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Enhanced phytoremediation of heavy metal polluted soil with native crops of Punjab, Pakistan

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Key words: Chelates, Fungi, Heavy metals, Maize, Mustard.

Abstract

In the present study, chemically and biologically enhanced phytoremediation potentials of maize (*Zea mays*) and mustard (*Brassica campestris*) were evaluated by cropping them on two different metals contaminated soils of Lahore and Gujranwala for 75 days. Soils were treated with varying amounts of DTPA (Diethylene Triamine Penta Acetic acid) and three different fungal species to facilitate metal uptake by plants. In pot experiment under green house, addition of fungi and DTPA chelate significantly increased the Cu, Pb, Cr and Cd concentrations in roots and shoots. Maximum maize shoot biomass was obtained in the presence of *Aspergillus fumigatus* and maximum root biomass was produced in the presence of *Aspergillus niger*. In case of mustard crop both shoot and root biomass was maximum produced in the presence of *Aspergillus fumigatus*. It was found that maximum Cd and Cu were solubilized in *Aspergillus flavus* inoculated soils while Pb and Cr were solubilized maximum in *Aspergillus fumigatus* inoculated soils in case of maize experiment. In case of mustard maximum Cd and Cu were solubilized in *Aspergillus niger* inoculated soils while Pb and Cr were solubilized maximum in *Aspergillus flavus* inoculated soils. Increased metals uptake, bioconcentration factor, and phytoextraction rate and phytoextraction efficiency were noticed over the control in both fungal and DTPA amended soils. Based on obtained data a phytoremdiation model was developed.

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Introduction

A variety of treatment technologies has been developed for the remediation of metals contaminated soils and phytoextraction is an economically feasible technique. It is publically more acceptable and also has potential to clean the environment (Chen and Cutright, 2002). Phytoremediation is a plant based remediation strategy which uses the plants for the environmental remediation (Rauf *et al.,* 2009). Many chemical and biological treatments, such as inoculums of fungi and bacteria, EDTA, DTPA, NTA and other organic compound have been reported to used in pot and field experiments to facilitate the heavy metals extraction and to acquire the higher efficiency of phytoextraction/phytoremediation (Blaylock *et al.*, 1997; Huang *et al.*, 1997; Kayser *etal.*, 2000; Ke *et al.*, 2006 and Wu *et al.*, 2006). It is known that microbial populations affect the solubilization of heavy metals and their availability to plants, through acidification, releasing chelator and reduction-oxidation changes (Peer *et. al.*, 2006). It is reported that presence of microbes in the rhizosphere increases the levels of Zn, Cu, Pb, Ni and Cd in plants. Heavy metals tolerance and production of biomass could be enhanced by improved interaction among the plants and rhizosphere microbes. So it is considered an important phytoremediation technology factor (Whiting *et. al.*, 2003).

Fungi play a very important role in solubilization and fixation of heavy metal ions and change the availability of these ions for the plants (Birch and Bachofen 1990). Different soil and plant factors affect the phytoremediation process and these also include the soil fungi. There is a need of information about symbiotic relationships between soil microbes (bacteria and fungi) and roots of plants. Heavy metals are also in compound forms in soil which also affects the metals behavior in their solubilization and uptake processes (Boruvka and Drabek, 2004). Different types of components present in the fungal cell walls like carboxyl, hydroxyl, amino and other functional groups. Through these functional groups fungi can bind with toxic heavy metals such as Pb, Cu, Cd and Ni etc. (Kapoor and Viraraghavan, 1995). The phytoremediation effectiveness limited often because of low solubility of metals and theirsorption on surfaces of soil particals, however metals solubilization could be increased by adding complexing/chelating agents with the time (Pivetz, 2001). In the literature several chelating agents has been reported which enhance the rate of phytoextraction. However EDTA and DTPA has been investigated widely and they have high chelating ability towards the most of the metals, like Cd, Cu, Cr and Pb, which ultimately leads to increased translocation of metals from soil to plant (Barlow *et al.*, 2000; Wong *et al.*, 2004 and Begonia *et al.*, 2005). A lot of researches on phytoextraction are based on green house experiments few tested the hyperaccomulators plants in the field and actually determined their heavy metals accumulation potential (Hammer and Keller, 2003; McGrath *et al.*, 2006 and Zhuang *et al.*, 2007). Pakistan is a developing country and its population is more as compared to resources. Most of the area is used for agriculture and urbanization purpose. Due to industrialization the increase level of contamination is making land useless for better yield production. Contamination of lands with toxic heavy metals is a widespread environmental issue. The present study was planned by keeping in view the heavy metals exposure directly to the water and agriculture soils. The proposed study was done in laboratory and under green house conditions with the objectives to evaluate the efficacy of a synthetic chelator and isolated fungi in the uptake and translocation of heavy metals as well as on the total biomass of plants.

Materials and methods

Soil sampling

Bulk surface samples of two soils used for arable agriculture were collected from Gujranwala and Lahore. These soils were polluted with heavy metals. Soil from Gujranwala(fine–loamy, mixed, hyperthermic Udic Haplustalf) were collected from peri-urban areas of Gujranwala (N 32°–06.262; E

74°–10.236) and Lahore (N 24°–57.212; E 69°–17.329).

Soil preparation

Contaminated soil samples were dried in open air, grinded and sieved through a stainless steel sieve of 2-mm. About 200 g portion of soil was again sieved from the <2-mm sieved and reground to obtain a <200-µm fraction of soil for chemical analysis. The Gujranwala surface soil was medium in texture with higher soil percentage organic matter content. Both soils are calcareous in nature. Total heavy metal concentrations in the soils were determined by Flame Atomic Absorption Spectroscopy with graphite furnace of PerkinElmer AAnalyser 800 model.

