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Gautam, Meenu, Pandey, Divya and Agrawal, Madhoolika (2016) Phytoremediation of metals using lemongrass (Cymbopogon citratus (D.C.) Stapf.) grown under different levels of red mud in soil amended with biowastes. International journal of phytoremediation. pp. 555-562. ISSN: 1522-6514

https://doi.org/10.1080/15226514.2016.1267701

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Phytoremediation of metals using lemongrass (Cymbopogon citratus (D.C.) Stapf.) grown under different levels of red mud in soil amended with bio-wastes

Journal:	*International Journal of Phytoremediation*
Manuscript ID	BIJP-2016-0266.R2
Manuscript Type:	Original Article
Date Submitted by the Author:	06-Oct-2016
Complete List of Authors:	Gautam, Meenu; Science, Botany Pandey, Divya ; Stockholm Environment Institute Agrawal, Madhoolika; Banaras Hindu University, Ecology Research Laboratory, Department of Botany
Keywords:	Cymbopogon citratus, Phytoremediation, Red mud

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Phytoremediation of metals using lemongrass (*Cymbopogon citratus* (D.C.) Stapf.) grown under different levels of red mud in soil amended with bio-wastes

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Abstract

Due to hostile condition of red mud (RM), its utilization for vegetation is restricted. Therefore, RM with bio-wastes as soil amendment may offer suitable combination to support plant growth with reduced risk of metal toxicity. To evaluate the effects of RM on soil properties, plant growth performance and metal accumulation in lemongrass, a study was conducted using different RM concentrations (0, 5, 10 and 15 % w/w) in soil amended with bio-wastes (cowdung manure (CD) or sewage-sludge (SS)). Application of RM in soil with bio-wastes improved organic matter and nutrient contents, and caused reduction in phytoavailable metal contents. Total plant biomass was increased under all treatments, maximally at 5 % RM in soil with SS (51.7 %) and CD (91.4 %) compared to control (no RM and bio-wastes). Lemongrass acted as a potential metal tolerant plant due to metal tolerance index >100 %. Based on translocation and bioconcentration factors, lemongrass acted as potential phytostabilizer of Fe, Mn and Cu in roots and was found efficient in translocation of Al, Zn, Cd, Pb, Cr, As and Ni from roots to shoot. The study suggests that 5 % RM with bio-wastes preferably SS may be used to enhance phytoremediation potential of lemongrass.

Key words: *Cymbopogon citratus*; phytoremediation; red mud

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1 Introduction

Bauxite residue, commonly referred as red mud (RM) is generated in large quantities during Bayer's process of alumina extraction. It is highly alkaline and saline residue mainly composed of Fe_2O_3, Al_2O_3, CaO, Na_2O, TiO_2, SiO_2, P_2O_5 and VO_5 [1]. Globally, ≈ 150 million tons of RM is generated annually; management of such a quantity of waste is an increasing problem because utilization of only 2-3 % of generated residue has been estimated in a productive way [2]. Nowadays, several applications of RM have been suggested viz. manufacturing of ceramics, building materials, pigments, paints, as adsorbent and catalyst, but challenge remains to find economically viable options to utilize significant amount of residue generated every year [2]. A decade ago, slurry disposal was practiced in adjoining areas, nearby estuaries/lagoons, as filler at depleted mine- and quarry sites or stored in nearby dammed valley [3]. Improper disposal of poorly treated residue results in several environmental problems including contamination of surface and ground water through leaching, alteration in soil properties and plant community structure, and several other health issues related to human and wildlife [3]. For an instance, Ajka spill in Hungary caused contamination of vast areas of agricultural land with RM that contained elevated levels of toxic metals with consequent impacts on plants [4]. To date, dry stacking is the most popular choice of RM disposal with relatively lower risk of environmental contamination. However, study by Power *et al*. "see ref. 5" reported air pollution as major problem associated with its dry disposal practice.

To manage such waste dumps, different phytotechnologies have been developed, which are cost effective, and also offer sustainable and eco-friendly options [6]. Phytoremediation of RM is, however, a challenging task due to its high alkalinity, salinity, elevated levels of potentially toxic metals, poor water retention and nutrient supplying capacities that limit establishment of plants on RM dumps [7]. Therefore, prior to establishment of vegetation on

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such dumps, its unfavorable properties need to be improved to support plant growth coupled with enhanced phytoremediation potential. Studies have been conducted with RM in combination with bio-wastes (sewage-sludge, vegetative dry dust, animal manures, bacteria and mycorrhiza) as soil amendments, which improved soil properties and plant performance with low phytotoxic effects [7,8]. These studies have prompted us to assess the utilization of animal manure and sewage-sludge in combination with RM, which may boost the levels of dissolved organic carbon and nutrient availability, and lower phytoavilability of toxic metals. This strategy offers twin benefits in industrial as well as organic waste management [9,10].

Cymbopogon citratus (D.C.) Stapf., commonly known as lemongrass, is a metal tolerant plant that withstand the harsh environmental conditions [11]. Israila *et al.* "see ref. 12," have identified lemongrass as a potential metal (Cd, Ni and Pb) accumulator grown on scrap-metal dumpsite at Dakace, Zaria-Nigeria, and suggested its suitability for phytoremediation of metal contaminated sites. Lemongrass cultivation is also widely practiced for stabilization of slopes and restoration of alkaline and saline soils [13].

The present study was conducted using lemongrass grown under varying red mud treatments in soil amended with cowdung manure or sewage-sludge (1) to assess the physico-chemical properties of soil under different soil treatments (2) to evaluate the phytoremediation potential of lemongrass, and (3) to assess the influence of metals on plant growth performance under varying soil treatments.

2 Materials and methods

The experiment was conducted in Botanical Garden, Banaras Hindu University (25°18′ N 82° 01′ E and 76.19 m above sea level) from February 03 to August 03, 2013. During the experiment, mean monthly maximum and minimum temperature were 41.4 and 12.5° C,

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respectively. Mean monthly maximum and minimum relative humidity were 89.4 and 29.3 %, respectively, and total rainfall during the period was 110.0 mm.

Red mud was obtained in the form of dry lumps (≈70 % solid cake) from the dumping yard, situated nearly 900 m from the premises of HINDALCO Industries Ltd., Renukoot, India. Lemongrass, known for its medicinal value was found growing naturally in the planted areas of RM dumps under plantation project called "Sanjeevani." Sewage-sludge (SS) and cowdung manure (CD) were collected from Dinapur municipal sewage treatment plant, Varanasi and dairy farm, B.H.U., respectively. Garden soil (C) was dug out upto 30 cm depth from Botanical Garden, B.H.U. Lemongrass was incurred from Faculty of Ayurveda, Institute of Medical Sciences, B.H.U. After removing stone and plant materials, RM, SS, CD and C were air dried, crushed, passed through sieve (2 mm mesh size) and mixed in a definite proportion to obtain varying soil treatments (Table S1-1). Prior to the present study, an experiment was conducted using lemongrass grown under varying RM concentrations (0, 10, 20, 30, 40 and 50 % w/w) in soil amended with CD or SS and maximum biomass was obtained under 10 % RM treatment. Therefore, we have selected 5, 10 and 15 % RM w/w with CD or SS for the present study.

Different soil treatments thus obtained were filled into cylindrical plastic pots (diameter, 25 cm; height, 50 cm). In total, there were 90 pots (9 treatments × 10 replicates); each filled with 10 kg of different soil treatments. Pots were left at experimental site for 14 days for pathogen destruction, physico-chemical stabilization and proper conditioning of treated soil due to bio-waste amendments [14]. On fifteenth day, one plant slip (shoot length: 15 cm, root length: 5 cm) of lemongrass was transplanted into each pot. Amount of water was standardized to avoid the leakage from the pots at different ages. Amounts of watering were 0.4, 0.6, 0.8 and 1.0 L between 0-40, 41-80, 81-120, 121-180 DAT, respectively given in each pot, every alternate day.

