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Potential Toxic Elements Distribution Based on Interplay between Pollutants and Biochemical Remediative Amendments

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Abstract: The aim of this work is to measure in field experiment the distribution of potential toxic elements PTE's Cu, Ni and Zn in sewaged soil after application of chemical remediation technique represented by probentonite, bioremediation technique as thiobacillus thiooxidant, Mycorrhyzal (AM) conidia, and mixture of this treatments (PBC) applied before the plantation of Canola (Brassica Napus, L.), Indian Mustard (Brassica juncea Czern.) and Black nightshade (Solanum nigrum L) as a hyper accumulated plants. The obtained results showed that phytoremediation technique applied individually increased the readily available form after remediation arranged as canola> black nightshade> Indian mustard, meanwhile the application of Probentonite with phyto-accumulation plants increased the residual form in soil system. In the case of bioremediation materials applied, data showed decreasing in both readily and hardly available forms with margin increasing in moderately available forms. The final conclusion of this work documented as the application of PCB technique could be the best management practices in minimizing Zn equivalent to the save level. Different mechanisms of the remediation materials actions were discussed.

Keywords: Distribution, PTE's, remediation techniques, sewaged soils

INTRODUCTION

Nowadays, it is widely recognized that the impact of anthropogenic metal ions on the environment cannot be evaluated by measuring merely the total concentration of individual trace element species, because the mobility, bioavailability, and consequently, toxicity depend strongly on their chemical forms and type of binding. Thus, procedures for discrimination of different solid-phase associations of PTE's ore heavy metals distribution in soil system(s) are required. Barreled with the abovementioned line, such technique (Fractionation), consider to be the best tool in the evaluation of remediation technique(s) applying to minimize the hazards of such toxic pollutants in soil system. The accumulation of potential toxic elements (PTE's) in different compartments of the biosphere and their possible mobilization under environmentally changing conditions induces a perturbation of both the structure and function of the ecosystem and might cause adverse health effects to biota. Because it is at the interface between the atmosphere and the earth's crust, as well as being the substrate for natural and agricultural ecosystems, the soil is open to inputs of heavy metals from many sources [1].

Geochemical forms of heavy metals in soil affect their solubilities; therefore directly influence their potential bioavailability [2,3,4]. Thus, assessing environmental impacts of trace metals in soils by aqueous concentrations of trace metals is incomplete. In particular, it may be helpful to obtain information on the degree of bioavailability of these metals [4].

In Egypt, increasing awareness of the hazard that heavy metals in sewaged soils can cause to the environment and to humans health through presence of these pollutants in soils and its uptake by different agricultural crops, is presently pressuring society to comply with environmental regulations and develop management strategies to minimize their adverse impacts. These strategies, however, should be built according to distribution of these trace elements in soil system [6, 8].

Existing guidelines or regulations for the assessment of heavy metal contamination of soil are based on total soil metal concentrations. However, it is generally recognized that total concentrations do not necessarily provide good information on the potential bioavailability or mobility of metals in soils [9].

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Sequential fractionation techniques are being used increasingly to provide more useful assessments of soil heavy metal contamination than is possible with single extractions or total metal concentrations alone. In addition, heavy metals fractionation is a fairly widely used technique for understanding the mechanisms of heavy metals distribution in soils and to help assess bioavailability of trace metals in soils.

In pure system, [5] studied Pb distribution in three Egyptian soils to understand Pb reactions as affected by their chemical characteristics and to predict the bioavailability of Pb in theses soils. However, metals added as a salt do not undergo the same processes as those metals already present in the sewage entering the treatment plant. For this reason the studies exists with simple metal salts is controversial, since there is no certainty that the added metals will be present in the same forms as those metals already present in the sludge. This is important because there is evidence that the bioavailability of metals added to soil as metal salts may differ from the bioavailability of metals established from sludge treated soils [7,8].

The primary objectives of this study are to investigate the effect of remediation materials applied in Abo-Rawash sewage soil on PTE's distribution. This procedure will help in understanding the mode of action of these materials in such sewage soil types of soils. Also, to select the best management could be applied for minimizing the hazards of inorganic pollutants could be found in such soils.

MATERIAL AND METHODS

Soil Used

Barrier to discuss the results of PTE's distribution in the studied sewage soil, it should be mention that the studied soils were early planted with three hyperaccumulator plants commonly used to absorb PTE's i.e. Canola (Brassica Napus, L.), Indian Mustard (Brassica juncea Czern.) and Black nightshade (Solanum nigrum L). This soil was previously cultivated by artichoke and continually irrigated with untreated sewaged water from the 70th of last century and cultivated with different kind of trees and vegetables. The chemical characterization of the used soil with pH 8.67, EC 0.2 dse m-1, 2.6% OM, and the texture is sandy soil. The experiment consisted of 24 plots (3m x 3.5m =10.5 m = 1/400 fed. with 5 ridges (70cm) irrigated with inoculation only sewage effluent using furrow irrigation.

