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Pressmud Subdue Phytoremediation Indices in Lead-Contaminated Soils: A Human Health Perspective

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Abstract
Direct discharge of waste into water bodies and mining are two major sources of lead contamination in ecosystems. Water scarcity promoted the usage of industrial effluent-contaminated waters for crop production, mainly in peri-urban areas. These wastewaters may contain heavy metals and pollute crop ecosystems. These metals can reach the living cell via contaminated raw foodstuffs that grow under these conditions and cause various ill effects in metabolic activities. In this study, graded levels of pressmud (0, 2.5, 5, 10 g/kg) were applied on lead imposed soil with different contamination levels (0, 100, 150, 300 mg/kg) and metal dynamics was studied in spinach crop. Experimental results showed that the addition of pressmud upto 10 mg/kg had decreased different phytoremediation indices in spinach crop. Whereas, increasing Pb level enhanced the indices' values, indicating accumulation of significant amount of Pb in spinach biomass. However, application of pressmud (upto 10 mg/kg) reduced the bioconcentration factor (BCF) from 0.182 to 0.136, transfer factor (TF) from 0.221 to 0.191, translocation efficiency 66.11–59.34%; whereas, Pb removal enhanced from 0.063 to 0.072 over control treatment. These findings suggest that application of pressmud declined Pb concentration, the BCF and the TF in test crop which lead to less chances of adverse effect in human. These information are very useful for effectively managing wastewater irrigated agricultural crop production systems.

Keywords Food chain contamination · Lead contamination · Phytoremediation indices · Pressmud · Soil health

Abbreviations

Pb	Lead
PM	Pressmud
DHA	dehydrogenase activities
TF	Transfer factor
BCF	Bioconcentration factor
TE	Translocation efficiency
EC	Electrical conductivity
OC	Organic carbon
CEC	Cation exchange capacity
ICP OES	Inductively Coupled Plasma-Optical Emission Spectrometry
ICAR	Indian Council of Agricultural Research
DTPA	Diethylene triamine penta acetic acid
DAS	Days after sowing
LSD	Least significant difference
CRD	Complete randomized design
ANOVA	Analysis of variance

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Introduction

A growing population and lifestyle changes have led to an increase in anthropogenic-mediated trace metal contamination in natural ecosystems. Industrial units established for the economic growth of a country need to follow the strong binding norms of safe discharge of effluents. The treatment and safe disposal of industrial effluents are costly affairs and major issues that discourage small and medium-sized industries and result in the discharge of industrial effluent wastes into natural water bodies (Minhas et al. 2021). These effluents contain significant amounts of metals, affecting ecosystem services and human health. Meena et al. (2020) have reported that continuous application of contaminated water builds up toxicant in soil system and affects soil health properties. Dotaniya et al. (2022) have also mentioned that poor availability of fresh water for agricultural activities forced the farmers to depend on marginal quality water and long-term use of these marginal quality water has enhanced the pollutant level compared to tubewell irrigated soils. Accumulation of significant Pb in soil affects crop germination, root and shoot growth, biomass production, transpiration, inhibit photosynthesis, imbalance mineral balance, chlorophyll production, protein formation, etc. in plants (Collin et al. 2022). Lead toxicity declines soil microbial activities and their ecological services (Wan et al. 2022). Soil DHA (dehydrogenase) enzymatic activity is adversely affected by the addition of Pb in a vertisol and has reduced from 38.9 to 18.2 µg TPF/g soil/ 24 h (Dotaniya and Pipalde 2018). It is need of hours to regularly monitor the concentration of metal in crop fields that are irrigated with industrial effluent contaminated waters. Attention must be given to the build-up of metal concentration and its effect on soil-water ecosystem functioning (Saha et al. 2017a). Soil heavy metal analysis and interpretation may reveal the presence of contaminants, active form, geographical area, primary source of pollutant (Dotaniya et al. 2019a, b). This data type has been widely used to take preventive measures to conserve natural water bodies and ecosystems. The quantity of overall contamination does not always determine the toxicity of a pollutant with respect to a living organism. Chemical characteristics of metal as well as its interaction with soil constituents, may have an impact on their transfer/availability to organisms.