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For physico-chemical analyses of different soil treatments, samples from three pots per treatment were taken out using soil corer (5 cm diameter and 10 cm depth) just before transplantation. Each sample was air dried, crushed and passed through sieve of 2 mm mesh size. The pH and EC of samples were measured in aqueous suspension of 1:5 (w/v) using pH meter (Model EA940, Orion, U.S.A) and conductivity meter (Model 303, Systronics, India), respectively. Total organic carbon (TOC) and total nitrogen (TN) contents were determined following Walkley and Black's rapid titration method "see ref. 15" and Gerhardt automatic N analyzer (Model KB8S, Germany), respectively. Available phosphorous (AP) was estimated by Olsen's method [16]. Cation exchange capacity (CEC) through exchangeable cations extraction was determined following repeated leaching method [17]. Phytoavailable metals in different soil treatments were extracted using 0.05 M EDTA solution [18]. Exchangeable cations and EDTA extractable metal contents were determined using Atomic Absorption Spectrophotometer (Analyst-800, Perkin Elmer Inc., Norwalk, CT, USA).

Harvesting was done in triplicate by taking out entire plant along with roots and soil from the pots at 180 days after transplantation (DAT). Plants were gently jerked and after separating easily removable soil, roots were washed under running water to remove adhering soil particles. Thereafter, plant parts were washed twice using de-ionized water to avoid metal contamination. Afterwards, roots and shoot were separated and oven dried at 80^{o} C until constant weights were attained. Dry weights of roots and shoot were measured for biomass determination.

Dried samples of roots, shoot and soil in triplicate were homogenized by grinding to fine powder using mortar and pestle. Contents of Al, Fe, Zn, Cu, Mn, Ni, Pb, Cr and Cd were determined on Atomic Absorption Spectrophotometer (AAS) (Analyst-800, Perkin Elmer Inc., Norwalk, CT, USA) after acid digestion (HNO_3 and $HClO_4$ in 9:4 ratio) following method of Gaidajis "see ref. 19". Moreover, AAS equipped with mercury hydride system

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(MHS-15, Perkin Elmer Inc., Shelton, CT, USA) was used to determine As content in acid digested plant- and soil samples following the method by Welz and Šucmanová "see ref. 20". The choice of metals viz. Fe, Al, Ni, Cr, Cd, Ni, As and Pb was based on their high concentrations in RM and potentially phytotoxic effects [21]. Micronutrients such as Zn, Mn and Cu showed low phytoavailabilities at circum-neutral to alkaline pH [22].

Precision and accuracy of analysis was assured through repeated analysis of samples against National Institute of Standard and Technology, Standard Reference Material (NBS SRM-1570) for all metals. Blank and drift standards (Sisco Research Laboratories Pvt. Ltd., India) were run after every five sample runs to calibrate the instrument. Results were found within ±2 % of the certified value. Coefficients of variation of replicate analysis were determined for different determinations and precision of analysis. Variations were found to be less than 10 %.

Metal tolerance ability of the plant was determined through metal tolerance index (MTI) "see ref. 23," whereas its phytoextraction potential was estimated using translocation (TF) and bioconcentration factors (BCF_{plant}), calculated using the formulae by Qihang *et al*. "see ref. 24".

$$MTI\ (\%) = \frac{Total\ plant\ biomass\ under\ treatment}{Total\ plant\ biomass\ under\ control}$$

$$TF = \frac{Metal\ contents\ in\ shoot\ tissue}{Metal\ contents\ in\ root\ tissue}$$

$$BCF_{plant} = \frac{Metal\ contents\ in\ plant\ tissue}{Metal\ contents\ in\ soil}$$

Statistical significance of differences between physico-chemical properties of different soil treatments, total and phytoavailable metal contents, plant biomass, metal contents in plant, TF and BCF values for different treatments were tested by one way analysis of variance

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(ANOVA) followed by Duncan's multiple range test as post hoc. Prior to conducting significance testing, normality and homoscedasticity of data were tested with Kolmogorov-Smirnov and Levene's test, respectively and distribution was found normal based on resulted p values above 0.05 in all cases. Pearson's correlation and linear regression analysis were performed for metal contents in soil and plants grown under different treatments. All the statistical tests were performed using SPSS software, IBM SPSS Statistics 20.0 (IBM, Armonk, NY, USA).

3 Results and discussion

3.1 Soil properties before transplantation

Due to high alkalinity and EC value of RM (Table S1-2), its increasing concentrations in bio-waste amended soil resulted in significant increases in their levels (Table 1). The pH of RM was above, whereas EC value was below threshold levels suggested as remediation target [25]. However, RM treated soils exhibited circum-neutral pH and were in the range of remediation target (pH, 5.5-9.0 and EC, < 4mS cm^{-1}) [25]. High pH and EC may be ascribed to utilization of caustic soda during Bayer's process, resulting in an increase in soluble and free forms of caustic soda content in RM [1]. Insignificant change in CEC values from CD_{RM0} to CD_{RM15} and SS_{RM0} to SS_{RM15} treatments may be due to its low value in RM compared to CD and SS, respectively (Table 1). In RM, more than 50 % of exchangeable cations are dominated by Na^+, whereas in both CD and SS, the same is dominated by exchangeable Na^+, Ca^{2+} and Mg^{2+} ions. Bio-wastes in combination with RM are known to ameliorate the low CEC value of RM thereby prompting exchange of other macronutrients (Ca^{2+}, Mg^{2+} and K^+ ions), essential for better plant growth [26].

Notably higher levels of TOC, TN and AP were observed in CD and SS compared to RM (Table S1-2) and C (Table 1). Being low in TOC (0.63 %), TN (trace to 0.02 %) and AP

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(0.09 %), RM is limited in major nutrients required for plant's growth and development [27]. Organic matter and humus are crucial for soil pedogenesis [28]. Addition of CD and SS in soil prior to RM addition resulted in significant increases in the levels of TOC, TN and AP in CD_{RM0} and SS_{RM0} treatments, respectively compared to control (Table 1). However, increasing RM treatments in CD or SS amended soil caused gradual decline in TN and AP, whereas TOC showed insignificant change across treatments. However, their levels were significantly higher compared to control. Organic carbon in soil is of paramount importance in determining soil aggregate stability which consequently influences gaseous exchange, phytoavailable nutrients, water storage and transport [29]. Jones *et al.* "see ref. 9" found that addition of poultry manure to RM stimulated active microbial biomass, thus enhancing soil aggregate stability. RM was found deficient in TN content; therefore its increasing proportions in bio-waste amended soil may have caused reduction in its value across treatments. A decline in AP may be attributed to appreciable amount of sesquioxides (>50 %) in RM resulting in high P retention capacity due to formation of insoluble metal phosphates [22].

Contents of Al, Fe, Pb, Cd, As, Cr and Ni were higher, whereas Mn, Zn and Cu contents were found low in RM compared to CD and SS (Table S1-2). Xue *et al.* "see ref. 1" reported nutrient deficiency as a potential limiting factor for vegetative growth on RM. When compared with soil quality guidelines defined by NOAA "see ref. 30", contents of Cd and Cr (in RM and SS), Cu (in RM, SS and CD) and Zn (in SS) were higher than their suggested values. Increasing RM concentrations in bio-waste amended soil increased metal contents, but trends were different under CD and SS amendments (Table 1). Trend of total metal contents was Fe>Al>Mn>Zn>Cu>Cr>Pb>Cd>Ni>As under CD_{RM0} to CD_{RM15} and Fe>Al>Zn>Mn>Cr>Cu>Cd>Pb>Ni>As under SS_{RM0} to SS_{RM15} treatments. Higher contents of Zn, Cr and Cd than Mn, Cu and Pb, respectively in later case may be attributed to many

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folds higher Zn, Cr and Cd contents due to SS than CD amendment in soil following RM treatments. Contents of Cu, Zn, Cd and Cr under CD_{RM0} to CD_{RM15} treatments were within, whereas under SS_{RM0} to SS_{RM15} treatments were above the soil quality guidelines of NOAA [30]. Moreover, As, Pb and Ni contents in all soil treatments comply with NOAA soil quality guideline. Studied metal contents when compared with their screening levels in soil required for plants, Mn, Zn, Cu, Cd and Cr under all soil treatments, whereas Pb and Ni under SS_{RM5} to SS_{RM15} treatments exceeded their prescribed values by NOAA [30].