Green house pot experiment

Three kgof each of the contaminated Gujranwala and Lahore soils werefilled in polyethylene lined plastic pots, after that six seedsof each crop were sownin each pot. The soils were fertilized with500mg N kg^{-1} of dry soil as urea (in three splits),50 mg K kg^{-1} as KH_2PO_4. $2H_2O$and 70 mg P kg^{-1} as a basal dose. The pots were brought to near60 % of the water holding capacity by the addition of distilledwater and after that were maintained around this moisture levelthroughout the whole experiment. Chelating agent (DTPA) was applied at the rate of 0,1.25, 2.5, and 5.0 mM kg^{-1} of soil in two equal splits at 25th and 45th day after sowing in triplicate. Fifteen ml spores suspension of three fungal species *Aspergillus niger* (Specie A), *Aspergillus flavus* (Specie B) and *Aspergillus fumigatuse* (Species C) were applied in triplicates. Applied fungal cultures were two weeks old. Each fungal inoculated pot was also amended with 100 mL potato broth of 10^7strength at the time of sowing.

Plantswere harvested after 75 day of sowing, and shoots and roots were washed with simple and de-ionized water. Fresh and dry weights (after drying at40 °C) of shoots and roots were recorded periodically and analyzed for Cr, Cu, Cd and Pb. After harvesting, the soil was analyzed for DTPA extractable Cd, Pb, Cu and Cr concentrations.

Analysis of data

In order to compare the phytoextraction efficiency of all the plant species after the addition of different chelating material concentrations to soils, the Bioconcentration Factor (BCF), Phytoextraction Rate (PR) and extraction factor (EF) were calculated. A phytoremediation model was developed based on generated data and time period for the reclamation of metals contaminated soils was predicted using obtained information.

Results and discussion

Maize shoot biomass

Inoculation with *Aspergillus niger* showed 13.8% increase in maize shoot biomass, *Aspergillus flavus* showed 11.5% and *Aspergillus fumigatus* showed 18.6% increased in fresh shoot biomass as compared to control (Fig. 1).

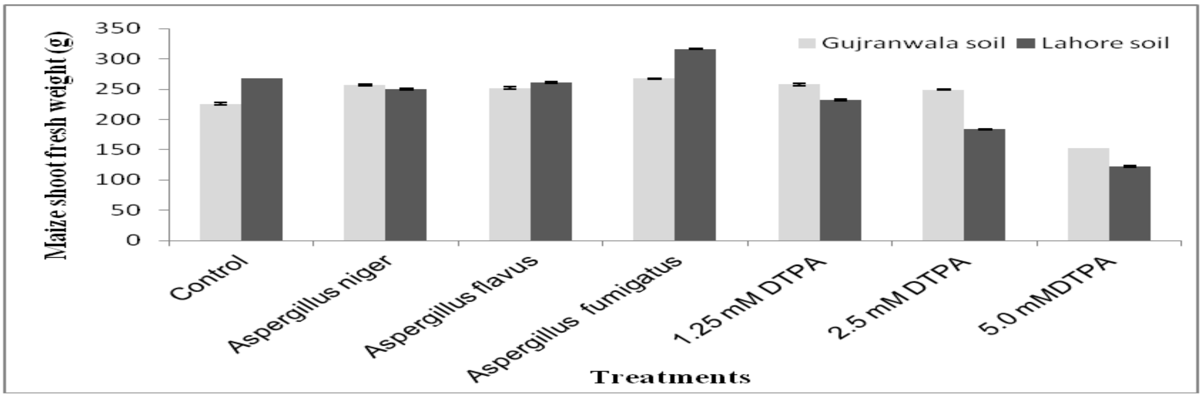

Fig. 1. Maize shoot fresh biomass production in Gujranwala and Lahore soils in the presence of fungi and DTPA concentrations.

It was also shown in the graphical presentation that experiment conducted under green house using Lahore contaminated soil 6.7% and 4.1 % decrease in maize shoot fresh weight was observed when *Aspergillus niger* and *Aspergillus flavus* were applied respectively but 18% increased was noticed in case of *Aspergillus fumigatus* innoculation.Overall *Aspergillus fumigatus* presented good results in plant grown in both Lahore and Gujranwala soils. In case of chemical treatment, DTPA amendment showed less shoot biomass growth as compared to the control in Lahore and Gujranwala soils only DTPA dose 1.25 mM kg^{-1} showed 14.2% more growth as compared to control in Gujranwala soil. Exactly same trend was notices in dry biomasses of maize shoot (Fig. 2).

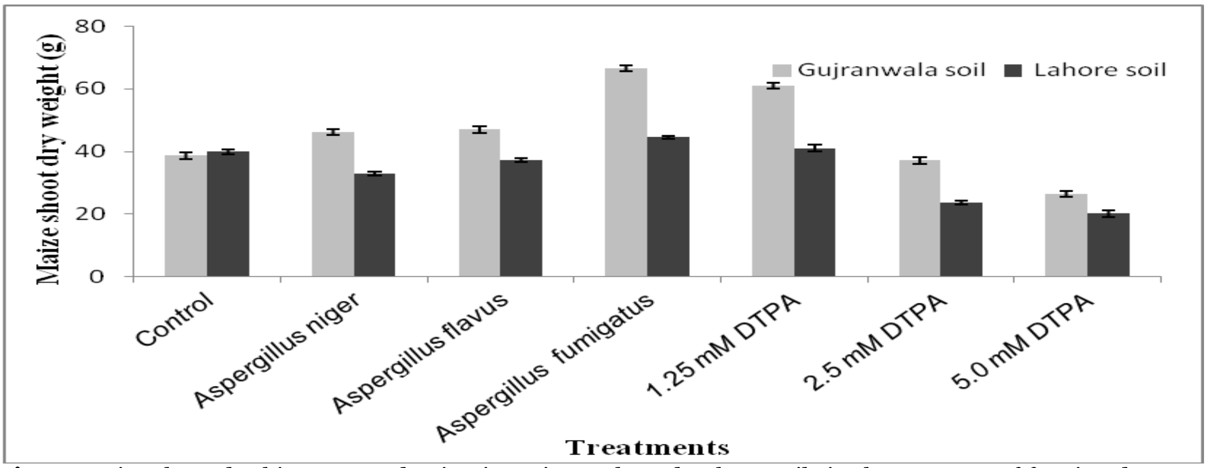

Fig. 2. Maize shoot dry biomass production in Gujranwala and Lahore soils in the presence of fungi and DTPA concentrations.