The uptake of trace metals by plants from the soil can be done using the soil-plant ratio/coefficient. These ratios or coefficients are calculated to select accumulator plants used in phytoremediation as well as crops grown in contaminated soil. Saha et al. (2017a) have mentioned that trace metal movement capacity by root from soil solution greatly depends on pollutant and crop species. Many indices are used to measure metals' translocation from soil to plant. The soil-plant partition ratio/ coefficient, also known as the transfer factor (TF), which is the concentration proportion of metal in plant to metal in soil (Saha et al. 2017b). Bunzl et al. (2000) have mentioned that such information is used to identify the phytoremediation characteristics of a plant and to forecast metal absorption potential from the soil for a particular crop ideotype in a predicted concentration of contamination. Sheppard and Sheppard (1985) have elaborated their findings that transfer factor (TF) is the quantity of a metal that would be anticipated to re-locate in above and below-ground biomass at an equilibrium situation. Further, Davis et al. (1999) has described that indices indicate the metal toxicity in a particular plant part and the behavior of a crop's root and shoot. If the TF is greater than one, it suggests that crops are accumulators; if the value is near one, their behavior toward these metals is pretty unremarkable; and if the value is below one, it indicates that the plants are excluder. Phytoremediation methods of metal removal/ immobilization are important with respect to environment and are cheaper and eco-friendly (Dotaniya et al. 2020). Phytoremediation technologies are very popular among small and marginal farmers. Phytoremediation plants are hyperaccumulators and less adversely affected by metal toxicity. However, some of the leafy vegetables are also accumulate Pb in their leaves and in the long run affect human health. A plant grown in metal-contaminated areas can accumulate more metals in its tissues and may behave like a hyper-accumulator that can be decided by the calculation of different metal indices like bioconcentration factor (BCF) - metal in plant part from natural bodies; transfer factor (TF) - movement root to shoot part; translocation efficiency (TE) - the potential of plant to absorb trace metal and metal removal (Saha et al. 2017b).

The availability and form of metal ions in soil solution may change with adding organic matter. Organic matter in soil acts as a bio-absorption material, binding site for active metal ions and immobilizing them; this leads to converting a metal from a dangerous toxic to a non-toxic form and concentration (Gupta et al. 2019). Pressmud (PM) is a sugarcane industrial byproduct that poses a storage difficulty. It has a greater organic carbon content and plant nutrients in its composition. For every 100 tonnes of crushed sugarcane, approximately 3 tonnes of PM cake are left over as a byproduct (Gupta et al. 2011). It enhances the plant biomass by improving soil health and plant nutrient status. The PM is a nutrient and soluble sugar-rich soil additive that is quickly metabolized (Gunjal and Gunjal 2021). However, due to its rapid ignition properties, it is predominantly burned in brick kilns across the nation, resulting in the loss of millions of tonnes of nutrients and eventually burning harms the ecology. The use of PM for remediation of Pb contaminated agricultural field is not given much attention. The application of PM may enhance the production potential and reduce the chemical fertilizer requirement in peri-urban agricultural belts.

With this context, in this study, it is aimed to investigate the effect of PM application on phytoremediation indices under Pb-contaminated soil in the soil-plant system using spinach as a test crop.

Materials and Methods

Properties of Experimental Soils

Bulk geo-referenced soil, most popularly known as black cotton soil, was collected from the research farm of the ICAR-Indian Institute of Soil Science, Bhopal, India. For more in-depth examination of the physico-chemical characteristics of the soil, the soil was completely ground, sieved (2 mm), and stored in a plastic container. Soil texture was measured by Bouyocos hydrometer method (1963) and sand, silt and clay contents of soil were 25.4%, 17.8%, and 56.7%, respectively, and classified as clay loam. Soil pH (7.81), and electrical conductivity (0.53 dS/m) were recorded, and cation exchange capacity 40.56 cmol(+)/kg was determined by the method of Reeuwijk (2002); total amount of C (0.66%) by wet oxidation method and organic carbon 0.44% by Walkley and Black (1934) method were measured. Available plant nutrients were also measured to compute the soil fertility status. As per Subbiah and Asija (1956) procedure available nitrogen content of soil was 142 kg/ha; P content (8.64 kg/ha) by Olsen et al. (1954) and available potassium by Hanway and Heidel (1952) procedure (216 kg/ha) were estimated. Extractable metals like zinc, iron, manganese, copper, lead and nickle were 0.88, 6.15, 5.64, 2.05, 1.26 and 0.65 mg/kg, respectively, measured as mentioned DTPA extraction procedure. The total lead concentration of soil (46.94 mg/kg) was also measured by adopting the procedure given by Singh et al. (2005).

