogen city.

Data shown in Table (2) indicated that *S. cepivorum* was able to infected Giza 6 onion. causing increased significantly damping off compared with control untreated as previously shown(. Embaby *et al.,*2006) *F.subglutinase* were infected Balady garlic causing increased significantly damping off garlic compared with control. as previously(**Crowe,**2006). Data in Table (3) indicated that *R, solani*, *F.proliferatum* and *F.solani* infected potato plants causing significantly increased damping off compared with control untreated. as previously shown. (Brewer2005; Almasia 2008**.** El-Kot2008**;** Uddin2011). Data also indicated that *F. solani* caused the highest total infection, pre, post emergence followed by *R.. solani* and *F. proliferatum*.Data presented in Table (4) indicated that significantly increase damping off oskarpoly sugar beet compared with control untreated. Data also indicate that *F. xylariodies* caused the highest total infection, pre, post emergence damping off followed by *F. compatoris* and *F.teritum*.

Data presented in Table (5) indicated that *F. proliferaitum* and *F*.oxysporium significantly increase pre, total infection of wheat compared with control as previews (El-**Kazzaz**2008). Data also indicated that *F. oxysprum* caused more pre, post damping off, total infection compared with *F. proliferatum*

Data presented in Table (6) indicated that *F. solani* and *F. moniliforme* caused significantly increased in pre damping off and total infection of soybean compared with control untreated. Data also indicate that *F. moniliform*e caused the heights, pre, post, total infection as previews(**El-Kazzaz**2008 compared with *F. solani*.

Table 2. Pathogen city test *Selrotium cepivorum* and *Fusarium subglutinase* the causal pathogen of damping of onion and garlic

Fungi	Host	Pre %	Post %	Total infection
Sclerotium cepivorum	Onion	15	2.5	17.5
Control		0	0	0
Fusarium subglutinase	Garlic	40	7.5	47.5
Control		0	0	0
L.S.D. 5%		5.496	5.332	5.961

Table (3): Pathogen city tests *Rhizoctonia solani*, *Fusarium solani* and *Fusarium proliferatum.* The causal pathogen of damping off potato

Fungi	Host	Pre %	Post %	Total infection
Rhizoconia solani	Potato.	30	7.5	37.5
Control		0	0	0
Fusarium proliferatum		20	5	25
Control		0	0	0
Fusarium solani		32.5	10	42.5
Control		0	0	0
L.S.D. 5%		3.975	5.496	5.961

Table 4. Pathogen city tests *Fusarium lateritium, Fusarium xylarioides and Fusarium campasitras* the causal pathogen of damping off Sugar beet

Fungi	Host	Pre %	Post %	Total infection
Fusarium lateritium	Sugar beet	42.5	5	47.5
Control		0	0	0
Fusarium xylarioides		55	5	60
Control		0	0	0
Fusarium campasitras		47.5	5	52.5
Control		0	0	0
L.S.D. 5%		7.054	7.054	11.923

Table 5. Pathogen city tests of *Fusarium proliferatum* and *Fusarium oxysporum* the causal pathogen of damping of Wheat.

Fungi	Host	Pre %	Post %	Total infection
Fusarium profiraitum	Wheat	20	5	25
Control			0	0
Fusarium oxysporum		25	5	30
Control		0	0	0
L.S.D. 5%		5.767	N.S	11.535

Table 6. Pathogen city test of *Fusarium solani* and *Fusarium moniliforme* the causal pathogen of damping of Soybean.

Fungi	Host	Pre %	Post %	Total infection
Fusarium solani	Soybean	35	7.5	42.5
Control		0	0	0
Fusarium moniliforme		45	5	50
Control		0	0	0
L.S.D.5%		9.9800	N.S	14.984

Effect of the leaves powder of sweet orange on the tested pathogenic soil born fungi *Invitro*

Data presented in Table (7) indicated that the leaves powder of sweet orange inhibited the growth of the all tested pathogenic soil born fungi *in vitro* this work is in agreement with the work of Amadioha andObi(1988) they mentioned that extract of citrus plant contain antifungal compound that can be used as alternative to synthetic fungicides Okwu *et al.,(2007)*they found The leaves of sweet orange contain alkaloids, phenols ,flavonoids ,tannins and saponins inhibited the growth of *Fusarium oxusporum in vitro* which causes damping off disease of Okra plant.

Table 7. Effect of the leaves powder of sweet orange on the tested pathogenic soil born fungi *in vitro* under greenhouse conditions during growing seasons2010 and 2011

Fungi	**Plant soil**	% of growth reduction
Scelerotium cepivorum	Onion	15.5
Fusarium subglutinase	Garlic	37
Rhizoctionia solani	Potato	22
Fusarium proliferatum	Potato	16
Fusarium solani	Potato	30.6
Fusarium lateritium,,	sugar beet	24
Fusarium xylarioides	sugar beet	16
Fusarium campasitras	sugar beet	27
Fusarium solani	Soybean	15.3
Fusarium moniliforme	Soybean	18.3
Fusarium proliferatum	Wheat	27.6
Fusarium oxysporum	Wheat	31.3

L.S.D 5% 2.019

Data presented in Table (8, 9,10,11 and12) indicated that the leaves **powder** of sweet orange reduce damping off for all tested pathogenic soil born fungi compared with control untreated with leaves **powder** of sweet orange this result agree with Van Heerden *et a*l.,1995) they found that control of soil borne pathogens by using citrus waste compost

Table 8.Effect of the leaves powder of sweet orange on the damping off onion and garlic caused by *Selrotium cepivorum* and *Fusarium subglutinase*. Under greenhouse conditions during growing seasons2010and2011

Fungi	Emergence damping –off%				Total infection	
	Pre %	Pre %	Post %	Post %		
	2010	2011	2010	2011	2010	2011
Sclerotium cepivorum	10.5	5	0	0	10.5	5
Control	15	15	2.5	2.5	17.5	17.5
Fusarium subglutinase	22.5	20	2.5	2.5	25	22.5
Control	37.5	35	7.5	5	45	40
L.S.D. 5%	6.66	7.998	5.496	6.332	8.535	7.998

Table 9 Effect of the leaves powder of sweet orange on the damping off potato caused by*Rhizoctonia solani*, *Fusarium solani* and *Fusarium proliferatum.* Under greenhouse conditions during growing seasons2010and2011

Fungi	Emergence damping –off%				Total infection	
	Pre %	Pre %	Post %	Post %		
	2010	2011	2010	2011	2010	2011
Rhizoconia solani	22.5	20	5	2.5	27.5	22.5
Control	27.5	22.5	5	5	32.5	27.5
Fusarium proliferatum	15	12.5	2.5	2,5	17.5	15
Control	22.5	15	5	7.5	27.5	22.5
Fusarium solani	25	17.5	7.5	5	32.5	22.5
Control	30	25	7.5	5	37.5	30
L.S.D. 5%	6.879	6.502	6.879	5.673	10.775	9.264

Table 10. Effect of the leaves powder of sweet orange on the damping off sugar beet caused by *Fusarium lateritium, Fusarium xylarioides and Fusarium campasitras* Under greenhouse conditions during growing seasons2010and2011.

Fungi	Emergence damping –off				Total Infection	
	Pre %	Pre %	Post %	Post %		
	2010	2011	2010	2011	2010	2011
Fusarium lateritium	25	15	2.5	2.5	27.5	17.5
Control	27.5	22.5	5	5	32.5	27.5
Fusarium xylarioides	15.5	12.5	2.5	2.5	17.5	15
Control	20	17.5	7.5	5	27.5	22.5
Fusarium campasitras	27.5	20	2.5	2.5	30	22.5
Control	32,5	22.5	5	5	37.5	27.5
L.S.D. 5%	12.784	6.74	4.766	6.502	10.978	8.9519

Table 11. Effect of the leaves powder of sweet orange on the damping off wheat caused by *Fusarium proliferatum* and *Fusarium oxysporum*. Under greenhouse conditions during growing seasons2010and2011

Fungi	Emergence damping -off%				Total infection	
	Pre %	Pre %	Post %	Post %		
	2010	2011	2010	2011	2010	2011
Fusarium profiraitum	15	10	0	0	15	10
Control	22.5	20	2.5	2.5	25	22.5
Fusarium oxysporum	20	15	0	0	20	15
Control	30	22.5	2.5	2.5	30	25
L.S.D. 5%	6.665	5.495	4.618	4.618	6.53	7.658

Table 12. Effect of the leaves powder of sweet orange on the damping off soybean caused by *Fusarium solani* and *Fusarium moniliforme*. Under greenhouse conditions during growing seasons2010and2011

Fungi	Emergence damping -off%				Total Infection	
	Pre %	Pre %	Post %	Post %		
	2010	2011	2010	2011	2010	2011
Fusarium solani	27.5	20	2.5	2.5	30	22.5
Control	37.5	30	5	5	42.5	35
FusariumMoniliforme	32.5	27.5	2.5	2,5	35.2	30
Control	40	35	2.5	7.5	42.5	42.5
L.S.D.5%	6.665	6.665	3.999	5.496	7.54	10.664

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Developing Management Strategies in Boron Toxic Soils from Central Anatolia of Turkey