Phytoavailable metal contents were maximum in SS followed by CD, C and RM (Table S1-2). Micronutrients such as Fe, Mn, Cu, Zn and Ni, were available in lower concentrations in RM due to formation of immobile metal complexes under alkaline condition and adsorption of metals on the surface of RM [27]. In soil, CD and SS amendments caused significant increases in phytoavailable metals in CD_{RM0} and SS_{RM0} treatments, respectively compared to C. Furthermore, increasing RM concentrations reduced phytoavailable metal contents significantly compared to CD_{RM0} and SS_{RM0}, respectively (Table 1). Circum-neutral pH induced by RM treatments may favor precipitation of metals followed by increase in metal sorption by charged colloids of RM. Cancrinite and hematite are two principal phases of RM, which provide adsorption capacity to RM [31]. Phytoavailable metal contents under different soil treatments thus showed the trend in the order of Fe>Al>Mn>Zn>Cu>Cr>Ni>Cd>As>Pb. Low phytoavailable Mn is a major problem associated with oxidizing environment which favors oxidation of Mn^{2+} to insoluble Mn^{4+} in RM [32]. Moreover, increase in Fe-oxide content due to RM addition might have played a significant role in reducing phytoavailable Zn, Ni, Pb and Cd contents across treatments [22]. Relatively low phytoavailable Cu content may be ascribed to hydroxyl and carboxyl groups supplied by bio-waste amendments, which lead to formation of insoluble and immobile Cu-complexes, resulting in reduced risk of Cu-phytotoxicity [33]. However, higher phytoavailable Cu than Ni content may be because of

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increase in aqueous dissolved organic carbon content due to RM addition, which may have increased the mobility of Cu followed by Ni complexed to organic matter [21]. Reduced phytoavailable Cd and Pb contents may be ascribed to their strong antagonistic relationship with Mn and Cu, respectively. Whereas, appreciable amount of Fe-sulfate in RM at circum-neutral to alkaline pH is known to reduce labile As content efficiently in soil [34]. Thus, low phytoavailabilities of metals in aged bauxite residues are more pronounced than spiked one, which may be attributed to their complex nature and relative behavior of different metals [3].

3.2 Plant biomass

Root, shoot and total plant biomass were significantly higher under RM treatments compared to C (Fig. 1), indicating that they are efficient enough to tolerate high metal contents in soil and in their tissues [35]. Maximum increase in root and shoot biomass was observed under 5 % RM treatment followed by a decline under further treatments. Total plant biomass was significantly increased by 59.7 and 91.4 % under CD_{RM5} and SS_{RM5} treatments, respectively compared to control (Fig. 1). Reduction in plant biomass under 10 and 15 % RM treatments may be attributed to metal toxicity. In a similar study conducted with *Festuca rubra*, maximum increase in its biomass was found in soil treated with 5 % RM concentration, which was ascribed to decrease in metal contents in grass due to increased soil pH [36]. Decrease in biomass of plants growing in metal polluted soil is closely related to growth and development of roots, because roots are first organ exposed to elevated metal contents in soil. Due to metal toxicity, poorly developed roots may lead to decrease in nutrient transport and water uptake by plant, thereby affecting shoot and total plant biomass [37].

3.3 Metal contents in lemongrass

Among all metals, content of Fe was found maximum, whereas As content was found minimum in plant biomass under different RM treatments in soil amended with CD or SS

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(Table 2). Low As uptake by the plant under RM treatments may be due to its reduced phytoavailable contents [34]. Metal contents in lemongrass when compared to phytotoxic thresholds for medicinal plants showed that all the metals were within the prescribed threshold levels [38]. However, Cd content in plant under RM treatments in SS amended soil exceeded its threshold level (5-30 ppm) [38]. Increase in Cd content in plant may be due to its higher level in SS compared to CD amendments in soil and low adsorption capacity of Cd (1.35 mmol g^{-1}) onto adsorbent surface of RM resulting in its increased mobility and phytoavailability [31]. It should be noteworthy that although Cd content in the plant was higher than its threshold level, no such severe phytotoxic effect of Cd on total plant biomass was reflected which may be due to synthesis of Cd induced phytochelatin complex that masks Cd-phytotoxicity [39]. Moreover, Fe, Cu, Cr, Cd, Ni, Zn (except under C and CD_{RM0} to CD_{RM15} treatments) and Pb (except under C and CD_{RM0} treatments) contents in plant exceeded the WHO safe limits (20, 10, 1.5, 0.3, 1.5, 50 and 10 mg kg^{-1}, respectively) for medicinal plants, however, Mn and As were within their safe limits (200 and 5 mg kg^{-1}, respectively) under all treatments [40].

Pearson's correlation analysis showed that significant positive correlations exist between metal contents in plant biomass with their levels under different RM treatments in bio-waste amended soil (Table S1-3). Indeed, RM was found to be a predominant source of Al, Fe, Pb, Cd, Ni, Pb, Cr and As, whereas CD and SS acted as sources of Mn, Zn and Cu in different soil treatments (Table 1). Furthermore, linear regression models also confirmed that increasing concentrations of RM in soil amended with CD or SS significantly increased the metal contents in lemongrass (Figs. S1-1 and S1-2). Similar relationships were reported for lemongrass grown in soil treated with increasing contents of Cd, Hg and Pb [35]. A weak magnitude of relationship for Mn (R^2 = 0.657) and As (R^2 = 0.722) in plant under different RM treatments in CD amended soil may be attributed to their antagonistic relationship with

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Cd [14,41]. Moreover, moderate relation for Zn (R^2=0.776) and Ni (R^2=0.757) may be because of competitive behavior of Fe with Ni followed by Zn for binding sites in plant [42]. This could also be attributed to relatively low phytoavailable Zn and Ni contents compared to Fe in treated soil.

3.4 Metal tolerance index of lemongrass

Assessing metal tolerance is of paramount importance while selecting plant for phytoremediation [43]. To determine metal tolerant behavior of plant, one of the most common parameters used is metal tolerance index (MTI) [44]. Based on total plant biomass, MTI of lemongrass under different soil treatments showed remarkable differences in tolerance to high metal contents compared to C. The MTI under CD_{RM0}, CD_{RM5}, CD_{RM10}, CD_{RM15}, SS_{RM0}, SS_{RM5}, SS_{RM10} and SS_{RM15} treatments were 129.7, 151.7, 126.7, 115.1, 150.1, 191.4, 167.4 and 120.7 %, respectively. Although MTI varied with increasing metal contents due to RM addition in bio-waste amended soil, its value was found >100 % under all treatments, thereby categorizing lemongrass as metal tolerant plant (Table S1-4), capable to grow in metal polluted soil. Metal tolerant plants are known to elicit their ability to tolerate metal induced reactive oxygen species by increasing their enzymatic and non-enzymatic antioxidants, proline accumulation, synthesis of phytochelatins and metallothioneins for detoxification and homeostasis [35,45].

3.5 Translocation and bioconcentration factors for metals

The translocation factor (TF) determines effectiveness in metal movement from roots to shoot, whereas bioconcentration factor (BCF_{plant}) is used in determining the uptake and accumulation of metals from soil into plant biomass [46]. Plant with BCF_{plant} value >1, indicates its efficiency to uptake and accumulate metals from soil, while with BCF_{plant} value <1 is metal excluder [47]. Thus by comparing BCF_{plant} and TF values of metals under different soil

treatments, we can compare plant's ability in extracting metals from soil and then translocating them to easily harvestable part of the plant. Plant with both BCF_{plant} and TF values >1 can be used as a suitable candidate for hyper-accumulation of metals, while plants with TF <1 can phytostabilize metals in roots [48].