Collection and Analysis of Pressmud

Sugarcane PM was collected from sugarcane factories in Kanpur and analyzed for plant nutrient content and related properties. Standard approaches were used to analyse the properties and elemental composition of PM (Page et al. 1982). Physico-chemical properties of PM used in the study were pH (8.63) and EC 6.82 dS/m. Organic carbon was 16.21%; nitrogen, P, K, and S, concentrations were 1.06, 0.13, 0.39 and 0.43%, respectively. DTPA extracted micronutrients such as Fe, Zn, Cu, and Mn concentrations were 2900, 158, 52.3 and 196 mg/kg, respectively (Lindsay & Norvell 1978).

Treatment Details

Based on the preliminary observations on data related to Pb from the industrial effluents irrigated soils of peri-urban Bhopal, the doses of Pb were decided. Four treatments of Pb were taken T_1 (control-0 mg/kg), T_2 (100 mg/kg), T_3 (150 kg/ha) and T_4 (300 mg/kg). A cross-examination of the prepared concentration of the working solution using Inductively Coupled Plasma Optical Emission spectroscopy (ICP-OES) (Make: Perkin Elmer; model: 2100 DV with a minimum measuring level of one ppb) was done by following the QA/QC procedures. These treatments were incubated in soil for a month. After the thoroughly mixed spiked soil used for the QA/OC. The Pb concentrations of 100, 150, and 300 mg Pb/kg spiked soil measured as 100 ± 0.02 mg/kg, 149.7 ± 0.3, and 299.8 ± 0.5 mg/kg Pb, respectively. These concentrations were almost identical to the working solution. The compared limits of detection (LOD) was computed by the help of replicated Cr samples. The LOD value for treated metal (Cr) was improved (0.082 mg/kg) as comparison to the method used for calculation (0.39 mg/kg) by Stasinakis et al. (2002). Levels of PM were fixed based on organic carbon and nutrient contents as 0, 2.5, 5.0 and 10.0 g/kg. In total, there are 16 treatment combinations as below:

Pb_0PM_0	$Pb_{100}PM_0$	$Pb_{150}PM_0$	$Pb_{300}PM_0$
$Pb_0PM_{2.5}$	$Pb_{100}PM_{2.5}$	$Pb_{150}PM_{2.5}$	$Pb_{300}PM_{2.5}$
$Pb_0PM_{5.0}$	$Pb_{100}PM_{5.0}$	$Pb_{150}PM_{5.0}$	$Pb_{300}PM_{5.0}$
$Pb_0PM_{10.0}$	$Pb_{100}PM_{10.0}$	$Pb_{150}PM_{10.0}$	$Pb_{300}PM_{10.0}$

Crop Cultivation

The popular spinach variety "All Green" was planted in pot culture experiment. It had good germination (70%), and physical purity (minimum 98%). Healthy seeds of spinach were soaked in water for 24 h. The soaked seeds were treated with fungicide at the rate of 5 g per kg seeds. Seeds were placed at equal distances at a depth of less than 1 cm and covered with soil to avoid damage. The crop was fertilized with 75:40:40 kg NPK /ha. These nutrients were supplied urea, DAP, and MOP. Seedlings were thinned to have the desired plant population (5 plants/pot). Spinach was grown by following standard agronomic recommendations. The crop was harvested after 75 days of sowing (DAS) and both above ground and below ground biomasses were collected separately. The plant samples were dried in the oven (60 °C), weighed, ground and stored in polybags for further analysis of Pb concentration, plant nutrient concentration, and uptake.