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Abstract

The main objective of this research was to determine spatial variability in a boron toxic field irrigated by pivot sprinklers to develop a management strategy based on spatial variability of soil properties and rye performance. The location of study area was in Niğde Province, Turkey. Topsoil 0-30 cm) and subsoil 30-60 cm) were separately sampled to describe the variability at each of 56 predetermined sampling points based on a regular grid design in 43 ha area. The major constrains in plant production were high pH and boron contents of the soils. Average soil pH values were 8.29 and 8.82 and boron contents were 24.61 ppm and 56.29 ppm for surface and subsurface soils, respectively. Soil pH showed the lowest variability (CV=3.60% for top soil and 3.48% for subsoil), boron content showed the highest variability (CV= 67.94%) in surface soils of the study area. Boron content in surface soils had high significant positive correlations with electrical conductivity (EC) (r=0.737, P<0.01) and clay content (r=0.520, P<0.01), and negative correlations between sand content (r=*0.575, P<0.01) and aggregate stability (r=-0.378, P<0.01), whereas no significant correlation occurred between boron content and any of the textural components in subsurface soils. Range values indication of spatial correlation distance varied from 6.7 for organic matter to 704 m for silt content in surface soils. Clay content had the strongest spatial dependence in subsurface soils. Soil pH in surface soils and EC in subsurface soils showed the lowest spatial dependence based on sill over nugget variance ratio. The results revealed that the reclamation and management of problematic soils needs to be planned based on spatial distributions of soil properties. Since soil characteristics show spatial dependence, defining the variability in the field using classical statistical methods that assume the soil properties to have random variabilities might result in application of inappropriate management strategies.

Keywords: Boron, spatial distribution, geostatistics, variability, management

Mercury forms in typologically varied soils from arable lands of northern Poland

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Abstract
To understand the behaviour of mercury in soils and assess the risk of its toxicity, the forms in which the element is found in the environment must be detected. In the occurrence of favourable alkylation conditions (for example in floodplain soils) the content of methyl and ethyl mercury is higher. These Hg forms in comparison to mineral compounds, are toxic for the same organisms in concentrations from 10 to 100 times lower. The research concerned 8 selected soil profiles representing 6 types of soils (*Endogleyic Phaeozem, Endogleyic Fluvisol, Haplic Fluvisol, Cutanic Luvisols, Haplic Arenosol, Folic Podzol*), of different genesis used in plant production. The content of mobile (water soluble), available (DTPA-extractable) and bound with organic matter (NaOH-extractable) Hg forms were determined after thermal decomposition using AMA mercury analyser. The mobility as well as availability of mercury in the analysed soils was very low, with average percentage to 0.28 % and 2.45 % of the total content of this metal respectively. It was mainly dependent on texture, the amount of organic matter and soil pH. The percentage of mercury bound with organic matter ranged from 2.34 to 73.70 % of the total content of this metal, and was correlated with amount of clay and Fe oxides. Considering these results, the hazard of migration of this element into deeper horizons of the soil profile and ground water is very low. Moreover, crops from the investigated area are not at risk from mercury contamination.
Keywords: mercury, mobility, availability, soils, AMA.

Introduction

Mercury possesses unique physicochemical properties when compared to all chemical elements, and the variety of compounds it forms makes it both a very difficult and interesting object of study (Fitzgerald, 1995; Lindqvist et al., 1991; Schlüter, 2000; Schuster, 1991). Toxicity of mercury is linked to the form in which it is fund in the environment. Organic Hg compounds (methyl mercury and ethyl mercury mostly), in comparison to mineral compounds, are toxic for the same organisms in concentrations from 10 to 100 times lower (Boening, 2000; Gochfeld, 2003). The form of the highest toxicity, and at the same time of the highest bioacumulation is methyl and dimethyl mercury (Alloway, 1995; Gochfeld, 2003). Mercury is bioacumulated very easily. Plants accumulate relatively small amounts of Hg, much more animals (especially water animals) and fungi (Boening, 2000; Campbel et al., 2003; Kabata-Pendias and Pendias, 2001; Tsuchiya et al., 2008). This metal is accumulated mainly in plant roots, from which it is transported in varied pace to the parts above the surface (Boening, 2000; Gochfeld, 2003).

Mercury undergoes very specific transformations in different ecosystems. It is crucial to observe the content of different forms of this metal in soils. This is correlated with the variety of structure and composition and the dynamics of the soil system. This stems from different genesis of soils, as well as the variety and changeability of environmental conditions and anthropopression in spatial and temporal aspect.

The aim of this study was to assess the influence of selected soil components and physico-chemical properties affecting distribution, environmental mobility and potential toxicity of mercury in typologically varied soils used in plant production.

Materials and methods

The research material was collected from samples taken from 8 selected soil profiles representing 6 types of soils: one profile of *Endogleyic Phaeozem*, one profile of *Endogleyic Fluvisol*, one profile of *Haplic Fluvisol*, three profiles of *Cutanic Luvisols*, one profile of *Haplic Arenosol,* and one profile of *Folic Podzol* (IUSS 2007), formed form parent material of various origin. The analysed soils are arable soils which are used for cultivating plants typical for the region (sugar beet, wheat, rye). Only Haplic Podzol is a forest soil located in the area of relatively limited anthropogenic influence. The results presented are a continuation and extension of research on the content of mercury in the soil environment presented by Różański (2009) together with complete data on composition, physico-chemical properties, and the total content of mercury.

The content of water soluble, mobile mercury forms (Hg_{H2O}), extracted using distilled water (soil/water 1:2.5), the content of available mercury extracted by 1M Diethylene triamine pentaacetic acid (Hg_{DTPA}) (Lindsay and Norvell, 1978), and the content mercury bound to the soil organic matter extracted by 1M NaOH (Hg_{NaOH}) (Schnitzer and Khan, 1978) were determined after thermal decomposition in 700°C using AMA 254 mercury analyser.
All the results of the mercury content in soil samples were verified using certified material (reference soil sample TILL-3 and SO-4, Canada Center for Mineral and Energy Technology).

Results and discussion

The content of water soluble mercury forms in soils

It is assumed that the water soluble mercury compounds are mainly salts - eg. $HgCl_2$, $HgBr_2$, $Hg(CN)_2$, $Hg_2(C_2H_3O_2)_2$ (Barnett and Turner, 2001; Han et al., 2003; Schroeder and Munthe, 1998), and also low molecular weight organic Hg compounds (Wallschläger et al., 1998a). It should however be noted that the solubility of mercury compounds depends to a great extent on the composition and properties of the soil solution. For example cinnabar (HgS), which is sparingly soluble, may dissolve in solutions containing dissolved organic matter (fulvic acids) or compounds with thiol ligands (Jacobson et al., 2005). The content of soluble fractions in soils does not usually exceed 4% of the total humus content (Henderson et al., 1998).
The content of mobile, water soluble mercury forms was very low in the examined soils and ranged between 0 and 0.82 $\mu g \cdot kg^{-1}$ (table 1). The percentage of Hg_{H2O} in the total content of this metal was on average 0.28 %. Low share of readily soluble Hg compounds indicates that their migration into the deeper soil horizons is low. The highest content of Hg_{H2O} was found in surface horizons, especially organic horizons of *Folic Podzol* (profile 8). Slightly different results were found for the polluted soils of Europe, where the highest content of mobile mercury forms was found in subsurface horizons (below 20 cm). This was linked with Hg being washed together with humus acids from surface horizons to subsurface horizons, where these were sorbed by mineral components of soil sorption complex (Biester et al., 2002a, 2002b).
Forest soils of northern Poland were characterised by higher content of Hg than the content found in this research (Malczyk, 2000), while in the mercury polluted soils of Great Britain, the concentration of Hg_{H2O} was so low that it did not exceed the detection level in the analytic method used (Penyamentheekul, 2004). In the soils of central Spain the content of Hg_{H2O} did not exceed the level of 0,025 $\mu g \cdot kg^{-1}$ (using both the AMA and ICP-MS method) (Sánchez et al., 2005).
The content of readily soluble mercury compounds was positively correlated with the content of organic carbon ($r=0.51$; $p<0.05$, table 2). The lowest contents of Hg_{H2O} were determined in surface horizons of the analysed soils, rich in organic matter (except profile 6 and 7). The greatest percentage of water soluble forms in the total content of Hg was found in horizons with considerably low content of organic matter and poor in clay fraction (especially in *E* horizons of *Cutanic Luvisols*, profiles 5 and 6). Such results indicate considerable influence of both organic matter and clay fraction on the content of Hg_{H2O}.
The results of the research indicate that the amount of readily soluble Hg forms was relatively low in relatively heavy-textured soil, rich in clay minerals (profile 3). When comparing the content of Hg_{H2O} in surface horizons in both analysed *Fluvisols* (profiles 3 and 4) it could be concluded that the percentage of these Hg forms, despite comparable content of organic carbon, and considerably high total content of mercury in heavy-textured *Endogleyic Fluvisol* (profile 3), was significantly higher in fine-texture *Haplic Fluvisol* (profile 4, table 1). Clay fraction was therefore very influential in binding mercury in these soils. Moreover, the content of the analysed *Fluvisols* suggests, that in soils rich in humus and clay minerals, as a result of adsorption of positively charged mercury cations on the negatively charged surface of humus compounds and clay minerals, sparingly soluble in water complexes Hg-humus-clay minerals may be formed. Mercury is hence bound to the solid phase of the soil. This could account for a relatively high content of Hg_{H2O} in organic horizons (*Oi, Oe* and *Oa*) of the *Folic Podzol* (profile 8), in which no clay minerals were found. The above hypothesis stating the role of clay minerals in binding mercury in soil is also confirmed by other authors (Biester et al., 2002b; Boszke et al., 2004; Inácio et al., 1998).