No significant change in TF value for Cu was found under all treatments, whereas Al, Zn, Pb, Cd, Ni, Cr and As showed maximum TF values under CD_{RM0} and SS_{RM0} followed by a gradual decline under increasing RM treatments (Table S1-5). A decline in TF values under RM treatments may be attributed to decrease in available metals absorbed by the roots and their further transport to shoot. The TF values of Fe, Mn and Cu were found < 1 indicating restricted movement of metals from roots to shoot. However, TF values of Al, Zn, Pb, Cd, Ni, Cr and As were found >1 which illustrates the efficiency of plant in transport of metals from roots to shoot. Effectiveness of lemongrass in metal (Al, Zn, Cd, Pb, Ni, Cr and As) movement from roots to shoot is more likely due to an efficient metal transporter "see ref. 49" and possibly due to metal sequestration in leaf vacuole and apoplast "see ref. 50". TF values of Pb, Cd and Ni were higher, whereas that of Cu and Zn (except in C) were found lower than those reported in lemongrass grown on scrap metal dumpsite [12]. Thus based on TF values, lemongrass was found efficient in translocation of Al, Zn, Cd, Pb, Ni, Cr and As from roots to shoot and acted as a potential phytostabilizer of Fe, Mn and Cu in roots.

The BCF_{plant} values for Mn, Zn, Cu, Ni and Pb increased, whereas for Al, Fe, Cd, Cr and As decreased with increase in RM concentration in bio-waste amended soil, BCF_{plant} values for all metals were < 1 under all soil treatments (Table S1-5). BCF_{plant} values for Zn, Cu, Pb, Cd and Ni in the plant were found less than those reported in lemongrass collected from scrap metal dumpsite [12]. Low BCF_{plant} values for studied metals in the plant under different soil treatments may be attributed to decrease in their phytoavailable contents with increase in soil

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pH and metal adsorption capacity of RM [1]. In the present study, based on BCF_{plant} values, lemongrass acted as a potential metal excluder.

4 Conclusions

The study showed that red mud in combination with sewage-sludge in soil led to more significant improvement in organic matter and nutrient contents compared to cowdung amended soil. Increasing red mud concentrations in bio-waste amended soil increased total metal contents with simultaneous reduction in their phytoavailable contents. Studied metals except Cd in lemongrass were found within the phytotoxic thresholds for medicinal plants. All metals except Mn exceeded the WHO safe limit for consumption in medicinal plants. A significant improvement in plant biomass was observed under red mud treatments compared to control soil with maximum increase under 5 % RM treatment. Lemongrass was found to be a potential metal tolerant plant as metal tolerance index was more than 100 %. Based on translocation factor, lemongrass acted as a potential phytostabilizer of Fe, Mn and Cu in roots, whereas it was found efficient in translocation of Al, Zn, Cd, Pb, Ni, Cr and As from roots to shoot. Moreover, lemongrass was identified as a potential metal excluder due to bioconcentration factors <1 for studied metals. The study suggests that 5 % RM in combination with bio-wastes; preferably SS may be used as a soil quality enhancer by reducing metal toxicity and also make it suitable for lemongrass cultivation coupled with enhanced phytoremediation efficiency. However, a much more detailed investigation over the likely environmental impacts of this strategy under field condition is needed.

6 Acknowledgements

Authors are thankful to the Head, Department of Botany, Institute of Science, B.H.U., Varanasi and HINDALCO Industries Ltd., Renukoot, U.P. for providing all the necessary facilities during the research work. We are thankful to AXA Junior Research (Post-doctoral)

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Fellowship, York, United Kingdom and Council of Scientific and Industrial Research (CSIR), New Delhi, India for financial assistance in the form of Research Associateship (Divya Pandey) and Senior Research Fellowship (Meenu Gautam), respectively. We are also grateful to anonymous reviewers for their critical comments and helpful suggestions which helped us in improving the manuscript.

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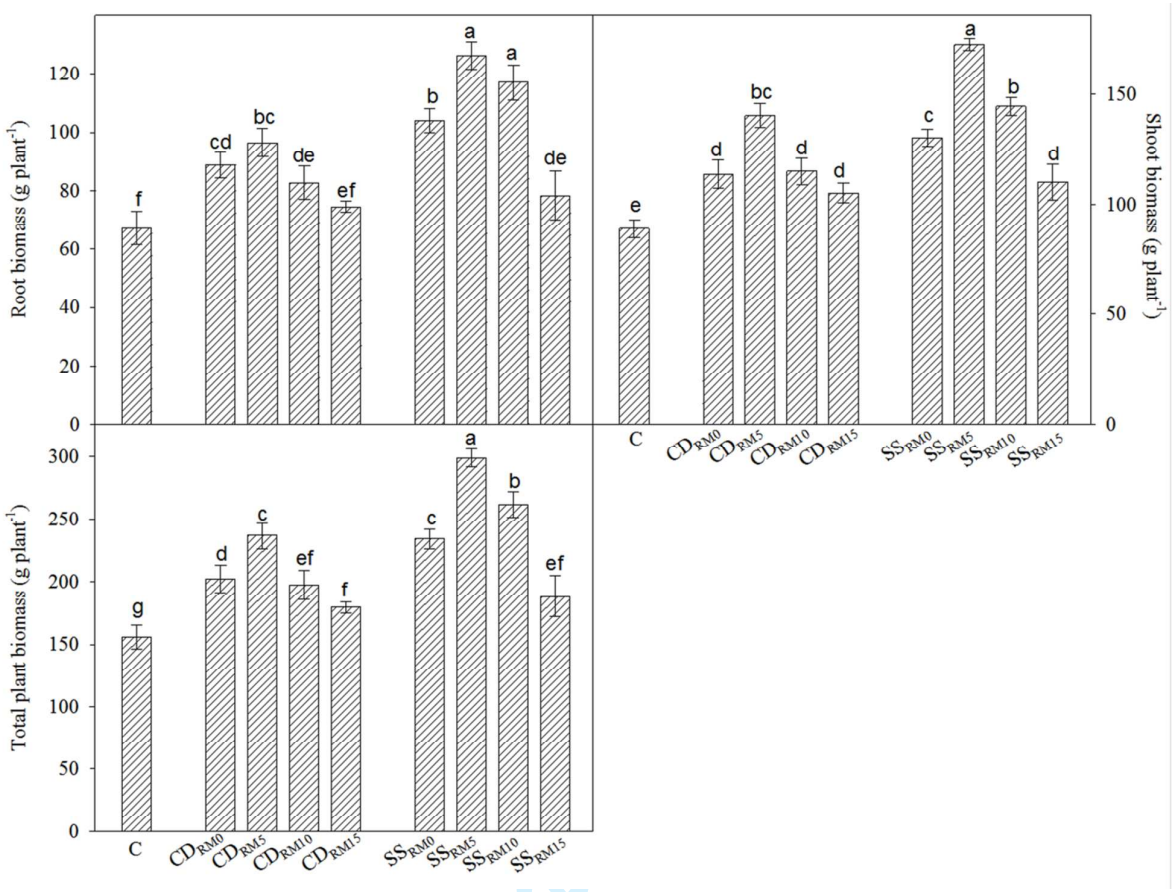

Fig. 1 Root, shoot and total plant biomass of lemongrass grown under different RM treatments in soil amended with CD or SS. Values are mean ± SE. Different letters indicate significant differences at $p<0.05$ according to Duncan's test. RM: red mud; C: control; CD_{RM0}: soil with CD; CD_{RM5}: 5 % RM in CD_{RM0}; CD_{RM10}: 10 % RM in CD_{RM0}; CD_{RM15}: 15 % RM in CD_{RM0}, SS_{RM0}: soil with SS; SS_{RM5}: 5 % RM in SS_{RM0}; SS_{RM10}:10 % RM in SS_{RM0} and SS_{RM15}:15 % RM in SS_{RM0}.