Total Heavy Metal Analysis

The total contents of metals in soil were extracted using aqua-regia (comprised of 2/3 part of concentrated HCl in conjugation with 1/3 part of AR grade HNO_3). Plant samples (0.25 g) were in flask, five mL of HNO_3 was added and kept for two hours for pre-digestion. After that 5 mL of aqua-regia was added and heated. The digested content was made up to the desired volume using 0.1 *N* HCl. Then

Table 1 BCF factor of Pb influenced by Pb and PM levels (n = 3)

Pb (mg/kg)	Pressmud (g/kg)				Mean
	0	2.5	5	10	
0	0.233	0.229	0.219	0.226	0.227
100	0.163	0.170	0.156	0.133	0.155
150	0.177	0.172	0.142	0.102	0.148
300	0.156	0.153	0.120	0.082	0.128
Mean	0.182	0.181	0.159	0.136	
SD±	0.034	0.033	0.043	0.064	
lsd (0.05)	Pb = 0.007; PM = 0.007; Pb × PM = 0.014				

digested material was filtered with filter paper Whatman No. 42 and the metal concentration in the solution was measured using ICP-OES (Make: Perkin Elmer; model: 2100 DV with minimum measuring level of one ppb).

Calculation of Phytoremediation Indices

The Pb concentration in soil and different plant parts under different treatments were used to calculate the phytoremrediation indices, which indicate the dynamics of Pb from soil to plant parts. The following formulae can be used to compute various phytoremediation indices i.e., BCF, TF, TE, and crop removal to determine Pb's hyperaccumulation properties and dynamics in plant-soil interaction systems. The bioconcentration factor (BCF), which was derived by Zhuang et al. (2007), described the capacity of plant to remove contaminants proportionally from harvested tissue over to soil/water bodies. Similarly, Adesodun et al. (2010) mentioned that the lead concentration ratio in shoot and root depicted the TF. Meers et al. (2004) further improved the methods, and plant phytoremediation efficiency was measured as total concentration in shoot vis-a-vis soil application. Total plant uptake with respect to the total applied Pb was known as lead removal.

Statistical Analysis

Factorial design with two factors with four levels was adopted to study different treatments' main effect and interactive effects. The analysis of variance (ANOVA) was analyzed for different parameters in the study. To compare treatment means least significant difference (lsd) was employed at a 5% levels of significance. The SAS version 9.3 was used for the full statistical data analysis (Gomez and Gomez 1983).

Results and Discussion

When the application of Pb levels increased from control to 300 mg/kg, the BCF value decreased from 0.227 to 0.128 (300 mg/kg soils) and nearly 77.34% reduction in BCF was observed due to Pb toxicity (Table 1). Similarly, the application of PM reduced the BCF value from 0.182 to 0.136. Minimum doses of application of PM upto 2.5 g/kg soil did not affect the BCF value. Whereas, a higher application rate of PM 5 and 10 g/kg reduced BCF value to 12.63% and 25.27%, respectively.

Table 2 Transfer factor of lead in spinach affected by different doses of PM (n = 3)

Lead (mg/kg)	Pressmud (g/kg)				Mean
	0	2.5	5	10	
0	0.281	0.433	0.429	0.324	0.367
100	0.236	0.201	0.174	0.160	0.193
150	0.211	0.179	0.147	0.125	0.166
300	0.157	0.154	0.174	0.156	0.160
Mean	0.221	0.242	0.231	0.191	
SD±	0.052	0.129	0.133	0.090	
lsd (0.05)	Pb = 0.018 ; PM = 0.018; Pb × PM = 0.037				

Table 3 Translocation efficiency (%) of lead in spinach affected by different doses of PM (n = 3)

Lead (mg/kg)	Pressmud (g/kg)				Mean
	0	2.5	5	10	
0	69.143	75.201	73.923	68.787	71.763
100	64.853	60.473	60.910	61.933	62.043
150	66.797	58.700	57.343	52.590	58.858
300	63.663	60.053	61.727	54.047	59.873
Mean	66.114	63.607	63.476	59.339	
SD±	2.397	7.766	7.220	7.518	
lsd (0.05)	Pb = 2.519; PM = 2.519; Pb × PM = 5.038				