Table 1. The content of mercury forms in analysed soils.

Profile no	Soil	Horizon	Depth	Hgtot.*	Hg_{H2O}	Hg_{DTPA}	Hg_{NaOH}	Hg_{H2O}	Hg_{DTPA}	Hg_{NaOH}
			[cm]	[µg·kg^{-1}]				[%]		
1	*Cutanic Luvisol*	Ap	0-22	48.62	0.23	0.27	35.83	0.47	0.56	73.70
		Bts	22-61	36.26	0.06	0.13	16.68	0.17	0.36	46.01
		Ck	61-95	19.07	0.02	0.15	8.99	0.10	0.78	47.13
		2Ck	<95	19.50	0.05	0.12	7.86	0.26	0.64	40.31
2	*Endogleyic Phaeozem*	Ap	0-35	36.66	0.20	0.39	23.46	0.54	1.07	63.99
		AC	35-48	21.66	0.09	0.28	8.83	0.41	1.29	40.77
		Cg1	48-95	23.26	0.00	0.31	9.46	0.00	1.32	40.65
		Cg2	95-140	17.60	0.01	0.44	6.43	0.06	2.52	36.51
		Cgk3	<140	18.13	0.02	0.32	3.82	0.11	1.74	21.07
3	*Endogleyic Fluvisol*	Ap	0-20	1438.00	0.18	0.20	33.70	0.01	0.01	2.34
		AC	20-45	280.50	0.05	0.21	113.82	0.02	0.07	40.58
		Cg1	45-70	384.70	0.08	0.19	159.37	0.02	0.05	41.43
		Cg2	70-100	292.00	0.04	0.24	142.49	0.01	0.08	48.80
		Cg3	<100	238.40	0.04	0.24	96.70	0.02	0.10	40.56
4	*Haplic Fluvisol*	Ap	0-15	142.90	0.42	0.49	76.00	0.29	0.35	53.18
		C1	15-55	80.60	0.08	0.12	43.97	0.10	0.15	54.56
		C2	55-73	88.93	0.04	0.12	48.36	0.04	0.14	54.38
		C3	73-90	71.90	0.03	0.14	35.23	0.04	0.19	49.00
		C4	<90	3.73	0.04	0.38	1.18	1.07	10.19	31.59
5	*Cutanic Luvisol*	Ap	0-26	26.93	0.13	0.61	15.62	0.48	2.26	57.99
		E	26-36	16.01	0.08	0.28	6.51	0.50	1.76	40.68
		E/B	36-57	21.60	0.04	0.21	9.06	0.18	0.97	41.96
		Bts1	57-90	22.15	0.04	0.10	7.32	0.18	0.47	33.04
		Bts2	90-120	33.04	0.01	0.09	8.77	0.03	0.26	26.54
		Ck	<120	17.67	0.01	0.11	4.64	0.06	0.62	26.26
6	*Cutanic Luvisol*	Ap	0-27	26.61	0.09	0.30	14.01	0.34	1.13	52.66
		Eg	27-40	14.19	0.10	0.34	4.90	0.70	2.36	34.53
		Btgs1	40-76	36.85	0.08	0.08	10.79	0.22	0.22	29.28
		Bts2	76-105	29.38	0.09	0.09	6.68	0.31	0.31	22.74
		BC	105-135	31.30	0.02	0.09	7.73	0.06	0.29	24.71
		Ck	<135	17.08	0.02	0.16	4.02	0.12	0.94	23.52
7	*Haplic Arenosol*	Ap	0-29	24.64	0.09	2.20	14.36	0.37	8.92	58.26
		A/B	29-37	22.20	0.11	0.53	13.03	0.50	2.40	58.71
		Bs	37-65	21.71	0.05	0.36	11.55	0.23	1.64	53.19
		BC	65-77	8.95	0.09	0.29	2.75	1.00	3.21	30.68
		C	<77	6.98	0.10	0.28	0.78	1.43	3.96	11.22
8	*Folic Podzol*	Oi	10-9	126.50	0.59	5.45	58.40	0.46	4.31	46.17
		Oe	9-3	266.70	0.38	1.21	85.86	0.14	0.46	32.19
		Oa	3-0	322.00	0.82	6.83	76.06	0.25	2.12	23.62
		AE	0-12	21.12	0.09	1.46	9.93	0.43	6.93	47.00
		Bh	12-18	19.56	0.01	0.99	8.82	0.05	5.08	45.11
		Bs	18-36	23.11	0.02	0.73	12.57	0.09	3.14	54.38
		B/C	36-84	7.40	0.00	0.98	1.81	0.00	13.26	24.52
		C1	84-125	6.88	0.00	0.74	1.40	0.00	10.79	20.39
		C2	<125	5.56	0.01	0.61	0.85	0.18	10.98	15.34

* - Total content of mercury published by Różański (2009).

The content of available forms of mercury

The contents of DTPA-extractable metals in soils are considered as fractions available for plants (Kabata-Pendias and Pendias, 2001; Lindsay and Norvell, 1978). The percentage of Hg_{DTPA} in the total content of mercury was low, and ranged between 0.01 to 13.26 % (2.45 % on average, Table 1). A comparable participation of available forms was concluded by Barnett and Turner (2001) in the soils polluted by this metal (0.3-14 %, 3.2 % on average), however, the horizons with the greatest amount of these forms were subsurface horizons. In the profiles of these examined soils, the horizons with the greatest participation of available forms of mercury in the total content of this metal were both surface horizons (profiles 5 and 7) as well as deeper horizons (usually the parent material horizons – profiles 1, 2, 3 and 4).

Table 2. Statistically significant correlation coefficients ($p < 0.05$).

Hg form	$Hg_{tot.}$	Organic carbon	pH H_2O	pH KCl	H^+	$Fe_{tot.}$	Fe_d	Fe_o	Clay	CEC
Hg_{H2O}		0.51								
Hg_{DTPA}			-0.72	-0.58	0.81	-0.59				-0.49
Hg_{NaOH}	0.90	0.77					0.44	0.74	0.40	0.66

The concentration of bioavailable forms of Hg in the examined soils ranged between 0.09 and 2.20 $\mu g \cdot kg^{-1}$ in mineral horizons (0.39 $\mu g \cdot kg^{-1}$ on average) and from 1.21 to 6.83 $\mu g \cdot kg^{-1}$ in organic horizons (4.50 $\mu g \cdot kg^{-1\ on}$ on average, table 1). The profile distribution of Hg_{DTPA} was varied. No statistically significant relation was concluded between the content of this mercury fraction and its total content (insignificant correlation coefficient). Significant correlations were calculated between Hg_{DTPA} and acidity (pH in H_2O, pH in KCl, the total content of exchangeable H^+ cations), total content of Fe and the content of clay fraction (r=-0.72, r=-0.58, r=0.81, r=-0.59, r=-0.49; $p<0.05$ respectively (Table 2).

In the examined soils of acid reaction (*Haplic Arenosol* and *Folic Podzol*, profiles 7 and 8) the content of available, DTPA extractable mercury forms was higher in comparison to soils with neutral reaction (*Endogleyic Phaeozem* and *Fluvisols*, profiles 2, 3 and 4). Higher content of Hg_{DTPA} with acid reaction may be the result of the release of mercury from Fe and Al complexes (Schlüter, 1997). Moreover, in the acid reaction increases the content of soluble, low molecular weight fulvic acids, mainly responsible for binding of Hg in the soil solution (Biester et al., 2002b; Wallschläger et al., 1998a). It is also important that the fluctuations of pH more influence methyl mercury than Hg^{2+} sorption (Boszke et al., 2003).

A relatively low content of DTPA extractable mercury should be noted in *Fluvisols* and *Bts* horizons of *Luvisols* (profiles 3-6). These soils were characterized by considerably high content of clay fraction and amorphous Fe oxides in comparison to other examined samples. High content of clay fraction and Fe_o, which form soil sorption complex, lead to binding of mercury by solid phase of the soil (Boszke et al., 2003; Dreher and Follmer, 2004), yet in low amount of organic matter it does not limit its bioavailability (exchangeable form of Hg) (Biester et al., 2002b; Wang et al., 1997). The process of mercury binding by clay minerals is observed in soils containing minimum 15% of clay fraction (Wang et al., 1997).