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Table 1 Selected physico-chemical properties, total and phytoavailable metal contents in control and RM treatments in soil amended with CD or SS (Mean ± SE)

Parameters	C	CD_{RM0}	CD_{RM5}	CD_{RM10}	CD_{RM15}	SS_{RM0}	SS_{RM5}	SS_{RM10}	SS_{RM15}
pH	7.87±0.05^{cd}	7.33±0.01^{f}	7.82 ±0.02^{d}	8.09 ±0.02^{b}	8.27 ±0.01^{a}	7.71±0.04^{e}	7.82 ±0.01^{d}	7.94±0.01^{c}	8.21±0.04^{a}
EC (mS cm^{-1})	0.18 ±0.01^{i}	0.50±0.00^{e}	0.54±0.00^{d}	0.56±0.00^{c}	0.86±0.01^{b}	0.32±0.00^{h}	0.43±0.00^{g}	0.47±0.00^{f}	1.15 ±0.00^{a}
TOC (%)	0.56 ±0.10^{d}	4.19±0.06^{c}	4.23±0.28^{c}	4.56 ±0.07^{bc}	4.40±0.10^{bc}	4.77±0.21^{ab}	5.15±0.06^{a}	5.18±0.11^{a}	5.09±0.06^{a}
TN (%)	0.12±0.01^{f}	0.54±0.02^{a}	0.48±0.02^{abc}	0.30±0.01^{d}	0.21±0.01^{e}	0.53±0.001^{ab}	0.50±0.03^{abc}	0.46±0.03^{bc}	0.45±0.04^{c}
AP (mg kg^{-1})	31.83±1.64^{g}	443.04±37.98^{cd}	416.49±14.97^{de}	393.78±6.31^{ef}	377.26±12.17^{f}	566.64±18.93^{a}	520.32±5.04^{ab}	485.22±4.54^{bc}	463.39±5.74^{cd}
CEC (meq100 g^{-1})	4.16±0.11^{bc}	3.76±0.06^{c}	3.78±0.03^{c}	3.99±0.37^{c}	4.14±0.02^{bc}	4.50±0.06^{ab}	4.57±0.05^{ab}	4.65±0.03^{a}	4.71±0.02^{a}
Total metal contents (mg kg^{-1})									
Al*	0.08±0.00^{g}	0.37±0.01^{g}	8.58±0.21^{e}	19.05±0.22^{c}	24.31±0.19^{b}	3.14±0.24^{f}	16.62±0.13^{d}	24.70±0.20^{b}	32.95±0.23^{a}
Fe*	2.57±0.03^{h}	3.38±0.22^{h}	16.26±0.11^{g}	31.84±0.48^{e}	48.65±0.56^{c}	21.81±0.23^{f}	35.80±1.12^{d}	51.91±1.06^{b}	67.39±1.38^{a}
Mn	260.67±1.76^{e}	285.43±1.57^{d}	286.68±0.89^{d}	291.42±4.57^{d}	303.42±2.02^{c}	308.39±2.19^{bc}	310.27±4.53^{abc}	314.97±1.49^{ab}	318.16±1.35^{a}
Zn	134.49±7.49^{e}	146.07±2.16^{e}	147.54±3.21^{de}	163.14±2.60^{cd}	170.61±4.75^{c}	540.94±7.01^{b}	536.03±5.28^{b}	550.94±7.01^{ab}	560.32±5.31^{a}
Cu	70.55±0.79^{f}	76.20±1.38^{e}	79.92±2.08^{e}	88.57±1.93^{d}	116.28±2.07^{c}	177.57±2.27^{b}	179.63±2.55^{ab}	181.79±1.30^{ab}	184.24±0.62^{a}
Pb	18.27±0.48^{i}	18.79±0.72^{h}	25.53±0.46^{g}	43.26±0.75^{e}	45.94±0.43^{d}	36.49±0.23^{f}	49.03±0.08^{c}	61.80±0.11^{b}	73.31±0.21^{a}
Cd	1.46±0.05^{h}	2.14±0.02^{h}	12.75±0.06^{g}	33.50±0.11^{f}	40.25±0.17^{e}	50.98±0.83^{d}	73.30±1.65^{c}	93.08±2.89^{b}	116.58±3.51^{a}
Ni	11.69±0.81^{f}	12.48±0.34^{f}	13.22±0.25^{f}	16.79±0.38^{e}	18.78±0.75^{e}	26.55±0.25^{d}	35.27±1.24^{c}	39.68±1.04^{b}	49.75±0.27^{a}
Cr	16.94±0.75^{g}	17.97±0.39^{g}	68.31±0.96^{f}	84.07±1.42^{ef}	112.12±3.79^{e}	310.05±5.03^{d}	351.06±3.89^{c}	410.96±0.43^{b}	476.42±37.34^{a}
As	0.13±0.01^{g}	0.59±0.01^{fg}	0.99±0.06^{ef}	1.37±0.06^{e}	1.88±0.06^{d}	3.47±0.24^{c}	3.80±0.20^{c}	4.80±0.35^{b}	5.73±0.07^{a}
Phytoavailable metal contents (mg kg^{-1})									
Al	57.86±0.79^{c}	58.21±0.83^{c}	47.00±2.52^{e}	42.33±1.45^{f}	36.00±1.00^{g}	78.51±1.87^{a}	63.67±0.88^{b}	57.89±0.48^{c}	52.36±1.06^{d}
Fe	86.43±0.86^{cd}	103.30±3.43^{b}	82.16±0.90^{de}	72.44±1.27^{ef}	66.67±0.33^{f}	135.96±3.59^{a}	111.67±4.41^{b}	92.88±1.28^{c}	81.98±0.56^{de}
Mn	35.71±1.17^{b}	35.69±0.24^{b}	32.70±0.36^{cd}	31.34±0.47^{de}	28.33±0.88^{f}	41.69±0.24^{a}	37.44±0.08^{b}	34.67±1.02^{bc}	29.63±1.93^{ef}
Zn	5.07±0.20^{g}	11.19±0.01^{d}	9.98±0.11^{e}	8.44±0.03^{f}	8.22±0.40^{f}	18.47±0.21^{a}	16.42±0.24^{b}	15.90±0.34^{b}	14.56±0.73^{c}
Cu	4.24±0.20^{ef}	6.97±0.12^{d}	4.61±0.29^{e}	3.64±0.25^{f}	2.77±0.08^{g}	12.65±0.03^{a}	10.67±0.15^{b}	9.80±0.45^{c}	9.49±0.26^{c}
Pb	0.03±0.00^{f}	0.04±0.00^{bc}	0.03±0.00^{de}	0.03±0.00^{ef}	0.03±0.00^{g}	0.05±0.00^{a}	0.04±0.00^{b}	0.04±0.00^{bc}	0.03±0.00^{cd}
Cd	0.27±0.01^{e}	0.35±0.00^{b}	0.32±0.00^{d}	0.27±0.00^{e}	0.25±0.00^{f}	0.47±0.00^{a}	0.35±0.00^{b}	0.33±0.00^{c}	0.32±0.00^{d}
Ni	2.14±0.06^{de}	2.34±0.02^{c}	2.16±0.01^{de}	2.07±0.01^{e}	1.83±0.04^{f}	2.84±0.01^{a}	2.43±0.01^{b}	2.23±0.01^{d}	2.17±0.02^{d}
Cr	4.27±0.13^{d}	2.68±0.24^{e}	2.41±0.15^{ef}	1.89±0.15^{f}	1.07±0.05^{g}	6.97±0.15^{a}	6.57±0.28^{ab}	5.96±0.06^{b}	5.23±0.03^{c}
As	0.01±0.001^{f}	0.08±0.004^{c}	0.07±0.002^{cd}	0.06±0.004^{d}	0.03±0.001^{e}	0.13±0.01^{a}	0.10±0.001^{b}	0.07±0.003^{c}	0.04±0.002^{e}

*g kg^{-1}; EC: electrical conductivity; TOC: total organic carbon; TN: total nitrogen; AP: available phosphorous; CEC: cation exchange capacity; RM: red mud; CD: cowdung manure; SS: sewage sludge; C: control; CD_{RM0}: soil with CD; CD_{RM5}: 5 % RM in CD_{RM0}; CD_{RM10}: 10 % RM in CD_{RM0}; CD_{RM15}: 15 % RM in CD_{RM0}, SS_{RM0}: soil with SS; SS_{RM5}: 5% RM in SS_{RM0}; SS_{RM10}:10 % RM in SS_{RM0} and SS_{RM15}:15 % RM in SS_{RM0}. Numbers with different letters in same row differ significantly at $p < 0.05$ as per the Duncan's test.