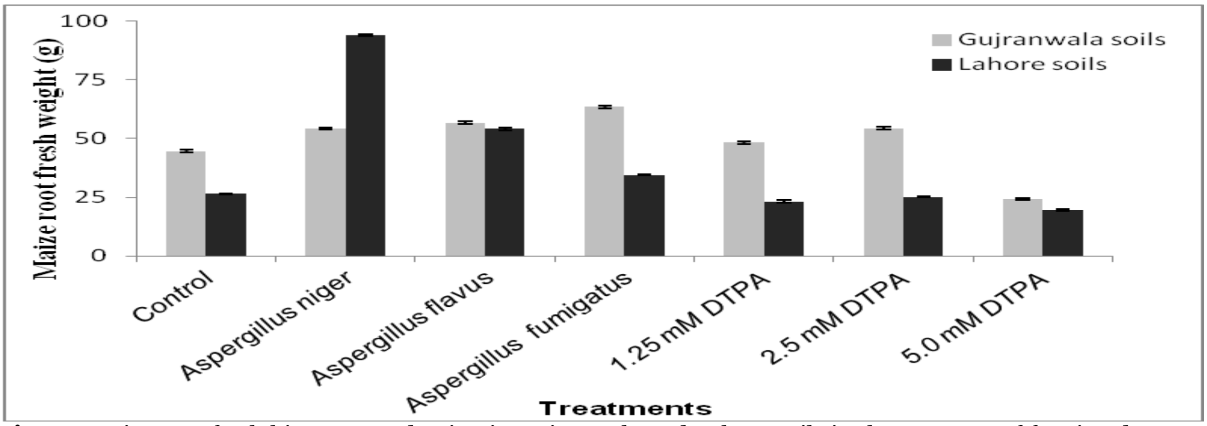

Fig. 3. Maize root fresh biomass production in Gujranwala and Lahore soils in the presence of fungi and DTPA concentrations.

Maize root biomass

With the addition of spore suspension of fungal strains growth was affected significantly and plants inoculated with *Aspergillus niger* showed 22.7% increase in root biomass as compared to control in Gujranwala soil and similarly plants inoculated with *Aspergillus flavus* and *Aspergillus fumigatus* also showed 27.2% and 43.1% increment in biomass respectively. With DTPA amendment root biomass growth increased with increasing dose from 1.25 mM Kg^{-1} to 2.5 mM Kg^{-1} as compared to control whereas root growth was reduced at 5.0 mM Kg^{-1} DTPA treatment in Gujranwala soils. It was observed from the results obtained from the experiment conducted in Lahore soil, *Aspergillus niger* enhanced the root growth two times as compared to the control.

Similarly *Aspergillus flavus* and *Aspergillus fumigatus* also showed significant increase in root biomass. On the other hand addition of chemical amendment stressed the root growth as compared to the control plants.

Mustard shoot biomass

It is clear from results that biological agents (fungi) have enhanced the shoot biomass production and maximum shoot biomass was observed in those pots which were inoculated with *Aspergillus fumigatus*.

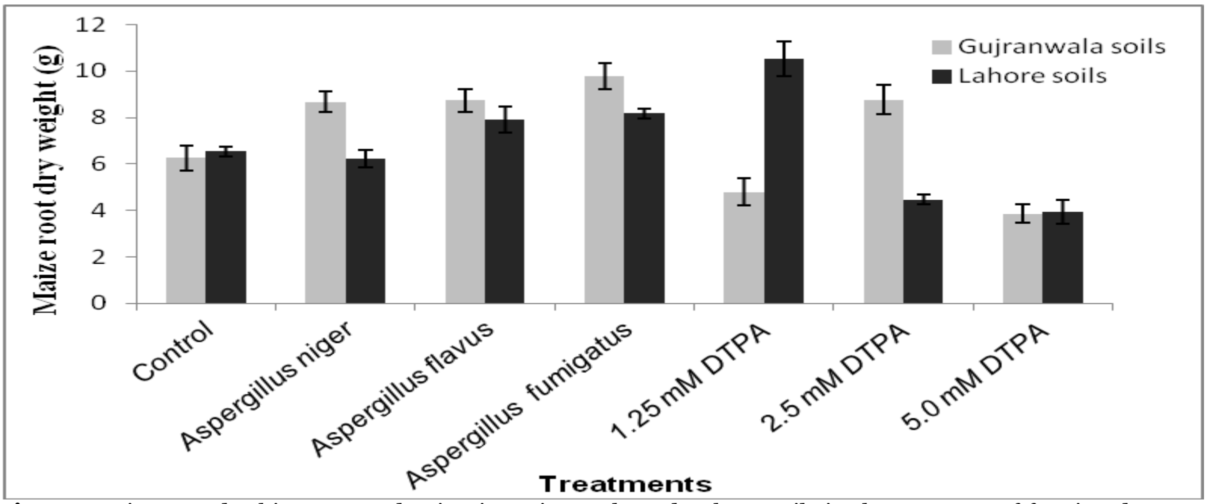

Fig. 4. Maize root dry biomass production in Gujranwala and Lahore soils in the presence of fungi and DTPA concentrations.

It is also noticed that Gujranwala soils has supported more mustard plants growth as compared to Lahore soils. *Aspergillus fumigatus* has increased the shoot growth four times higher as compared to control pots. Maximum fresh shoot biomass in Lahore was noticed 206.7 grams. In the DTPA amended pots maximum shoot biomass was observed at 1.25 mM Kg^{-1} of soil application and it reduced with increasing DTPA doses. Except the pot which was inoculated with *Aspergillus fumigatus* Lahore soil showed better biomass production as compared to Gujranwala. Dry biomass estimation showed that Lahore soil supporting more mustard plant growth. Maximum dry biomass was also produced in the presence of *Aspergillus fumigatus* (Fig. 5 and 6).