As for the toxicity of mercury, forming compounds with alkyl groups (the most often methyl group) is a very important process. It usually takes place in alluvial soils, in which this process, due to high content of organic matter, high level of ground water and seasonal floods may occur quite fast (Montgomery et al., 2000). Such a direction in transformation of mercury compounds may have influenced the distribution of DTPA extractable mercury forms in the profiles of *Fluvisols* and *Endogleyic Phaeozem* (profiles 3, 4 and 2). In horizons of gleyic characteristics (*g*) in *Endogleyic Phaeozem* (profile 2), heavy-textured *Endogleyic Fluvisol* (profile 3) and in *Cutanic Luvisol* (profile 6), the percentage of Hg_{DTPA} forms was greater in the entire profile. This could be the result of *in situ* formed alkyl mercury compounds, and the migration of mobile mercury forms from higher horizons (Barnett and Turner, 2001; Montgomery et al., 2000). The consequence of the occurrence of favourable alkylation conditions may be the increase in the content of especially toxic methyl and ethyl mercury, which as bioavailable forms constitute greater danger for all living organisms (Boening, 2000; Han et al., 2003; Tsiros and Ambrose, 1999). According to Gilmour and Henry (1991) and Petersonet et al. (1990) even in such conditions, the share of alkyl mercury forms usually does not exceed 3% of the total content of this metal in soils.

The content of mercury bound with organic matter

According to some authors (Biester et al., 2002b; Dmytriw et al., 1995) the forms of mercury extractable by NaOH, are mainly typical humus compounds.

The concentration of Hg_{NaOH} in the analysed soils ranged between 0.78 and 159.37 $\mu g \cdot kg^{-1}$, and the profile distribution was proportionally close to the distribution of total Hg content (table 1). It confirms the positive significant correlation coefficient between $Hg_{tot.}$ and Hg_{NaOH} ($r=0.90$; $p<0.05$, table 2). Similar results were determined by Malczyk (2000) in unpolluted forest soils. This indicates that organic matter plays a dominant role in binding of mercury in soils. Mercury often forms stable complexes with organic ligands of stability constant ranging from 18.4 to 22.1 (Stein et al., 1996).

Due to the complexity of the organic matter transformations in soil, scientific reports give contradictory information in this aspect. Wang et al. (1997) observed that the increase of humus in soils affects the decrease of Hg content in plants, which could indicate strong binding of this element by organic matter. Montgomery et al. (2000) however find relatively high concentration of mobile and available mercury forms in soils with comparatively high amount of organic matter. The influence of soil humus on binding of mercury is dependent on the clay fraction content, and if clay fraction is high may even play the dominant role in this process (Inácio et al., 1998; Wang et al., 1997).

The proved lack of pH influence on the content of Hg_{NaOH} in the analysed soils (insignificant correlation coefficient) may be caused by either the binding of mercury by organic matter regardless of pH value, or by the fact that determined pH range favoured such complexation. Gabriel and Williamson (2004) noticed the dominant influence of organic matter on binding mercury in soils, in which pH was lower than 7. This process occurred in both aerobic and anaerobic conditions.

The percentage of Hg_{NaOH} forms in the examined soils ranged from 2.34 to 73.70 % of the total content of mercury (Table 1). Other authors indicate that in some soils the organic matter binds about 30% (Munthe et al., 2001), and even 80–85% of mercury (Dmytriw et al., 1995; Henderson et al., 1998; Rennenberg and Dudas, 2001).

The highest percentage of Hg_{NaOH} in the total content of mercury was determined in surface horizons of the examined soils. The share decreased with the depth in the soil profiles. Such Hg_{NaOH} distribution was mainly connected with the content of organic carbon in profiles of soil horizon, which is confirmed by a significant positive correlation coefficient between this form of mercury and organic carbon content ($r=0.77$; $p<0.05$, table 2). A rather low percentage content of mercury bound with organic matter in enrichment horizons of *Luvisols* (profiles 1, 5 and 6) could be a result of formation of Fe-clay complexes responsible for binding mercury in these horizons, and not of greater Fe-humus-clay or humus-clay complexes (Dmytriw, 1995; Schlüter, 1997).

In *Fluvisols* the percentage of Hg_{NaOH} in the total content of mercury in separate layers, despite varied concentration, was relatively even. An exception was the surface horizon of *Endogleyic Fluvisol* (profile 3), in which the percentage of this mercury form (2.34%) was the lowest in all of the horizons of the examined soils (in the remaining horizons of profile 40.56-48.80%). Also a considerably lower percentage of Hg_{NaOH} as compared with the remaining part of the profile was determined in the deepest horizon of the *Haplic Fluvisol* (31.59%, in the remaining part of the profile 49.0-54.56%, profile 4). This was most probably due to different texture of these horizons in comparison to the rest of the profile (especially varied content of clay fraction), or the difference in humus composition (Boszke et al., 2004; Wallschläger et al., 1998b).

Statistically confirmed correlations between the content of Hg_{NaOH} and the content of clay fraction as well as free (Fe_d) and amorphous (Fe_o) ferric oxides ($r=0.40$, $r=0.44$, $r=0.74$; $p<0.05$ respectively, table 2) may result from the fact that during extraction procedure (1M NaOH), the solution contained exchangeable forms of mercury, bound to these soil elements (Wang et al., 1997). Still their percentage in soils does not usually exceed 3% of the total content of mercury (Panyametheekul, 2004). Moreover, the application of 1M NaOH solution, in comparison to other reagents used for extraction of typical humus fractions (eg. $Na_4P_2O_7$) allows to determine mercury bound with organic matter, the amount of which is the closest to the actual content of this Hg form in soil (Hall and Pelchat, 1997; Schnitzer and Khan, 1978).

The content of Hg_{NaOH} forms was significantly, positively correlated with cation exchange capacity ($r=0.66$; $p<0.05$, table 2), which is linked with high sorption capacity of organic matter (Gabriel and Williamson, 2004) and high affinity of Hg to functional groups containing sulphur (Kabata-Pendias and Pendias, 2001; Xia et al., 1999). Skyllberg et al. (2003) proved in their research that from 50 to 70% of the total sulphur content in soils was included in the functional groups (mainly thiol) of organic matter, which bind metallic mercury as well as alkyl compounds very easily.

Conclusions

The availability of mercury for plants in the analysed soils was very low, on average amounting to 2.45% of the total content of this metal. It was mainly dependent on texture, the amount of organic matter and soil pH. In the horizons rich in both organic carbon and clay fraction the share of water soluble Hg and DTPA-extractable forms was the lowest. Considering low percentage of mobile mercury forms in total content, the risk that the metal shall migrate into deeper horizons of the soil profile and ground water is rather little. Moreover, even in soils with comparatively high total content of mercury, its availability for plants is very low. Only considerable decrease of pH value simultaneously with more intensive mineralization of organic matter, responsible for binding majority of mercury in the analysed soils (up to 73.7%), could significantly affect mobility and/or toxicity of this metal.

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Effect of Two Pesticides on the Content of Some Heavy Metals in Clay Soil

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Abstract:
Pesticides are considered one of the main sources adding heavy metals to the soil. Commercial samples of a fungicide (copper hydroxide) and an insecticide (aldicarb) were collected to evaluate their contents of certain heavy metals (Cu, Zn, Cd, Ni, Pb, and Cr). The sample of the fungicide, copper hydroxide contained 48.00, 28.19 and 27.64 ppm of Ni, Pb and Cr, respectively. While the sample of the insecticide, aldicarb contained high concentration of Cd (14.21 ppm). The two tested pesticides were applied to the soil either once or three times (two weeks intervals). All experiments were carried out on a clay soil in the experimental farm of Assiut University, Assiut, Egypt. Soil samples were collected from different depths to determine the heavy metal contents (Cd, Cr, Ni, Pb, Cu and Zn). The obtained results indicated that the soil surface layer was the most contaminated layer with the investigated metals. Both tested pesticides raised the amounts of extractable Cd, Ni, Cu, Cr, Pb and Zn of soil samples. Levels of extractable Cd and Cr in the soil were significantly affected by the application time of the pesticide.
Keywords: pesticides, heavy metals, clay soil.

Introduction
Heavy metal-contaminated pesticides may affect the contents of soils and crops of these metals resulting in a probable increase of their harmful impacts on human health through the food supply chain. Even low annual accumulations may finally build up undesired concentrations in soil, especially where pesticides with high heavy metal or rare earth element concentrations are used (Mortevedt, 1987). Degradable chemicals, such as pesticides are, sometimes, potentially hazardeous to some forms of life. In New Zealand, dairy product samples contaminated with certain pesticide residues were also found to be contaminated with some heavy metals (Morrison 1988).
One source of soil pollution results from the accumulation of the repeated application of pesticides. Gimeno Garcia et al. (1996) found that levels of Cd in two herbicides and one fungicide were similar to its content in superphosphate fertilizer; highest levels of Fe, Mn, Zn, Pb and Ni were in the herbicides. In their studies on the pesticide residues and heavy metal content of soils, plants and wastewater in Karak, Jordan, Jiries et al. (2002), indicated that the investigated area was heavily contaminated with heavy metals and various types of pesticides and their metabolities. Some investigators have suggested that there is a relationship between the levels of heavy metals and the persistence of pesticides (Schuhmacher et al., 1997 and Gimeno-Garcia et al., 1996).
The objective of this study is to evaluate levels of some heavy metals in some pesticides that are widely used in Egyptian agriculture and there effect on the amount of extractable heavy metals.