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Table 2 Metal contents (mg kg^{-1}) in lemongrass grown under control and RM treatments in soil amended with CD or SS (Mean ± SE).

Metals	C	CD_{RM0}	CD_{RM5}	CD_{RM10}	CD_{RM15}	SS_{RM0}	SS_{RM5}	SS_{RM10}	SS_{RM15}
Al	0.01 ± 0.00^{i}	0.19 ± 0.00^{h}	1.66 ± 0.02^{f}	2.83 ± 0.05^{d}	3.55 ± 0.06^{b}	0.45 ± 0.02^{g}	2.37 ± 0.02^{e}	3.27 ± 0.05^{c}	3.85 ± 0.13^{a}
Fe	0.63 ± 0.02^{f}	0.65 ± 0.01^{f}	2.54 ± 0.07^{e}	3.18 ± 0.01^{c}	3.58 ± 0.03^{b}	2.76 ± 0.03^{d}	3.12 ± 0.10^{c}	3.49 ± 0.06^{b}	4.06 ± 0.11^{a}
Mn	52.03 ± 2.46^{e}	51.23 ± 2.77^{e}	62.54 ± 2.95^{d}	75.28 ± 0.26^{c}	84.64 ± 1.77^{b}	74.58 ± 0.96^{c}	88.87 ± 2.36^{b}	97.54 ± 3.11^{a}	102.39 ± 0.23^{a}
Zn	25.35 ± 1.30^{g}	29.03 ± 0.67^{fg}	29.59 ± 1.88^{f}	31.45 ± 0.67^{f}	37.70 ± 0.83^{e}	139.87 ± 1.79^{d}	143.67 ± 1.54^{c}	154.58 ± 0.91^{b}	161.09 ± 1.06^{a}
Cu	41.84 ± 2.77^{f}	45.21 ± 1.49^{ef}	47.14 ± 0.66^{ef}	51.40 ± 2.01^{e}	58.87 ± 0.21^{d}	69.60 ± 0.50^{c}	77.85 ± 3.96^{b}	81.68 ± 1.25^{ab}	85.98 ± 2.98^{a}
Pb	2.16 ± 0.01^{h}	6.00 ± 0.38^{g}	10.18 ± 0.18^{f}	21.96 ± 0.27^{d}	25.27 ± 0.43^{c}	16.52 ± 1.47^{e}	29.69 ± 0.10^{c}	43.35 ± 1.87^{b}	52.47 ± 1.22^{a}
Cd	1.32 ± 0.04^{g}	1.89 ± 0.03^{g}	10.33 ± 0.52^{f}	23.93 ± 0.74^{e}	26.99 ± 1.1^{e}	38.16 ± 0.54^{d}	53.82 ± 3.32^{c}	68.12 ± 2.45^{b}	79.43 ± 0.48^{a}
Ni	3.78 ± 0.02^{g}	5.95 ± 0.09^{f}	6.44 ± 0.07^{f}	7.69 ± 0.20^{e}	8.18 ± 0.16^{de}	8.69 ± 0.17^{d}	13.31 ± 0.48^{c}	17.59 ± 0.05^{b}	22.56 ± 0.06^{a}
Cr	8.38 ± 0.08^{f}	9.34 ± 0.03^{e}	9.68 ± 0.29^{e}	10.64 ± 0.19^{d}	10.74 ± 0.11^{d}	37.07 ± 0.02^{c}	38.82 ± 0.61^{b}	40.32 ± 0.02^{a}	41.03 ± 0.16^{a}
As	0.10 ± 0.01^{f}	0.74 ± 0.04^{e}	0.86 ± 0.03^{de}	0.94 ± 0.03^{d}	0.98 ± 0.02^{d}	2.15 ± 0.01^{c}	2.49 ± 0.11^{b}	2.53 ± 0.01^{b}	2.72 ± 0.01^{a}

RM: red mud; CD: cowdung manure; SS: sewage-sludge; C: control; CD_{RM0}: soil with CD; CD_{RM5}: 5 % RM in CD_{RM0}; CD_{RM10}: 10 % RM in CD_{RM0}; CD_{RM15}: 15 % RM in CD_{RM0}, SS_{RM0}: soil with SS; SS_{RM5}: 5% RM in SS_{RM0}; SS_{RM10}:10 % RM in SS_{RM0} and SS_{RM15}:15 % RM in SS_{RM0}. Numbers with different letters in same row differ significantly at $p < 0.05$ as per the Duncan's test.

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Table S1-1 Details of different treatments

Treatment	Description
C	Unamended soil (Control)
CD_{RM0}	Unamended soil and cowdung manure (2:1 w/w)
CD_{RM5}	5% red mud + 95% CD_{RM0}
CD_{RM10}	10% red mud + 90 % CD_{RM0}
CD_{RM15}	15% red mud + 85% CD_{RM0}
SS_{RM0}	Unamended soil and sewage-sludge (2:1 w/w)
SS_{RM5}	5% red mud + 95% SS_{RM0}
SS_{RM10}	10% red mud + 90 % SS_{RM0}
SS_{RM15}	15% red mud + 85% SS_{RM0}

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Table S1-2 Physico-chemical properties, total and phytoavailable metal contents in red mud, sewage-sludge and cowdung manure

Physicochemical properties	Red mud	Sewage-sludge	Cowdung manure
pH	9.21±0.01	7.72±0.02	7.77±0.01
EC (mS cm^{-1})	2.96±0.02	1.95±0.01	0.943±0.01
TOC (%)	0.30 ±0.24	6.58±0.02	5.44±0.02
TN (%)	nd	1.05±0.03	1.09±0.11
AP (mg kg^{-1})	6.46±0.02	638.32 ±8.22	572.24±3.08
CEC (meq 100^{-1} g)	4.39±0.01	6.10±0.06	5.49±0.06
Total metals (mg kg^{-1})			
Al*	187.7±4.79	2.42±0.04	0.01±0.00
Fe*	335.60±13.84	22.85±0.84	0.35±0.03
Mn	215.29±6.14	382.28±10.48	355.44±2.99
Zn	179.80±4.69	1087.66±54.44	204.87±7.71
Cu	122.61±2.95	325.01±8.79	188.63±4.34
Pb	162.86±5.93	66.26±3.64	6.91±0.51
Cd	252.18±4.24	169.76 ±7.56	0.64 ±0.05
Ni	58.51±0.96	56.90±2.61	4.72±0.16
Cr	418.75±8.32	315.42±8.42	8.04±0.23
As	18.07±1.15	10.67±0.36	0.04±0.002
Phytoavailable metals (mg kg^{-1})			
Al	187.7±4.79	2.42±0.04	0.01±0.0001
Fe	83.41±0.52	283.64±5.90	220.12±4.04
Mn	21.91±2.15	54.91±0.11	42.79±0.12
Zn	5.07±0.32	64.81±0.22	16.42±0.02
Cu	4.62±0.12	42.74±1.04	28.74±1.55
Pb	0.03±0.001	0.07±0.001	0.04±0.001
Cd	0.27±0.02	0.71±0.02	0.44 ±0.01
Ni	1.45±0.04	3.62±0.07	3.24±0.05
Cr	1.04±0.08	5.12±0.48	2.75±0.16
As	0.05±0.002	1.24±0.05	0.02±0.001