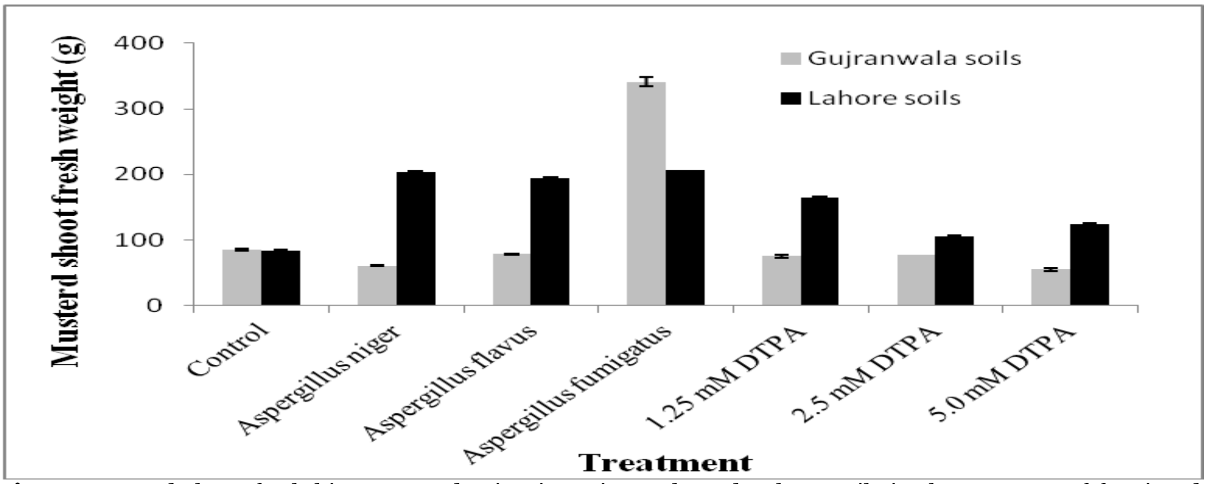

Fig. 5. Mustard shoot fresh biomass production in Gujranwala and Lahore soils in the presence of fungi and DTPA concentrations.

Mustard root biomass

Pots of Lahore soil showed more root growth as compared to Gujranwala soil. Maximum root biomass was produced in the presence of *Aspergillus fumigatus* in Lahore soil and *Aspergillus niger* in Gujranwala soil. In case of DTPA maximum root biomass production was observed at 2.5 mM Kg^{-1} of Gujranwala and Lahore soil. Similar results were predicted in case of root dry biomass. Maximum biomass produced was 29.67 grams in Lahore soil pots and 7.94 in Gujranwala soil. With the addition of spore suspension of fungal strains growth was affected significantly. When DTPA was applied root biomass production was increased as compared to the control in Lahore soil whereas, in Gujranwala soil growth was decreased. Overall, in this experiment root biomass growth was improved 60% as compared to the control.

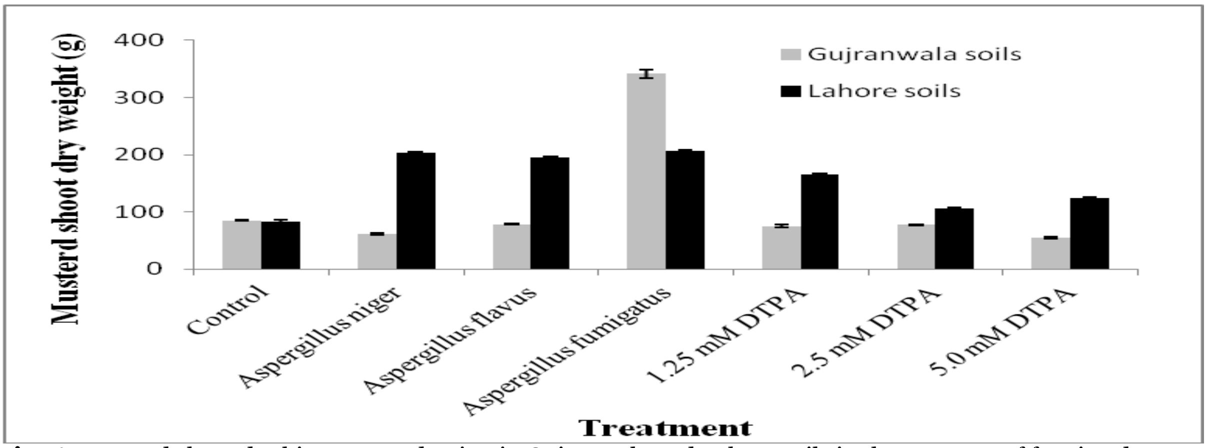

Fig. 6. Mustard shoot dry biomass production in Gujranwala and Lahore soils in the presence of fungi and DTPA concentrations.

In the present study *Aspergillus niger, Aspergillus fumigatus* and *Aspergillus flavus* showed potential for enhancing maize and mustard crops growth and phytoremediation of heavy metals (Cd, Cr, Pb and Cu) contaminated soils. Addition of fungal inoculums to the contaminated soil of Gujranwala and Lahore increased shoot biomass of maize and mustard crops more effectively as compared to the chemical amendment (DTPA).

Among the three fungal strains *Aspergillus fumigatus* has highest capacity to enhance the shoot growth. Based on above findings, it was concluded that, *Aspergillus fumigatus* could be used for phytoremediation of experiments in the heavy metals contaminated calcareous soils.

Many reports are available in the literature on enhancing the shoot, root and grain yield of crops treated with microbial inoculums and chemicals. Wu and Cheung (2005) reported that microbial inoculums not just expanded the nutrients uptake by the plants (for example N, P and K) but in addition to these they enhanced the soil properties, for example, organic matter substance and aggregate nitrogen in the soil. Maximum root biomass of maize and mustard crops grown under green house in the pot experiment was produced in the presence of *Aspergillus fumigatus* and DTPA applied at the rate of 1.25 mM kg^{-1} of soils to Gujranwala and Lahore contaminated soils.

Results of present study demonstrated that addition of DTPA and fungal inoculums enhanced root growth which can increase phytoextraction potential of heavy metals from contaminated soils with high efficiency. Results demonstrated that the addition of fungal inoculums and DTPA to the two soils induced Cu, Pb, Cr and Cd solubilization in the soil which ultimately stimulated phytoaccumulation and facilitated phytoextraction.