In the control treatment, a higher Pb transfer factor (0.367) was observed, and increasing the Pb application rate reduced the TF value up to 0.160 in maximum level (300 mg/kg soils). However, a drastic reduction was observed in 100 mg/kg soil with 47.41% (Table 2). A similar trend was observed with the application of PM in soil and reduced the transfer factor value from 0.221, 0.242, 0.231 and 0.191 in the 0, 2.5, 5, 10 g/kg soil applied treatments, respectively. Increasing level of PM from the control to the highest level (10 g/kg) in the experiment decreased the translocation efficiency from 66.11 to 59.34%. The percent of the decrease in TE was found to be 3.78%, 3.97% and 10.24% in 2.5, 5 and 10 g/kg PM applied pots, respectively, over control (Table 3). However, raising the graded application of Pb lowered the ability of the plant to mobilize the Pb from the lower part of the plant to the above plant biomass. Increasing the level of Pb in soil reduced the Pb transfer capacity from root to shoot. The maximum or drastic reduction in Pb translocation capacity was noticed in 100 mg Pb/kg applied treatment, after that no significant changes in TE were observed, increasing the level of Pb application in soil. Increasing the doses of PM elevated the organic carbon levels in soil (Gunjal and Gunjal 2021). The physico-chemical and microbial properties of the soil were also improved by the addition of organic matter, which was crucial for improving crop yield and soil health. Soil fertility parameters (nutrient concentration and availability), physical properties (air and water retention, structure, moisture retention), and biological health (population and diversity, enzymatic activities) might have been enhanced by an increase in organic matter content by application of PM (Dotaniya et al. 2018). Increasing biological activities in soil improved the plant nutrient dynamics and reduced the Pb uptake patter in spinach crop. Priming effect of soil organic matter also reduced the Pb active ion concentration in soil solution through immobilization process (Dotaniya et al. 2018). An increase in metal levels in plant roots might have interfered with plant uptake systems, however, translocation from root to shoot did not work in all treatment combinations. The plant possesses a specific ion absorption system, that is critical for biomass and yield. The BCF and the TF were reported to be less than one in all treatment combinations in the experiment, which indicated that spinach crops grown in the presence of Pb and PM did not perform as hyperaccumulators (Lin et al. 2007; Saha et al. 2017a). For the removal of trace metals from soil, plant spends energy. However, higher metal concentrations in soil or plant parts have been shown to trigger cell death. When PM was added to soil, it mineralized and produced many different kinds of low-molecular-weight-organic acids (Imran et al. 2021), that might have mediated reduction in heavy metal toxicity (Minhas et al. 2021). Various researchers reported similar types of findings in various crops: Khan et al. (1999) in spinach under hydroponic culture, Lin et al. (2007) in wheat, Dotaniya et al. (2019a, b, c) in spinach and Dotaniya et al. (2020) in Indian mustard.

In the experiment, it was observed that enhancing doses of lead reduced the Pb removal capacity of spinach from 0.149 to 0.037 (Table 4). A significant difference was observed at every Pb treatment, i.e., T_1 (control), T_2 (100 mg/kg), T_3 (150 mg/kg), and T_4 (300 mg/kg); whereas, Pb removal was 0.149, 0.057, 0.047 and 0.037, respectively (Fig. 1). Whereas, increasing doses of PM application had non-significantly enhanced the Pb removal by spinach crop. The interaction effect was also analysed, and it was found that the lowest Pb removal was 0.031 in the maximum dose of Pb and PM applied pot. While, the maximum dose of PM applied soil in conjugation with a minimum concentration of Pb (control) was reported to have maximum Pb removal. The incorporation of PM in soil enhanced the soil organic matter, improving the plant nutrient dynamics over a period (Gunjal and Gunjal 2021). The PM contains significant amount of plant nutrients, which support plant growth and metabolic activities. The accelerated growth of spinach might have secreted elevated levels of low molecular organic acids and bind up the active form of Pb (Dotaniya et al. 2019a, b, c). These acids enhanced the microbial population and diversity of the microorganism and converted the active form of Pb into a passive form (Wan et al. 2022). Adding organic carbon enhanced microbial growth and bio-absorption edges for the Pb adsorption and mediated the immobilization mechanism in soil (Meena et al. 2019). It was found from the results that the addition of PM declined Pb uptake from the soil by the spinach both in above and below plant parts, indicating that Pb was immobilized in the