*g kg^{-1}; nd: not detected; EC: electrical conductivity; TOC: total organic carbon; TN: total nitrogen; AP: available phosphorous; CEC: cation exchange capacity

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Table S1-3 Pearson's correlation coefficient between metal contents in soil and plant biomass under control and different red mud treatments in soil amended with both cowdung manure and sewage-sludge

Metals	R-values	
	Cowdung manure	Sewage-sludge
Al	0.994***	0.991***
Fe	0.935***	0.930***
Mn	0.749***	0.909***
Zn	0.681**	0.991***
Cu	0.911***	0.925***
Pb	0.967***	0.994***
Cd	0.992***	0.994***
Ni	0.870***	0.980***
Cr	0.865***	0.955***
As	0.849***	0.945***

*p<0.05, **p<0.01, ***p<0.001, ns: insignificant,

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Table S1-4 Classification of plant based on metal tolerance index

No.	MTI (%)	Classification of plant
1	$0 \leq 25$	Highly sensitive
2	$25 \leq 50$	Sensitive
3	$50 \leq 75$	Moderate
4	$75 \leq 100$	Tolerant
5	≥ 100	Highly tolerant

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Table S1-5 Translocation (TF) and bioconcentration (BCF_{plant}) factors for studied metals in lemongrass grown under control and RM treatments in soil amended with CD or SS (Mean ± SE).

Metals	Parameters	C	CD_{RM0}	CD_{RM5}	CD_{RM10}	CD_{RM15}	SS_{RM0}	SS_{RM5}	SS_{RM10}	SS_{RM15}
Al	TF	1.48±0.12^{d}	2.26±0.08^{a}	2.09±0.00^{ab}	2.08±0.07^{ab}	2.08±0.00^{ab}	2.10±0.15^{ab}	1.88±0.07^{bc}	1.85±0.07^{bc}	1.77±0.09^{c}
	BCF_{plant}	0.08±0.00^{e}	0.52±0.01^{a}	0.19±0.00^{b}	0.15±0.01^{c}	0.15±0.00^{c}	0.15±0.01^{c}	0.14±0.00^{c}	0.13±0.01^{cd}	0.12±0.00^{d}
Fe	TF	0.16±0.00^{d}	0.17±0.00^{cd}	0.17±0.01^{cd}	0.19±0.01^{bc}	0.23±0.00^{a}	0.12±0.00^{e}	0.15±0.01^{d}	0.17±0.00^{d}	0.21±0.00^{b}
	BCF_{plant}	0.25±0.01^{a}	0.19±0.03^{b}	0.16±0.01^{c}	0.10±0.00^{e}	0.07±0.00^{g}	0.13±0.00^{d}	0.09±0.00^{f}	0.07±0.00^{gh}	0.06±0.00^{h}
Mn	TF	0.47±0.01^{d}	0.56±0.06^{cd}	0.75±0.08^{ab}	0.80±0.04^{ab}	0.81±0.03^{a}	0.48±0.04^{d}	0.66±0.04^{bc}	0.79±0.05^{ab}	0.77±0.03^{ab}
	BCF_{plant}	0.20±0.01^{de}	0.18±0.01^{e}	0.22±0.01^{d}	0.25±0.00^{c}	0.28±0.01^{b}	0.24±0.01^{c}	0.29±0.00^{b}	0.31±0.01^{a}	0.32±0.01^{a}
Zn	TF	2.61±0.12^{d}	1.74±0.07^{de}	1.56±0.59^{ef}	1.16±0.09^{ef}	1.04±0.04^{f}	1.52±0.06^{a}	1.41±0.07^{ab}	1.32±0.09^{b}	1.30±0.05^{c}
	BCF_{plant}	0.22±0.02^{cd}	0.17±0.01^{e}	0.20±0.01^{cd}	0.19±0.00^{de}	0.22±0.00^{c}	0.26±0.01^{b}	0.27±0.01^{ab}	0.28±0.01^{ab}	0.29±0.00^{a}
Cu	TF	0.56±0.03^{a}	0.63±0.01^{a}	0.66±0.01^{a}	0.66±0.04^{a}	0.68±0.05^{a}	0.66±0.00^{a}	0.68±0.00^{a}	0.69±0.00^{a}	0.74±0.14^{a}
	BCF_{plant}	0.59±0.04^{a}	0.59±0.01^{a}	0.59±0.02^{a}	0.58±0.02^{a}	0.51±0.01^{b}	0.39±0.00^{d}	0.43±0.03^{cd}	0.45±0.01^{bcd}	0.47±0.02^{bc}
Pb	TF	2.24 ± 0.05^{b}	2.34 ± 0.14^{b}	1.80 ± 0.08^{c}	1.17±0.04^{de}	1.04±0.05^{e}	2.81±0.27^{a}	1.80±0.00^{c}	1.42±0.05^{d}	1.34±0.03^{de}
	BCF_{plant}	0.12 ± 0.00^{f}	0.33 ± 0.02^{e}	0.40 ± 0.01^{d}	0.51 ± 0.01^{c}	0.59 ± 0.02^{b}	0.45 ± 0.04^{cd}	0.61 ± 0.00^{b}	0.70 ± 0.03^{a}	0.72± 0.02^{a}
Cd	TF	1.64±0.16^{a}	1.60±0.05^{a}	1.53±0.14^{ab}	1.43±0.05^{ab}	1.29±0.02^{b}	1.59±0.01^{a}	1.51±0.11^{ab}	1.40±0.03^{ab}	1.46±0.09^{ab}
	BCF_{plant}	0.90±0.03^{a}	0.89±0.02^{a}	0.81±0.04^{b}	0.71±0.02^{c}	0.63±0.01^{d}	0.75±0.02^{bc}	0.73±0.03^{c}	0.73±0.03^{c}	0.68±0.02^{cd}
Ni	TF	2.03±0.05^{a}	2.00±0.12^{a}	1.77±0.24^{ab}	1.70±0.18^{ab}	1.68±0.03^{ab}	1.60±0.13^{b}	1.45±0.09^{b}	1.39±0.04^{b}	1.40±0.03^{b}
	BCF_{plant}	0.33 ± 0.02^{d}	0.48 ± 0.01^{ab}	0.49 ± 0.01^{a}	0.46 ± 0.01^{ab}	0.44 ± 0.02^{b}	0.33 ± 0.00^{d}	0.38 ± 0.02^{c}	0.44 ± 0.01^{b}	0.45± 0.00^{ab}
Cr	TF	1.21±0.00^{a}	1.21±0.00^{a}	1.13±0.00^{b}	1.08±0.00^{d}	1.04±0.00^{g}	1.08±0.00^{c}	1.05±0.00^{e}	1.04±0.00^{f}	1.03±0.00^{h}
	BCF_{plant}	0.50±0.02^{a}	0.52±0.01^{a}	0.14±0.01^{b}	0.13±0.00^{bc}	0.10±0.00^{de}	0.12±0.00^{bcd}	0.11±0.00^{cde}	0.10±0.00^{cde}	0.09±0.01^{e}
As	TF	1.18±0.01^{ab}	1.06±0.00^{c}	1.05±0.00^{c}	1.04±0.00^{c}	1.04±0.00^{c}	1.20±0.00a	1.19±0.01^{ab}	1.17±0.04^{ab}	1.13±0.03^{b}
	BCF_{plant}	0.78±0.09^{bc}	1.26±0.10^{a}	0.88±0.06^{b}	0.69±0.04^{cd}	0.52±0.01^{de}	0.63±0.05^{cde}	0.66±0.06^{cde}	0.53±0.04^{de}	0.47±0.01^{e}

RM: red mud; CD: cowdung manure; SS: sewage-sludge; C: control; CD_{RM0}: soil with CD; CD_{RM5}: 5 % RM in CD_{RM0}; CD_{RM10}: 10 % RM in CD_{RM0}; CD_{RM15}: 15 % RM in CD_{RM0}, SS_{RM0}: soil with SS; SS_{RM5}: 5% RM in SS_{RM0}; SS_{RM10}:10 % RM in SS_{RM0} and SS_{RM15}:15 % RM in SS_{RM0}. Numbers with different letters in same row differ significantly at $p < 0.05$ as per the Duncan's test.