The higher solubilizable heavy metals concentrations in the Gujranwala soil than the Lahore soils was most probably because of the high initial total and extractable metals concentrations of these soils. Thus, there was also a possibility of metals to leaching down the soil profile and this also may pose a high environmental risk to groundwater quality. A few investigations has been made to reduce the leaching risk associated with the use of synthetic chelators.

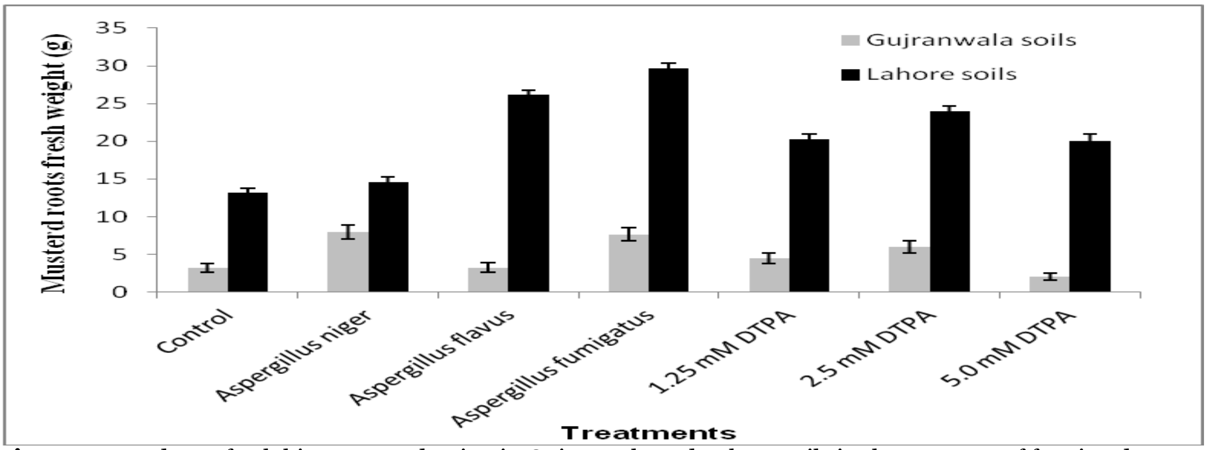

Fig. 7. Mustard root fresh biomass production in Gujranwala and Lahore soils in the presence of fungi and DTPA concentrations.

Residual metal contents in maize growing soil

It was observed that soil Cd, Cr, Pb and Cu concentrations significantly higher than those of the control in Lahore experimental soils but in Gujranwala experimental soil above metals concentrations were not significantly higher than control (Fig. 9 and 10).

This increase in extractable metals concentrations with increasing rates of DTPA and with different fungal strains suggests that the metals remained solubilized in the soil solutions even at the termination of the experiment. In case of Cd and Cu *Aspergillus flavus* showed more solubilization in post harvest soil of Gujranwala and *Aspergillus fumigates* showed maximum availability of Cr and Pb in Lahore and Gujranwala soils respectively.

Residual metal contents in mustard growing soil

It was noticed that extractable Pb content was higher than control in Gujranwala soils where as lower than control in Lahore experimental soils.

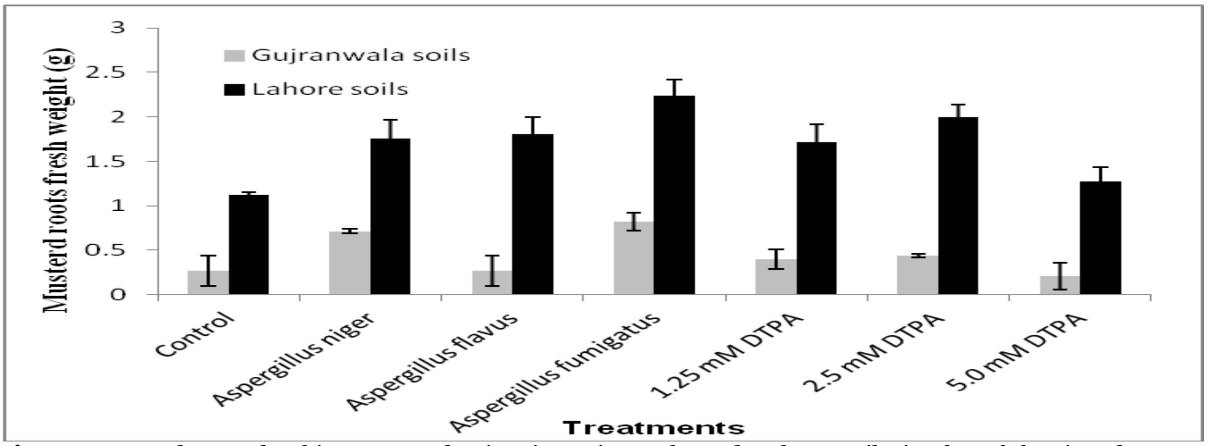

Fig. 8. Mustard root dry biomass production in Gujranwala and Lahore soils in the of fungi and DTPA concentrations.

The concentration of Cu was found approximately lower than control experiment in both Gujranwala and Lahore soils except in one treatment when *Aspergillus niger* was applied. In case of Cd availability, it was observed that Cd concentration was found higher in those soils of Gujranwal wher *Aspergillus niger, Aspergillus flavus* and 5.0 mM of DTPA Kg^{-1} of soil were applied. In contrast this, in Lahore soils Cd was found less then control in all the treated samples.