Table 4 Different treatments mediated the Pb removal capacity (μg/g) of spinach (n = 3)

Lead (mg/kg)	Pressmud (g/kg)				Mean
	0	2.5	5	10	
0	0.103	0.155	0.165	0.172	0.149
100	0.058	0.064	0.058	0.049	0.057
150	0.053	0.055	0.046	0.035	0.047
300	0.036	0.040	0.040	0.031	0.037
Mean	0.063	0.079	0.077	0.072	
SD±	0.029	0.052	0.059	0.067	
lsd (0.05)	Pb = 0.004; PM = 0.004; Pb × PM = 0.008				

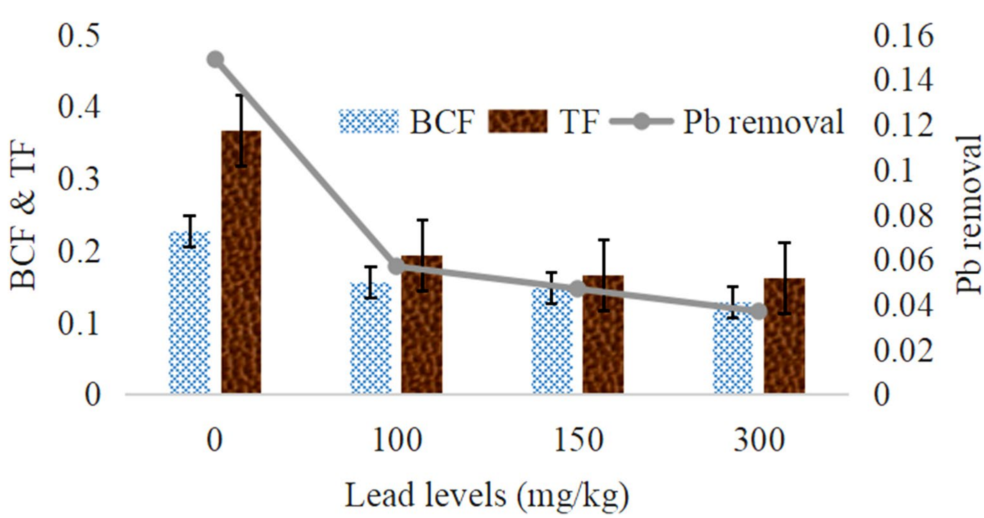

Fig. 1 Effect of pressmud on BCF, TF and Pb removal in spinach (n = 3)

soil solution. Lower Pb removal mechanism in spinach crop showed that spinach cultivated in Pb-contaminated soils could be safe to use.

Conclusions

Environmental pollution is directly associated with industrial development, population growth and lifestyle changes. However, these are necessary evil, but scientific management of generated effluent waste should be done before it is discharged into natural resources. Growing leafy vegetables in peri-urban belts of metropolitan cities' utilization of wastewater and their use for human consumption is a challenge to human health. Shortage of irrigation water and poor awareness among farmers on safe usage of industrial effluents or sewage mixed industrial effluents can be major threat. Significant amount of Pb enter from consumption of vegetable/food contaminated with Pb. In this experiment, sugarcane industrial waste PM is used for reducing the Pb uptake in spinach crop. Applying 10 g/kg PM has average reduction in the BCF (25.27%), TF (13.57%), TE (10.25%) and enhanced Pb removal (14.29%) value over control plots. In overall, Pb toxicity can be minimized in spinach crop that grown under Pb-contaminated soils with the help of PM application (10 g/kg) for crop production. These results are very important for minimizing Pb contamination in food chain and for formulating environmental policy mostly in peri-urban.

Data Availability All data generated or analysed during this study are included in this MS; publishing agency should have right to use for public domain. My manuscript has no associated data.

Declarations

Conflict of Interest The authors declare that they have no conflict of interest.

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