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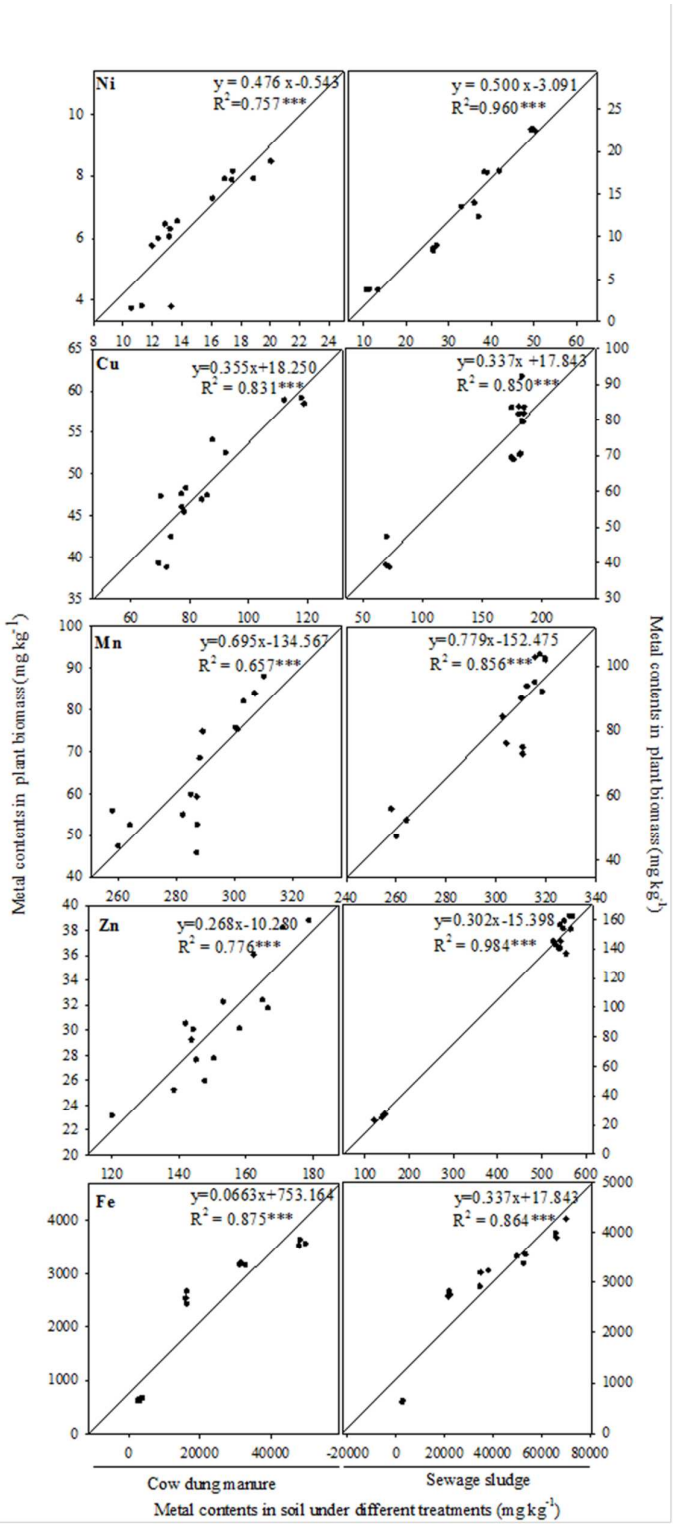

Fig. S1-1 Linear regression between metal content in soil (independent variable) and entire plant biomass (dependent variable) under control and red mud treatments soil amended with cowdung manure or sewage-sludge (n=3). Levels of significance are indicated: ns, not significant; *, $p< 0.05$; **, $p< 0.01$; ***, $p< 0.001$.

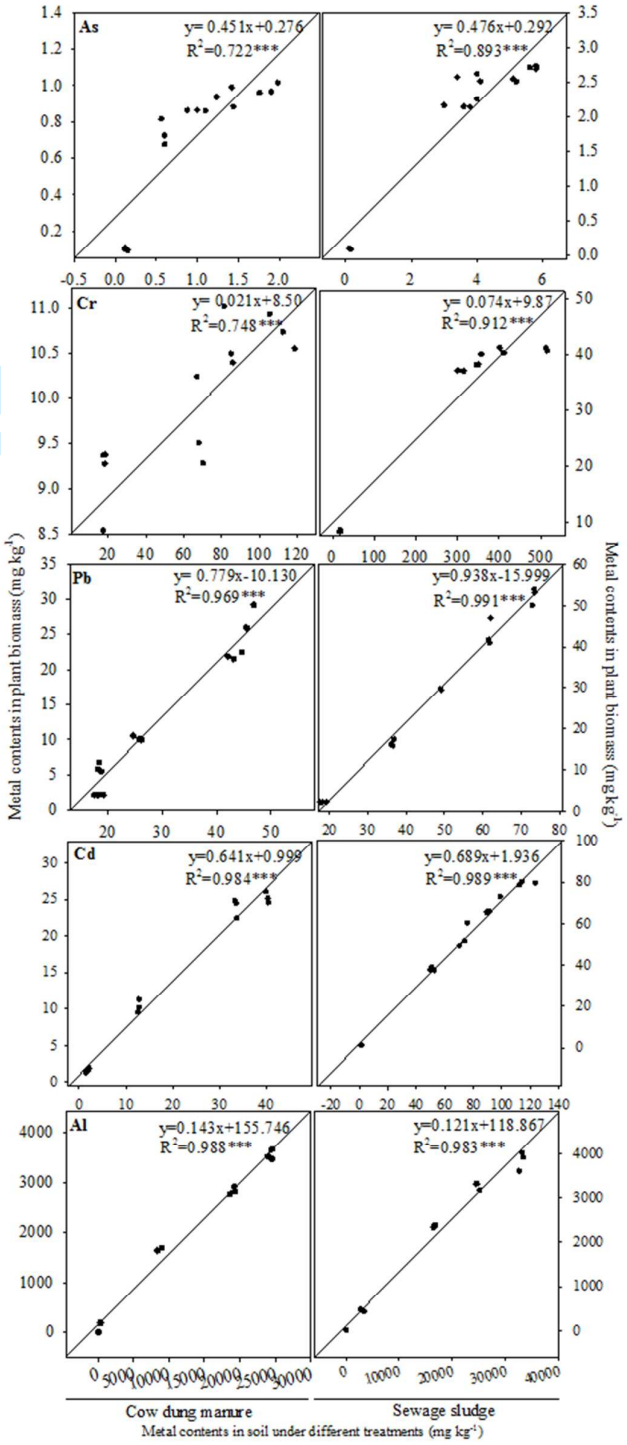

Fig. S1-2 Linear regression between metal content in soil (independent variable) and entire plant biomass (dependent variable) under control and red mud treatments soil amended with cowdung manure or sewage-sludge (n=3). Levels of significance are indicated: ns, not significant; *, $p< 0.05$; **, $p< 0.01$; ***, $p< 0.001$.