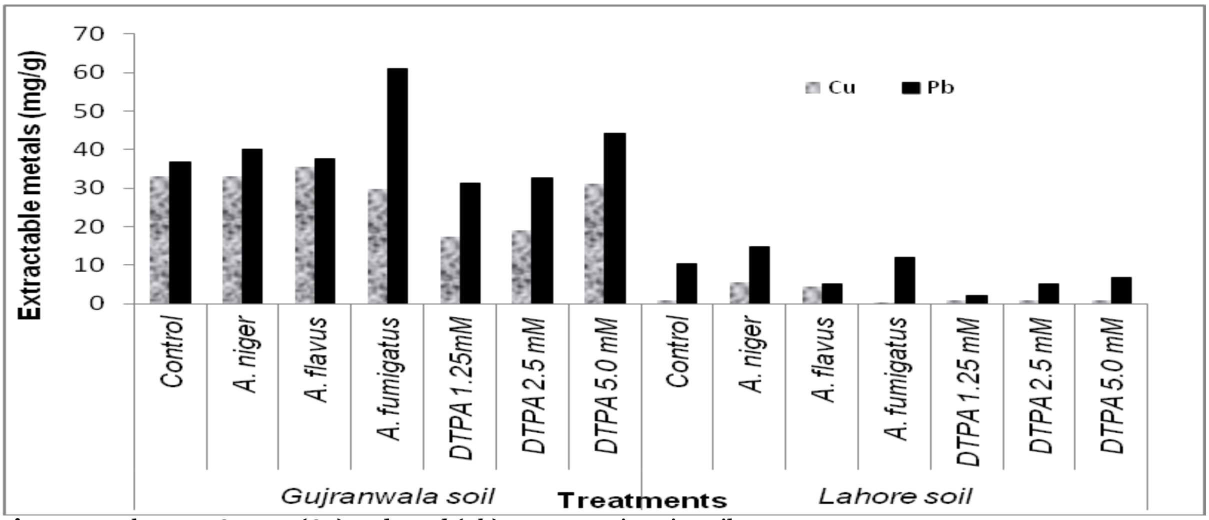

Fig. 9. Post harvest Copper (Cu) and Lead (Pb) concentrations in soils.

In case of Cr solubility Cr concentration in residual soil was noticed lower than control in Gujranwala soils but higher in Lahore soils. This may be due to different physiochemical properties of Lahore and Gujranwala soils (Fig. 11 and 12).

Metal analysis in maize shoots tissues

The highest increase in Cd, Pb, Cu and Cr concentrations in maize shoots were 2.01, 32.95, 28.57 and 3.02 mg g^{-1} respectivly in the Gujranwala soil and 5.57, 21.81, 2.08 and 7.98 mg g^{-1} respectivly in the Lahore soil when treated with *Aspergillus niger*, *Aspergillus flavus* and *Aspergillus fumigatus* and 1.25, 2.5 and 5.0 mM DTPA kg^{-1}. In Gujranwala soils *Aspergillus flavus* was proved more effective in removal of Cd, Pb and Cu while *Aspergillus fumigatus* gave good results in Cr uptake by maize plants. Similarly in Lahore soil Cd, Cu and Cr was significantly removed by maize plants when *Aspergillus flavus* was applied and maximum Pb was removed by maize crop when soil was inoculated with *Aspergillus fumigatus*. In case of chemical amendment DTPA dose 5.0 mM kg^{-1} was more suitable dose in metals solubilization which ultimately enhanced the metals uptake by maize plant shoots. It was noticed that after the completion of phytoremediation experiments metal concentrations were higher in Gujranwala soil as compared to Lahore soil. Whereas plant uptake were more grown in Lahore soils as compared to Gujranwala soil.

Metal analysis in maize roots tissues

Pb and Cr concentrations in roots of maize crop increased linearly with increasing rates of DTPA. Cd and Cu were more available to plants at DTPA dose 2.5 mg kg^{-1} of soil in case of chemical amendment. The maximum Cd, Pb and Cr concentrations were noticed 3.38, 67.75 and 37.67 mg $g^{-1}$of root dry matter in Gujranwala soil in the presence of *Aspergillus fumigatus* inoculum. Whereas, maximum Pb concentration in maize root dry matter was noticed in the presence of *Aspergillus flavus*. In maize roots grown in Lahore soil maximum Cd, Cu, Cr and Pb uptake was enhanced in the presence of *Aspergillus fumigates*.

Metal analysis in mustard shoots tissues

Maximum Cu uptake was noticed when *Aspergillus* *niger* and 2.5 mM of DTPA kg^{-1} of soil was applied in Gujranwala soils. When experiment conducted by using Lahore soils maximum Cu was found in shoots in the presence of *Aspergillus flavus* and 1.25 mM DTPA kg^{-1} of soil.

Maximum Cr concentration was observed in the presence of *Aspergillus fumigatus* in Gujranwala soils and in the presence of *Aspergillus flavus* in Lahore contaminated soil. Soil Cd availability to the mustard crop shoots was observed highest (1.85 mg kg^{-1}) in the presence of *Aspergillus niger* and *Aspergillus fumigatus* in Gujranwala soil. Chelate dose 2.5 mM proved best for Cd (2.07 g kg^{-1}) uptake from Gujranwala soil.

In Lahore soil maximum Cr was found in shoots of mustard when soil was inoculated with *Aspergillus flavus*. In case of Pb maximum shoot Pb concentration was found when Gujranwala soil was treated 1.25 and 25 mM DTPA kg^{-1} of soil and Lahore soil was treated with 2.5 mM DTPA kg^{-1} of soil.

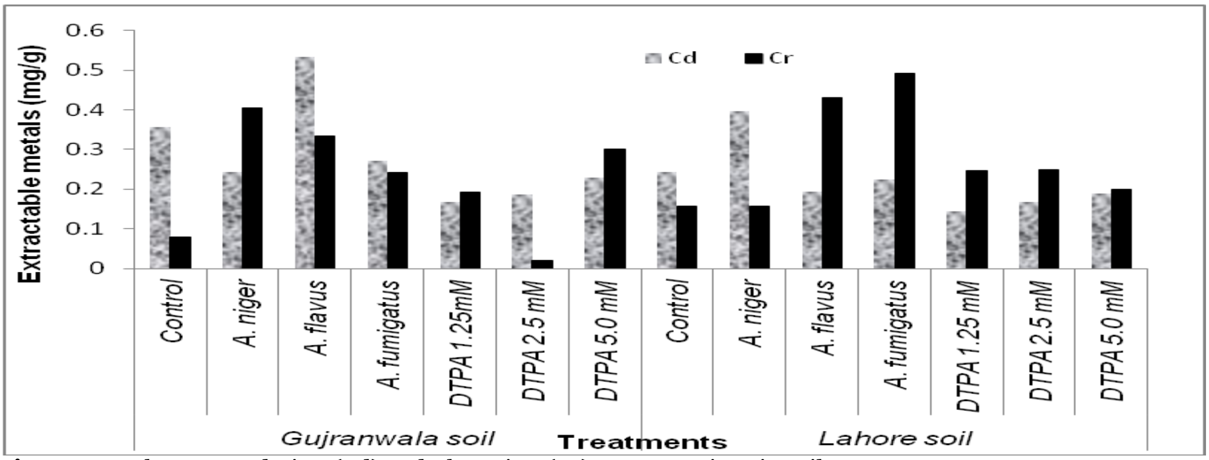

Fig. 10. Post harvest Cadmium(Cd) and Chromium(Cr) concentrations in soils.