Materials and Methods
Samples of the two tested pesticides (Copper hydroxide 77% W. P. and Aldicarb 10% granules) were collected to evaluate their contents of some heavy metals (Cd, Ni, Cr, Pb, Cu and Zn). Determinations of heavy metals in the pesticide samples were replicated three times. Five grams of samples were digested using 8 ml of concentrated hot nitric acid (Shaffer, 2001) and made up to 50 ml with distelled water. Then, the solution was filtered and centrifuged at 3000 Rpm for 10 min using the centrifuge (Universal 16) and filtered again. A sample containing all added chemicals except for pesticide was served as control. Heavy metal concentrations in the control sample were negligible. Heavy metals were determined in the two pesticide samples using the atomic absorption spectrophotometer (GBC 906 AA).
In order to evaluate the application effect of pesticides on heavy metal contents in the soil, an experiment was conducted at Assiut Experimental Station, Faculty of Agriculture, Assiut

University. Physical and chemical properties of the soil of the experimental site are summarized in Table (1)
Both pesticides were applied to the soil or sprayed on the plants (carot and pea plants) once (after 4 or 8 weeks from planting) or at three repeated times (after 4, 6 and 8 weeks from planting). The pesticides were either applied to the soil just before irrigation (aldicarb) or sprayed on the foliage (copper hydroxide) using knapsack sprayer fitted with one nozzle. A control treatment was made by applying water on the plants. The treatments were set up in a split-plot design. The main plots were allocated for pesticide treatment. The time of application treatments were in the sub-Plots. Each sub-Plot area was 1/400 fed. Each treatment was replicated 3 times. All agricultural practices were equally followed as recommended.
Pesticides used:
Cooper hydroxide 77% W.P.
Aldicarb 10% granules :2-methyl-2-(methylthio) propanal O-[(methylamino) carbonyl] oxime.

Table (1): Some physical and chemical properties of the experimental Soils.

Soil properties	Values
* Particle size distribution	
Sand %	19.30
Silt %	31.00
Clay %	49.70
Texture	Clay
* Field capacity (F. C.)%	42.7
* pH 1 : 1 (suspension)	7.88
* ECe (m mhos/cm)	1.42
* $CaCO_3$ %	3.13
* $NaHCO_3$ extractable P (ppm)	16.4
* Soluble ions (meq/100 gm)	-
$Ca^{++} + Mg^{++}$	0.58
Na^{+}	0.69
K^{+}	0.09
$CO_3^{2-} + HCO_3^{-}$	0.35
Cl^{-}	0.82
SO_4^{2-}	0.12

Soil samples were collected from three depths (0-5, 5-15 and 15-30 cm), air-dried, crushed to pass through a 2 mm sieve, and extracted using 0.1 M diethylene triamine penta acetic acid (DTPA) according to Lindsay and Norvell (1978). Cd, Ni, Cr, Pb, Cu and Zn were determined in soil extracts using a GBC 906AA Atomic Absorption Spectrophotometer.
Data were statistically analyzed according to Steel and Torrie (1980).

Results and Discussion

Heavy metals content in tested pesticides

Table (1) shows the content of heavy metals in the two tested pesticide samples. The insecticide aldicarb was more contaminated than copper hydroxide with Cd and Zn by 11.27 and 7.69 fold, respectively. While copper hydroxide was more contaminated than aldicarb with Pb, Cu, Cr and Ni by 26.35, 7068, 5.31 and 14.63 fold, respectively. Gimeno-Garcia et al. (1996) studied the levels of Cd, Co, Cu, Ni, Pb, Zn, Fe and Mn in different pesticides. Thier results showed that three tested pesticides contained high levels of Fe, Mn, Zn and

Table1. Heavy metal contents of the two tested pesticides .

Pesticide	Formulation type	Container type	Cd	Pb	Cu	Cr	Ni	Zn
			ppm					
1-Fungicides								
Copper hydroxide	W.P	Aluminum	1.26	28.19	405000.0	27.64	48.00	131.33
2-Insecticides								
Aldicarb	G	Aluminum	14.2	1.07	57.3	5.21	3.28	1010.5

Ni.Schuhmacher et al. (1997) indicated that the levels of heavy metals in soils near by pesticide factory were increased
with decreasing the distance from pesticides factory. In the present study, pesticide contamination with heavy metals may be attributed to the presence of some impurities with the pesticide forms especially those that all tested pesticides were formulated from chemical compounds. It is known that formulated pesticides contain some adjuvants, detergents or emulsifiers, which may include some heavy metals. The purity of active ingredients prepared to be added to the mixture of the other materials to produce the final pesticide formulation is usually less than 100%. So, probably there is some traces of unknown compounds even in very low concentration, but enough to contaminate the final formulation of pesticides with detectable amounts of heavy metals.

Effect of pecticides application on heavy metals availability

Most heavy metal levels were higher in the treatment with copper hydroxide and aldicarb compared with the control in both types of soil. The level of Cu was higher, as expected, with the application of Copper hydroxide (2.9 and 2.51 ppm) than the other heavy metals. These relatively high levels of extracted heavy metals may be attributed to the fact that some pesticides contain some heavy metals either as a part of their formulas (Copper hydroxide contains about 50% Cu) or as impurities.

Table (2): Effect of pesticide type on the DTPA-extractable heavy metals (ppm) of the soil cultivated with carrot and pea plants.

Pesticide type	Carrots						Peas					
	DTPA-extractable heavy metals (ppm)											
Heavy metal	Ni	Pb	Cd	Cr	Cu	Zn	Ni	Pb	Cd	Cr	Cu	Zn
Control	0.17	0.23	0.004	0.01	1.19	0.64	0.17	0.26	0.004	0.02	1.19	0.63
Copper hydroxide	0.23	1.07	0.021	0.03	2.09	0.68	0.20	0.86	0.016	0.27	2.51	0.62
Aldicarb	0.16	1.04	0.014	0.07	1.91	0.64	0.21	0.82	0.022	0.19	2.19	0.67
F-value	**	**	**	**	**	**	**	**	**	**	**	*

Distribution of DTPA-extractable heavy metals with soil depth

The distribution of DTPA-extractable Ni, Pb, Cd, Cr, Cu and Zn with depth in the soil cultivated with carrots and peas are present in Table 3. The surface layer (0-5 cm) was always the most contaminated layer with all tested heavy metals because of the repeated additions of contaminant sources of heavy metals. El-Desoky and Ghallab (2000) reported that DTPA-extractable Pb was decreased with increasing soil depth due to the contamination from a superphosphate factory. Levels of extractable Ni, Cd, Cr, Zn and Cu were significantly higher in the surface layer of the soil cultivated with peas (0.32, 0.024, 0.33, 0.74 and 2.45 ppm, respectively) compared with those of the soil cultivated with carrots (0.22, 0.020, 0.07, 0.71 and 1.91 ppm, respectively). This may be related to the variation in the root exudate composition between pea plants and carrots. Raskin et al. (1994) reported that plant roots can solubilize heavy metals by acidifying their soil environment with protons extruded from the roots. DTPA-extractable metals of both soils, cultivated with carrots and peas, significantly decreased with depth, except for Cd in the soil cultivated with carrots. One of the major factors governing cadmium, adsorption and distribution in soils is the presence of organic and inorganic ligands (OECD, 1994).

Table(3): Effect of soil depth on the DTPA-extractable heavy metals (ppm) of the soil cultivated with carrots and peas .

Soil depth (cm)	Carrots						Peas					
	DTPA-extractable heavy metals (ppm)											
	Ni	Pb	Cd	Cr	Cu	Zn	Ni	Pb	Cd	Cr	Cu	Zn
0 – 5	0.22	1.10	0.020	0.07	1.91	0.71	0.32	0.98	0.024	0.33	2.45	0.74
5 – 15	0.17	0.99	0.010	0.07	1.75	0.68	0.20	0.83	0.017	0.22	1.95	0.63
15 – 30	0.13	0.86	0.010	0.04	1.71	0.60	0.13	0.70	0.015	0.14	1.95	0.57
F-value	**	**	n.s	**	*	**	**	**	**	**	**	**
$LSD_{0.05}$	0.02	0.10	-	0.01	0.17	0.05	0.02	0.03	0.002	0.04	0.24	0.09

Influence of the application time of pesticide

DTPA-extractable heavy metals of the soil cultivated with carrots and peas as affected by the application time of the pesticide are shown in Table (4). Only levels of extractable Cd and Cr of the soil cultivated with carrots were significantly affected by the application time of the pesticide. Adding the pesticide once after 4 weeks from planting had the highest significant increase on the extracted Cd (0.019 ppm). Since the pesticide degradation is a time-dependant process, Cd from pesticides may release during this process from this treatment (after 4 weeks) resulting in a soluble form which is easy to be extracted. Chromium concentration in the soil was high (0.07 ppm) when pesticides were applied at 4 or 8 weeks after planting.

The extractable Cd of the soil cultivated with peas had high levels (0.20 ppm) at the early or late single pesticide application. The repeated applications of pesticides reduced the extracted Cd to 0.017 ppm. The repeated application might interfere with the degraded products of the former addition and interrupt releasing some heavy metals from the pesticide to the soil. Both early single application (after 4 weeks) and repeated applications (after 4, 6 and 8 weeks) increased the extractable Pb of the soil cultivated with peas. While the late single application (after 8 weeks) and the repeated applications increased the extractable Cr in this soil.

Table(4): Effect of time of pesticides applications on the DTPA-extractable heavy metals (ppm) in the soil cultivated with carrot and pea plants.