Metal analysis in mustard roots tissues

Maximum Cu was noticed in roots of mustard plants when grown in the presence of *Aspergillus niger* in Gujranwala soil and in the presence of *Aspergillus fumigatus* in Lahore soil.

Chelat concentration 2.5mM DTPA Kg^{-1} of soil was proved effective in making Cu available to the mustard roots in Gujranwala soil and 5.0 mM DTPA Kg^{-1} of soil in Lahore soil.

Maximum Cu, Cr, Cd and Pb were found in mustard roots in the presence of Aspergillus *fumigatus* inoculums in Lahore experimental soils. Whereas, in case of Gujranwala soils *Aspergillus fumigatus* was proved best for Cr and Pb accumulation by mustard roots. Similarly, DTPA dose 5.0 mM DTPA Kg^{-1} of soil was effective dose in making Cu, Cr, Cd and Pb available for the mustard uptake in Lahore soil.

Bioconcentration factor (BCF)

Overall mustard showed greater BCF value than maize for Cd, Cu and Cr however; maize depicted higher value of BCF for Pb. Maize and mustard showed maximum value for Pb 22.63 and 12.38 respectively and also in Lahore soils.

Maximum BCF values for Cu were observed 1.02 for maize and 484 for mustard. Mustard has high potential to accumulate Cu from soil. Similarly, Cr BCF values 61.50 and 107 were calculated for maize and mustard.

Phytoextraction rate (PR)

Maximum Cd phytoextraction rate of both crops maize and mustard were observed 50.29 and 72.80 in Lahore soil when soil was treated with *Aspergillus fumigatus*. Phytorextraction rate of Pb was noticed highest for maize and mustard (5.03 and 5.94) in the presence of *Aspergillus flavus* in Lahore soil.For Cu, it was shown that 220.17 was the highest phytoextraction rate for mustard when soil was amended with *Aspergillus flavus* and 135 for maize when *Aspergillus fumigatus* was applied. Only Cr phytoextraction rate for maize was noticed highest in Gujranwala soil but for mustard, maximum value was observed in Lahore soil.In case of DTPA treatments (1.25, 2.5 and 5.0 mM Kg^{-1} of soil), it was observed that with increasing DTPA dose metals solubility also increased and which caused in increased in phytoextraction rate of plants. In Gujranwala soils, most efficient dose was noticed 2.5 mM Kg^{-1} of soil and in Lahore soils 5.0 mM Kg^{-1} of soil was noticed more significant level of DTPA application.

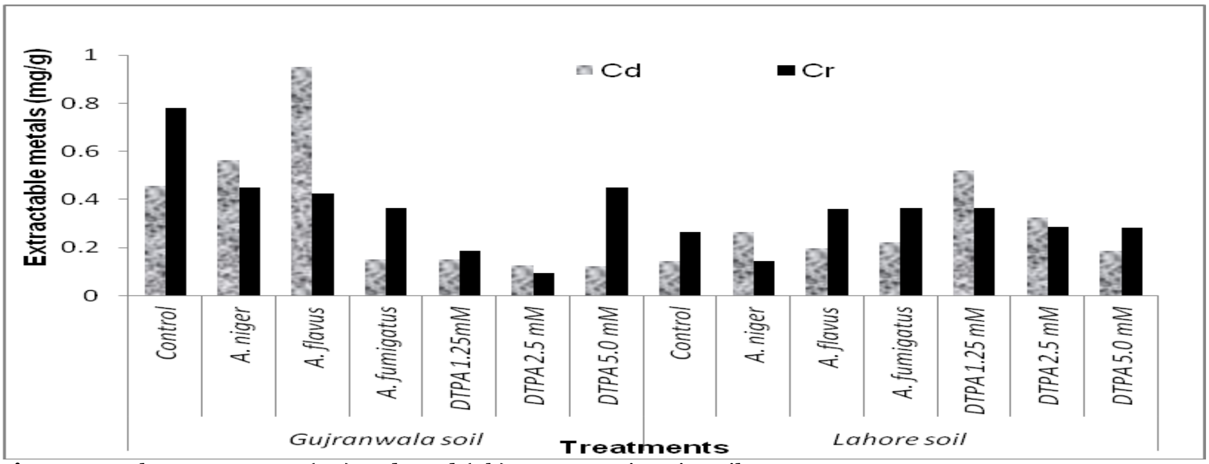

Fig. 11. Post harvest Copper (Cu) and Lead (Pb) concentrations in soils.

Extraction factor

The maximum Pb extraction factor of maize was calculated 63.2% in Gujranwala soil in the presence of *Aspergillus fumigatus* and 58% for mustard when DTPA dose was 5.0 mM Kg^{-1} of soil. In case of Lahore soils 40% extraction factor was shown by maize and 30% by mustard crop. Maximum Cu extraction factor (20.86% for maize and 34.3% for mustard) was observed in Gujranwala and Lahore soils respectively. The highest Cd extraction factor for maize crop was 16% in *Aspergillus flavus* amended soil and 28.56% in DTPA (2.5 mM Kg^{-1} of soil) amended soils of Gujranwala. While in Lahore soils 27.3 and 35.9% extraction factor were observed for maize and mustard both in the presence of *Aspergillus niger* application.