Application time	Carrots						Peas					
	DTPA-extractable heavy metals (ppm)											
	Ni	Pb	Cd	Cr	Cu	Zn	Ni	Pb	Cd	Cr	Cu	Zn
1	0.19	0.95	0.019	0.07	1.71	0.69	0.22	0.88	0.020	0.15	2.09	0.62
2	0.18	1.00	0.011	0.07	1.84	0.66	0.21	0.74	0.020	0.28	2.11	0.69
3	0.17	0.99	0.013	0.04	1.82	0.64	0.22	0.90	0.017	0.26	2.17	0.64
F-value	n.s.	n.s.	**	**	n.s.	n.s.	n.s.	**	**	**	n.s.	n.s.
$LSD_{0.05}$	-	-	0.006	0.01	-	-	-	0.09	0.002	0.06	-	-

1 after 4 weeks from palnting 2 After 8weeks of palnting

3 Repeated applications after 4,6 and 8 weeks from planting

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Heavy Metal Contents of Vineyards Soil in Alasehir, Manisa

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Abstract:
In Turkey, Sultana has been produced in especially Gediz Basin, Aegean region, Izmir, Denizli, Manisa and around (82%), has been provided (25%) from Alasehir, Manisa. Therefore, in consideration of the area's potential, the purpose of this work; it is to determine that total heavy metals (Fe, Zn, Mn, Cu, Ni, Co, Pb, Cd, Cr) contents, the level of contamination soil samples taken from vineyards which coached seedless raisin variety (*vitis vinifera L.*) as intense, in Alasehir, Manisa where is be semi-arid environment at Ege region. In this study, 26 soil samples where taken from both depths (0-30cm; 30-60cm) and from 13 garden had been taken in the period of the color term and analyzed. In terms of heavy metal contents, no Cd and Cr toxicity were determined in soils. In spite of that, there is no question Pb pollution in 93.2% of soil except number 2 which was detected pollution. But there is Co pollution in 23.1% and Ni pollution 15.4% of soil at first depth, at second depth, Co and Ni pollution in 53.8% and 30.8% of soil, respectively.

Keywords:Vineyards (*vitis vinifera L.),* Micro Element, Heavy Metal, Pollution, Contamination

Introduction

World of living things to division which has got a non-compliance with ecosystem around or any substance who gives a stress; is called "pollutants". In general, pollutants is domestic, industrial, agricultural and natural origin. In recent years, heavy metals which from segregation pollutants attracted the attention by investigators because it's hard, expensive to remove from soil and going to find out toxic effects on living things.

Industrial establishments and activites, which give rise to wastes including metal and heavy metal salts and agricultural applications (chemical fertilizer, pestiside, hormone) are the main sources of soil pollution (Forstner et al., 1991; Jarqusch-Wehrheim et al., 2001; Elmaci et al., 2002). Industrial wastes contain trace elements and heavy metals such as Fe, Zn, Cu, Mn, B, Pb, Cd, Ni, Cr and Co depending on the type of the establishment.

Trace element and heavy metal are microelement needed by animals and plants but they can become toxic above certain limits (Alloway, 1990). There is, thus, a complex movement of elements around the environment which needs to be monitored. In addition, the uptake of heavy metals by plants is soupled with factors like plant variety, species, plant part, age of organ or plant, growt period, stability to heavy metals and transfer factor of heavy metals (Fergusson, 1990; Secer et al., 2002).

Turkey takes place between rare grape producers and exporting country both amount of production and the width of vineyards areas and in terms of grape varieties because of it's the availability of ecological conditions in a semi-arid environment. Especially, Sultana has been produced in Gediz Basin, Ege region, Izmir, Denizli, Manisa and around (82%), has been provided (25%) from Alasehir town of Manisa. Also, vineyards which has got a round seedless grape variety *(vitis vinifera L.)* are forms the existing vineyards of 90% in region (Aydin et al., 2007). All kinds of wrong agricultural applications to make large a grapes, increase the yield and quality reveals some symptoms in product. This led to problems in foreign market over the years. Soil, water and air pollution which is defined as environmental pollutions are usually lead to soil pollution and negative effecf for quaity and quantity in plant production in especially agricultural areas. This threatens human health by food chain.

Therefore, in consideration of the area's potential, the purpose of this work ;is to determine total heavy metals (Cd, Co, Cr, Ni, Pb) contents, the level of contamination in taken soil samples where taken from vineyards which coached seedless raisin variety (*vitis vinifera L.*) as intense, in Alasehir town of Manisa where is be semi-arid environment at Ege region.

Materials and Methods

The Gediz plain has important agricultural potential in the Aegean region. According to the new soil taxonomy system the investigated soils can be classified as Typic xerofluvent soils (Altinbas et al., 1994) . In this study, 26 soil samples where taken from both depths (0-30cm; 30-60cm) and from 13 garden had been taken in the period of the color term and analyzed (Jackson, 1967). The places of vineyards are given Table-1. Samples of soil were taken in period of becoming coloured in August 2010. In soil samples, total heavy metal and some trace element contents (Cd, Co, Cr, Ni, Pb, Fe, Zn, Mn, Cu) were extract by aqua regia extraction method and has been determined in Atomic Absorption device (Slavin, 1968; Kick et al, 1980).

Table-1. Soil samples taken at locations

Sapmle No	Location	Name
1	Kovalık	Mehmet OK (10da)
2	Mezarlık	Mustafa Onal (4da)
3	Saglık ocagı	Mehmet Aktaş (6a)
4	Kovalık	Ahmet YILDIRIM (5da)
5	Sellik	Süleyman CEYLAN (10da)
6	River of Alasehir (Philadelphia)	Cevdet YAVUZ (8da)
7	River of Alasehir (Philadelphia)	Hüseyin BASTURK (8da)
8	River of Alasehir (Philadelphia)	Süleyman BASTURK (6da)
9	River of Alasehir (Philadelphia)	Faruk OZTURK (10da)
10	River of Alasehir (Philadelphia)	Cengiz OZTURK (8da)
11	River of Alasehir (Philadelphia)	İzzet CETİN (8da)
12	River of Alasehir (Philadelphia)	Hüseyin TOPALOGLU(10da)
13	River of Alasehir (Philadelphia)	Cengiz SAHİN (6da)

Results and Discussion

Total Micro Element and Heavy Metal Contents of Soil

Minumum, maximum and average values of total micro element and heavy metal (Fe, Zn, Cu, Cd, Co, Ni, Pb) contents of soil samples taken from vineyards plantations at both depths (0-30cm ve 30-60cm) are given in Table-2.

Limit values of Fe content are seen as 0.5-5.0% Fe to Scheffer and Schachtschabel (1989) in soils. According to these reference values; there is no toxicity problem in vineyard soils at both depths. In terms of total Zn; when 300 $mg.kg^{-1}$ value considered given by Kloke (1980) for soils, there is no question for Zn pollution at 0-30 cm and 30-60 cm depths of vineyard soils. For total Mn; when 1500 $mg.kg^{-1}$ - 3000 $mg.kg^{-1}$ of Mn values considered given by Pendias and Pendias (1992), in this context; ıt was determined that there is no any pollution in vineyard soils at both depths. Also, the remarkable amount of Mn which, is in the samples contained is well below of critical Mn value in this context. In studied soils, when the examined total Cu content which is extrated with the aqua regia ($3HCl+HNO_3$), Kloke (1980) reported that Cu as the critical value is 100 $mg.kg^{-1}$, Pendias and Pendias (1992) did that 60-125 $mg.kg^{-1}$. When we consider that critical values, Cu value were below those value in studied vineyard soils (Table-2). Total Cu contents of soils are generally showed up decrease from first depth to second depth. Tuna et al., (2005) obtained the same results which is the similar to our findings in terms of total Fe, Zn, Mn and Cu to investigate the heavy metal pollution in agricultural areas. Cd pollution which is caused by industry where is the intense in this location, especially from chemical fertilizers (superphosphate) and exhaust gases were seen distinct. Kloke (1980) and Pendias and Pendias (1992) explained the critical value of 3 $mg.kg^{-1}$ in terms of pollution of Cd. When we consider that value, we can say that there is no Cd toxicity in soils. Alloway, (1990) and Altinbas et al., (1994) explained that Cd is more intense in especially upper soil layers but mobility is more than Pb and Cu along the profile and shown up similar situation for Ni and Zn. In terms of total Co; there is no Co pollution according to the critical value (50 $mg.kg^{-1}$) given by Kloke (1980) in soils at the both depths, according to the critical values (30-60 mg.kg^{-1}), Co pollution was determined that 23.1% of soils at the first depth and at the second depth it is 53.8% of soils (Table-2).

Table 2. Total micro elements and heavy metal contents of soil samples taken from vineyard plantations

Sample No	Depth (cm)	Total Fe (%)	Total (mg.kg^{-1})							
			Cu	Zn	Mn	Co	Pb	Cr	Cd	Ni
1	0-30	1.89	38.5	112.4	495.0	28.0	69.3	28.2	2.26	43.8
	30-60	1.91	34.8	65.9	522.5	25.4	48.9	27.3	1.93	41.0
2	0-30	1.89	33.6	73.6	481.3	27.1	124.7	23.0	2.04	36.6
	30-60	1.78	35.8	62.0	495.0	28.0	185.4	25.3	2.04	38.0
3	0-30	1.86	24.9	62.0	357.5	22.3	40.3	25.0	1.82	37.2
	30-60	1.65	22.4	46.5	275.0	23.6	41.4	23.9	1.93	34.6
4	0-30	1.65	23.6	81.4	440.0	25.4	34.9	27.6	2.37	37.6
	30-60	1.54	22.9	73.6	288.8	25.8	34.9	26.2	2.36	38.3
5	0-30	1.69	34.9	62.0	316.2	20.6	22.6	23.9	1.21	33.4
	30-60	2.29	27.5	58.1	343.7	21.9	29.6	24.4	1.27	37.2
6	0-30	1.89	3.01	73.6	398.8	20.6	17.2	31.6	0.66	45.3
	30-60	2.08	23.6	100.7	343.8	18.4	9.7	27.9	0.61	34.8
7	0-30	2.81	32.7	143.3	426.3	22.3	16.7	35.1	0.72	47.2
	30-60	2.53	29.6	93.0	398.8	21.9	15.6	26.0	1.05	41.2
8	0-30	2.88	34.9	108.5	440.0	22.8	18.3	35.1	0.66	53.8
	30-60	2.96	39.7	155.0	495.0	25.8	19.9	41.7	0.88	50.4
9	0-30	2.67	30.1	174.4	343.7	23.1	14.5	34.2	0.83	34.4
	30-60	3.28	37.1	162.7	440.0	25.8	18.3	42.8	0.99	52.7
10	0-30	2.91	39.3	104.6	440.0	24.1	16.7	37.7	0.88	48.4
	30-60	3.02	35.2	132.6	360.0	25.6	17.2	39.4	0.96	56.5
11	0-30	2.12	45.2	93.0	412.5	21.0	19.9	36.8	0.88	44.8
	30-60	2.79	34.5	96.9	398.0	25.4	19.4	37.1	0.77	46.3
12	0-30	2.72	39.5	135.6	522.5	23.2	18.8	35.4	0.83	55.5
	30-60	2.72	37.9	166.7	495.0	24.1	18.8	34.2	0.77	56.1
13	0-30	3.00	35.1	135.6	440.0	24.5	15.6	38.5	1.05	45.7
	30-60	2.83	30.7	155.0	398.8	21.9	14.5	26.5	0.66	40.8

These were determined in most of the soils contained 5-1000 mg.kg^{-1} Cr and some cases where the concentration of up to 300 mg.kg^{-1} (Scheffer and Shachtschabel, 1989). In this context, there is no question from 75-100 mg.kg^{-1} Cr population given by Pendias and Pendias (1992) and 100 mg.kg^{-1} Cr pollution given by Kloke (1980). Pendias and Pendias (1992) explained the critical value is 100 mg.kg^{-1}, Kloke (1980) explained the critical value is 50 mg.kg^{-1} in terms of Ni for soils. When we consider the 50 mg.kg^{-1} Ni consantration as a critical value, Ni pollution was determined in the 15.4% of soils at the first depth, at the second depth in also 30.8% of soils (Table-2). Besides, amount of the Ni of soil samples taken from at the both depths, was seen with close to reported the limit value (50 mg.kg^{-1}), (Table-2). Altinbas et al., (1994) found between 28.7-86.2 mg.kg^{-1} total Ni in the agricultural soils of the Gediz region and in 2002, Elmaci et al (2002) 22.3-962.5 mg.kg^{-1} in the Manisa and Menemen plain. Also, especially Cd, Pb and Ni pollution was found to occur in relation with agricultural soil's distance from the highway as well as agricultural and industrial activities (Ndiokwere, 1984; Hakerlerler et al, 1994; Okur et al, 1995; Yagmur and Okur, 2011). Lead was found between 1-20 mg.kg^{-1} in polluted and 25-95 mg.kg^{-1} in slightly polluted soils (Scheffer and Schachtschabel, 1989). Kloke (1980) stated that the critical value of Pb pollution is 100 mg.kg^{-1} in soils. According to this critical value, there is a Pb pollution in the 92.3% of soils at the both depths, except vineyard which is number 2.The source of Pb pollution can be due to the use of phosphorus fertilizer, Pb containing fuel and Pb carried with air movements.

Conclusion

As a result; in generally, soil properties show up light alkaline and medium alkaline reaction, there is no saltness danger, show up that it is lime and sandy-loam texture and in terms of organic matter is poor. In terms of total N content, a large portion of soils are medium and good level, all of it is good in terms of P, available Ca and it is adequate 38.5% of soils in terms of available K. However, the fact that a large of portion of soils are light and medium alkaline reaction and it is close to half of some with lime reflected to be phosphorus fixation. In this regard, phosphorus fertilization was suggested to apply to the band and the 20-30 cm deep in vineyard where is located at Kavaklıdere. When the micro element contents of soils are examined, there is a inadequacy in terms of Fe and Zn and it was determined that available Mn and Cu contents are sufficient level. Available boron is high level to create a problem in 76.9% of examined soils. In addition, there is a boron pollution and boron toxicity in a large portion of vineyards because ground water of location has got a high boron. Therefore, the quality of irrigation water needs to be careful.

When the total micro element contents of soil examined in a aqua regia, there is no question for pollution in terms of total Fe, Zn, Mn and Cu. In terms of heavy metal contents, there are no Cd and Cr toxicities in soils. However, there is no problem for Pb pollution of 93.2% of soils except vineyard which is number 2. However, 23.1% of soils Co and 15.4% of soils Ni pollution were determined at the first depth, at the second depth 53.8% of soils Co and 30.8% of soils Ni pollution. While Co pollution is origin from industry organizations, Ni and Pb pollutions becomes from exhaust gases and it was thought to pass to the soil and plants by air pollution. The domestic and industrial wastes of Salihli where the industrial companies become dense and is near studied location, are emptied to the stream of Alasehir. So, there is a heavy metal pollution, intensely at the junction of Gediz river and Alasehir stream.

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Phytoremediation of Heavy Metal Polluted Soil in Contaminated Mine Sites, a Case Study from Izmir, Turkey

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Abstract
The effects of mine wastes can be multiple, such as soil erosion, air and water pollution, toxicity, geo-environmental disasters, loss of biodiversity, and ultimately loss of economic wealth. Mine reclamation has become important parts of the sustainable development strategy in many countries because there is a need to reduce the negative effects of post-mine areas by reclamation. Reclamation is the process to restore the ecological integrity of disturbed mine land areas. There are many approaches to rehabilitate the post-mining areas. Phytoremedion is more cost-effective and aesthetically pleasing than physical and chemical reclamation approaches. Phytoremedion reduce soil pollution and remedy the polluted soil. There are many mine sites in Izmir city which cause environmental damage and need reclamation. The aim of this study is to introduce phytoremediation technique and emphasize the suitability of contaminated sites in Izmir.
Keywords: *Soil reclamation, Phytoremediation, post-mining, Izmir, landscape reclamation, heavy-metals*

Introduction

Since the beginning of the industrial revolution, soil pollution by toxic metals has accelerated dramatically. According to (Nriagu, 1996) about 90% of the anthropogenic emissions of heavy metals have occurred since 1900; it is now well recognized that human activities lead to a substantial accumulation of heavy metals in soils on a global scale. Man.s exposure to heavy metals comes from industrial activities like mining, smelting, refining and manufacturing processes (Nriagu, 1996). Currently, conventional remediation methods of heavy metal contaminated soils are expensive and environmentally destructive (Bio-Wise, 2003; Aboulroos et al., 2006). But there are some remediation techniques which are nature friendly. These techniques using either chemical and or biological techniques (such as hyperaccumulator plants, or high biomass crop species after soil treatment with chelating compounds). The technologies applicable to a contaminated site depend on the cleanup goals, the form of pollutants presented the volume and physical/chemical properties of the polluted soils. In many cases soil cleanup goals depend on the concentration of pollutants (Sabour, 2007). In this study the remediation techniques are analyzed from different points of view for the environmental, aesthetical and economical aspects. Soil washing, solidification, in situ immobilization, chemical reduction and bioremediation are the analyzed techniques. This study is introducing the bioremediation method for post mining sites.
Bioremediation was considered because of being the most cost effective, sustainable, aesthetic method. Bioremediation is also full scale field application and long term applicable method. Bioremediation can be defined as any process that uses microorganisms, fungi, green plants or their enzymes to return the natural environment altered by contaminants to its original condition (Cunningham et al., 1997). In this study it is focused on the mining sites as a case study area because of being one of the most altered contaminated site. Phytoremediation has many environmental and economical benefits such as rehabilitating of soil, preventing the soil erosion; presenting nice scene, making the soil fertilized and creating job opportunities (recycle of the heavy metals). In the last 20 years, remediation techniques for the treatment of sites which were contaminated with heavy metals has been showing a considerable progress. But very large contaminated sites represent up today a problem due to the high costs for their remediation (Chhotu et al., 2008). Some researchers suggest that the incineration of harvested plant tissue dramatically reduces the volume of the material requiring disposal. In some cases valuable metals can be extracted from the metal-rich ash and serve as a source of revenue, thereby offsetting the expense of remediation (Cunningham and Ow, 1996). To use native plans in mining sites as a phytoremediation material is almost very helpful for not needing extra maintenance (saving money on fertelizer and pesticides, substantial for climatic factors, do not need extra water). The case

study area İzmir has potential plant diversity for bioremediation technique in post mine sites. Hence, rehabilitation of the post mine sites in Izmir city by phytoremediation technique would not be a problem.
The aim of this study is to enhance the environmental protection, landscape management and to increase the awareness of sustainable remediation methods. Also this study emphasizes the need of remediation of the mining sites.