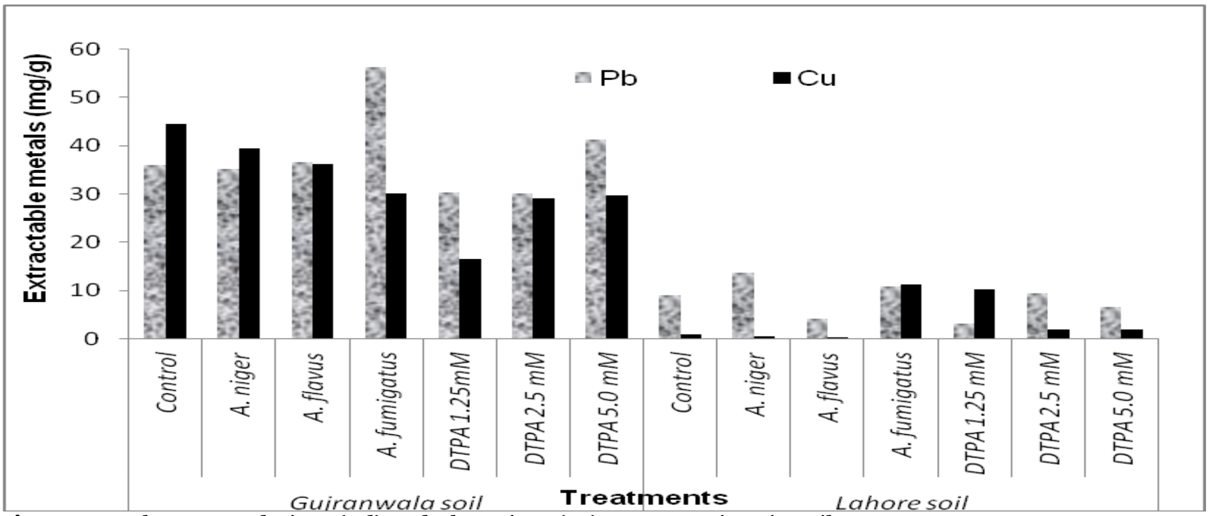

Fig. 12. Post harvest Cadmium (Cd) and Chromium (Cr) concentrations in soils.

Phyto-remediation model

Based on metals concentrations in the soil system and the threshold values targeted for the remediation, phytoremediation could be done repeatedly to bring the metals contamination level under the safe limits. The potential of the plant to extract metals from soil is a function of metal uptake by plants and plants biomass production. An important aspect of the present day is the ability to adequately model the behavior of system and before committing to a large scale investment. For developing a good model, there is a need of basic theoretical understanding of the system, experimental observation and measurement of the system. Model Assumptions were as follows:

1. The amount of metals in soil system during the experiment remains same.
2. The rate of effective uptake of metals by the plants roots becomes saturated as the amount of soil amendments increase.
3. The harvest cycle is a fixed period of months.
4. Each plant has the same quantity of biomass.
5. The death rate of plants during a harvest period is constant.
6. The amount of soil in the environment remains constant (the environment does not change)
7. The amount of plants harvested at the end of each harvest cycle remains the same (not dependent on number of harvest cycles)
8. A number of other factors working together during field applications could be challenged all those were assumed constant.
9. This model would be consider for upper soil layer 0-20 cm
10. Bulk density of soil was considered 1.4 Mg m^{-3}, giving a total soil mass of 2600 Mg ha^{-1}.

Based on above assumptions and calculated data following model can b proposed:

$$t = \frac{\text{Mi} - \text{Mf}}{\text{Mp} \times \text{Wp}}$$

t = phytoremediation time (years), Mi = metals concentration in soil (g/ha), Mf = acceptable metals concentration (g/ha), Mp = metals concentration in plants (g/ton), Wp = weight of plant (ton/ha/year).

The findings of present study also demonstrated that shoot growth of maize and mustard crops expanded more efficiently on the expansion of fungal strain, as compared to the chemical treatments with DTPA (1.25, 2.5 and 5.0mM kg^{-1} of soil) and to the control soil. The increase in shoot height could be because of increased activation of phosphorus made soluble by phosphorus solubilizing fungal isolates from the reserves of soil and rock phosphate (Singh *et. al.*, 2011). Metals uptake by plant tissues, their mobilization and storage in plant biomass is refereed bioconcentration factor (BCF) in phytoremediation technology (McGrath and Zhao, 2003). In the present study mustard crop showed higher BCF for Cd, Cu and Cr than maize. Whereas, maize crop showed higher BCF for Pb than mustard crop. This is in line with the view that the application of metal mobilizing agents to soils is a way of chemically enhancing root uptake and translocation of metals contaminants from soils to plants, thereby improving phytoextraction (Liphadzi *et al.*, 2003). Metal accumulation capability and agronomic practices to enhanced crops growth collectively termed as phytoextraction rate (PR) (Chaney *et al.,* 2000). Present study revealed that PR of Cd and Cu is more for mustard and PR of Pb and Cr was higher for maize crop. Ciura *et al.* (2005) showed that the crop's efficiency in cleaning soil polluted with metals was dependent on its biomass production and the metal distribution among its crop tissues. Potential risks due to unwanted leaching of metal complexes to groundwater during the fungal and chelant-enhanced phytoextraction process (Romkens *et al.*, 2002) can be assessed by comparing the RF with the extraction factor (EF), which represents the relative amount of metal that can be extracted with addition of varying amounts of EDTA. Chelate and fungal inoculums. In the current study conducted on maize and mustard crops for the phytoremediation of Cu, Cr, Cd and Pb values of EF for Pb, Cd and Cu were noticed higher for mustard crop and for Cr by maize crop. USEPA (2000) tested different plant species that have the potential to take up Pb and found that mustard has good ability to transport Pb from roots to shoots.

A study on the effects of different concentrations of Pb on the uptake and accumulation of this metal by the roots, hypocotyls, and shoots of mustard showed that it has considerable ability to remove Pb from solutions and accumulate it (Liu *et al.*, 2000). The uptake ability of maize and mustard to remove metals is highly considerable due to of its high biomass and genetic accumulation capacity.

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