Methodology
The method of this study is Phytoremediation as a Bioremediation technology. Bioremediation includes two different descriptions. One is technology description and the other one is design description.

In Technology Description there are 5 different techniques:

1 Phytotransformation
2 Rhizosphere Bioremediation
3 Phytostabilization
4 Phytoextraction
5 Rhizofiltration

Phytotransformation refers to the uptake of organic and nutrient contaminants from soil and groundwater and the subsequent transformation by plants. Phytotransformation depends on the direct uptake of contaminants from soil water and the accumulation of metabolites in plant tissue. For environmental application, it is important that the metabolites which accumulate in vegetation be non-toxic or at least significantly less toxic than the parent compound. Potential applications include phytotransformation of petrochemical sites and storage areas, ammunition wastes, fuel spills, chlorinated solvents, landfill leachates (including biochemical oxygen demand (BOD) and chemical oxygen demand (COD), and agricultural chemicals (pesticides and fertilizers).

Phytoremediation of the rhizosphere increases soil organic carbon, bacteria, and mycorrhizal fungi, all factors that encourage degradation of organic chemicals in soil. Rhizosphere bioremediation is also known as phytostimulation or plant-assisted bioremediation. Jordahl et al. (1997)

Phytostabilization refers to the holding of contaminated soils and sediments in place by vegetation, and to immobilizing toxic contaminants in soils. Establishment of rooted vegetation prevents windblown dust, an important pathway for human exposure at hazardous waste sites. Hydrauliccontrol is possible, in some cases, due to the large volume of water that is transpired through plants which prevents migration of leachate.towards.groundwater.or.receiving.waters. Phytostabilization is especially applicable for metal contaminants at waste sites where the best alternative is often to hold contaminants in place.

Phytoextraction refers to the use of metal-accumulating plants that translocate and concentrate metals from the soil in roots and above ground shoots or leaves. It has been used effectively by Phytotech at brownfields sites with relatively low level lead and cadmium contamination for soil remediation to below action levels (McGinty, 1996). It has also been proposed for extraction of radionuclides from sites with mixed wastes. Phytoextraction offers significant cost advantages over alternative schemes of soil excavation and treatment or disposal. An important issue in phytoextraction is whether the metals can be economically recovered from the plant tissue or whether disposal of the waste is required.

Rhizofiltration refers to the use of plant roots to sorb, concentrate, and precipitate metal contaminants from surface or groundwater. Roots of plants are capable of sorbing large quantities of lead and chromium from soil water or from water that is passed through the root zone of densely growing vegetation. The potential for treatment of radionuclide contaminants has received a great deal of attention in the press. Rhizofiltration has been employed by Phytotech® using sunflowers at

a U.S. Department of Energy (DOE) pilot project with uranium wastes at Ashtabula, Ohio, and on water from a pond near the Chernobyl nuclear plant in the Ukraine.
In Design description there are 7 steps:

1. Plant selection;
2. Treatability;
3. Planting density and pattern;
4. Irrigation, agronomic inputs, and maintenance;
5. Ground-water capture zone and transpiration rate;
6. Contaminant uptake rate and clean-up time required; and,
7. Analysis of failure modes

Design of a phytoremediation system varies according to the contaminant(s), the conditions at the site, the level of clean-up required, and the plant(s) that are used. Clearly, phytoextraction has different design requirements than phytostabilization or rhizosphere bioremediation. Nevertheless, it is possible to specify a few design considerations that are a part of most phytoremediation efforts.

Plant Selection
Plants are selected according to the needs of the application and the contaminants of concern. For phytotransformation of organics, the design requirements are that vegetation is fast growing and hardy, easy to plant and maintain, utilizes a large quantity of water by evapotranspiration (if groundwater is an issue), and transforms the contaminants of concern to non-toxic or less toxic products.

Treatability
It is necessary to utilize treatability studies prior to design in order to assure that the phytoremediation system will achieve desired results. Toxicity and transformation data are obtained in treatability studies. There is a large amount of variation in toxicity and transformation rates that can be expected from one plant species to another and even from one variety or cultivar to another.

Planting density and pattern
Planting density depends on the application. Louis Licht, Ecolotree, has pioneered the use of hybrid poplar trees as riparian zone buffer strips, landfill caps, and at hazardous waste sites. For hybrid poplar trees, 1000 to 2000 trees per acre are typically planted with a conventional tree planter at 12 to 18 inches depth or in trenched rows one to six feet deep.

Irrigation, agronomic inputs and maintenance
For terrestrial phytoremediation applications, it is often desirable to include irrigation costs in the design, on the order of 10 to 20 inches of water per year. Irrigation of the plants ensures a vigorous start to the system even in a drought. On the other hand, hydrologic modeling may be required to estimate the rate of percolation to groundwater under irrigation conditions. Over time, irrigation should be withdrawn from the site, provided the area receives sufficient rainfall to sustain the plants.

Ground water capture and transpiration
One must understand where the water is moving at a site in order to estimate contaminant fate and transport. For applications involving ground-water remediation, a simple capture zone calculation (Domenico and Schwartz, 1997) can be used to estimate whether the phytoremediation "pump" can be effective at entraining the plume of contaminants. Trees can be grouped for consideration as average withdrawal points. The goal of such a phytoremediation effort is to create a water table depression where contaminants will flow to the vegetation for uptake and treatment. It is important to realize that organic contaminants are not taken-up at the same concentration as in the soil or groundwater, rather there is a transpiration stream concentration factor (a fractional efficiency of

uptake) that accounts for the partial uptake of contaminant (due to membrane barriers at the root surface).

Contaminant Uptake Rate And Clean-Up Time
From equation (1) above, it is possible to estimate the uptake rate of the contaminant(s). First order kinetics can be assumed as an approximation for the time duration needed to achieve remediation goals. The uptake rate should be divided by the mass of contaminant remaining in the soil: k = U/Mo (2) ,where k = first order rate constant for uptake, yr-1 , U = contaminant uptake rate, kg/yr ,Mo = mass of contaminant initially, kg Then, an estimate for mass remaining at any time is expressed by equation (3) below.
M = Mo e-kt (3)
Where M = mass remaining, kg
t = time, yr
Solving for the time required to achieve clean-up of a known action level:
t = -(ln M/Mo)/k (4)
Where t = time required for clean-up to action level, yr
M = mass allowed at action level, kg
Mo = initial mass of contaminant, kg

Analysis of Failure Modes
Phytoremediation systems are like any other treatment scheme; one cannot simply walk away from them and expect success. There are events that can cause failure that should be realistically assessed at the outset. These include killing frosts, wind storms, animals (voles, deer, beaver), disease or infestation (fungus, insects), and latent toxicity. A contingency fund should be provided for periodic replanting of a certain percentage of the site in order to ensure a viable vegetation system.
In detail there is a further method to quantify the metals absorbed by plants. The machine for this procedure is called Atomic Absorption Spectroscopy. First of all in this method plant samples were thoroughly washed with running tap water and rinsed with deionized water to remove any soil particles attached to the plant surfaces, then oven dried (70ºC) to constant weight. The dried tissues were weighed and ground into powder for metal concentration analysis. Metal contents (70 different metal species) of the plant samples were extracted by acid digestion followed by measurement of total concentrations of all elements of interest using atomic absorption spectrophotometer.

Impact
The impact of this study has more than one area:

*Social impacts***:** This study will help to determine the heavy metals effects on natural life. The local managements will be aware about the effects of mining works in Southern Alps. Moreover this study will emphasize the value of the area and this will make people know more about the Trento and surroundings. The cultural landscape will be protected by this study and this will may increase the touristic activities and the advertisement of this area.

Environmental impacts: This study will preserve the endemic species in the study area because bioremediation technique will help them to survive. This is a low tech method to present to the disciplines which are working on environmental issues.

Economic impacts: To recycle the metals will help to resolve charges for remediation payments. And the payments of maintenance and watering of plants will be reduced by using native plants.

Results
In conclusion in every kind of discipline which are relevant to environment should take care of environmental pollution and find solutions for the future generation. Mining sites has big scale in all over the world and they create pressure on natural systems by heavy metal pollution.

Phytoremediation is one of the most efficient, beneficial, innovative and simply implementable solution for landscape protection in contaminated mine sites.
We cannot prevent the governments to stop building the mining sites because heavy metals are essential material of industry, architecture more areas. But what we can do is to reduce the effects of toxic heavy metals on environment. Toxic heavy metals in open mining sites can be transformed by the wind to air. Furthermore heavy metals can be reaching to the underground water resources. All the effects of toxic heavy metals create a threat on nature.
Landscape planning can help to create balance between nature and the effects of contaminated mine sites and bioremediation is one of the best planning proposal for ecology and environment. Phytoremediation as a bioremediation method should be implemented in the contaminated mine sites of Izmir city.

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