Thiobacillus

Acidithiobacillus Ferrooxidans: DSMZ medium 882 was used to isolate and grow Acidithiobacillus ferrooxidans.

Mycorrhyzal (AM) Conidia

AM fungi spores were extracted from soil by wet sieving and sucrose density gradient centrifugation [12]. Soil sample was placed in a plastic bag and air-dried (with the tops of the bags rolled 2mm mesh and stored at 4°C. The procedure included passage of 25 g of air-dried soil or 30 cm of harvested trap culture substrate through 1,000-, 500-, 125- and 32-µm sieves. The 1,000-µm sieves were checked for spores adjacent to or inside roots, while the 500-µm sieves was checked for larger spores, spore clusters and sporocarps. The contents of the 125- and 32-µm sieves were layered onto a water-sucrose solution (70% wt/vol) gradient and centrifuged at 900 µg for 2 min. The resulting supernatant was passed through the 32-µm sieves, washed with tap water and used to inoculate the sewage soil. Mycorrhyzal is not specific in terms of the partner plant they choose, which means that the same fungus could be grown on a down) for 24 hours under cover, then brushed through a large number of plant species.

Microbial Cultivation and Fortification: All microorganisms used in the bioremediation trails were grown in Bioflo and Celligen fermentor/bioreactor, each in suspension was impregnated on a proper mordant at the rate of 20 ml microbial suspension per 100 gm mordant oven dried soil. Sewage soils were solely inoculated with a single microorganism at the rate of 100 gm impregnated mordant/400 gm soil.

Instrumentation and analysis of PTEs

Flame atomic absorption spectrometry (FAAS) is a simple and well available technique frequently used in determining PTEs in natural aquatic samples. A Perkin–Elmer flame atomic absorption spectrometer (FAAS) and HACH DR890 colorimeter was used in this study. Atomic absorption measurements were carried out using air: acetylene flame while HACH colorimeter measurement with the provided test kits. The operating parameters for working elements were set of as recommended by the manufacturer.

Soil treatments

For the management of the experiment, in randomly complete plot design, the selected farm treated with probentonite 1.25 ton/ feddan, thiobacillus thiooxidant, Mycorrhyzal (AM) conidia, and mixture of these treatments. The abovementioned treatments were well mixed with the studied sandy soil and three replicates were took place for each treatments.

Heavy metals distribution

In the studied samples heavy metals distributions were conducted according to [13]. The following fractions were obtained: the water soluble, exchangeable, carbonate, Fe-Mn oxides, organic, and residual. The method could be summarized as follow:

A. Water-soluble: soil soluble, soil sample extracted with 15 ml of deionized water for 2 h.

B. Exchangeable: The residue from water-soluble fraction is extracted with 8 ml if 1M MgCl2 (pH7.0) for 1h.

C. Carbonate-Form: The residue from exchangeable fraction is extracted with 8 ml of 1 M NaOAc (adjusted to pH 5.0 with HOAc) for 5H.

D. Fe-Mn Oxides-Form: The residue from carbonate fraction is extracted with 0.04M NH2OH.HCL in 25% (v/v) HOAc at 96oCwith occasional agitation for 6H.

E. Organically form: The residue from Fe-Mn oxide fraction is extracted with 3 ml of 0.02M HNO3 and 5ml of 30% H2O2 (adjusted to pH 2 with HNO3). The mixture is heated to 85oC for 2h, with occasional agitation. A second 3-ml aliquot of 30% H2O2 (pH 2 with HNO3) is added and the mixture heated again to 85oC for 3h with intermittent agitation. After cooling, 5 ml of 3.2M

NH4OAc in 20% (v/v) HNO3 is added and the samples diluted to 20 ml and agitated continuously for 30 min.

F. Residual Fraction: The residues from organic fraction are digested using a HF-HCl/HNO3.

RESULTS AND DISCUSSION

Effect of chemical remediation treatments applied and phytoremediation plants used in distribution of PTE's in sewage soils.

Geochemical forms of heavy metals in soil affect their solubilities; therefore, directly influence their potential bioavailability [2, 3]. Thus, assessing environmental impacts of trace metals in soils by aqueous concentrations of trace metals is incomplete. In particular, it may be helpful to obtain information on the degree of bioavailability of these metals [5].

In Egypt, increasing awareness of the hazard that heavy metals can cause to the environment and to humans is presently pressuring society to comply with environmental regulations and develop management strategies to minimize their adverse impacts. These strategies, however, should be built according to distribution of these trace elements in soil system.

Existing guidelines or regulations for the assessment of heavy metal contamination of soil are based on total soil metal concentrations. However, it is generally recognized that total concentrations do not necessarily provide good information on the potential bioavailability or mobility of metals in soils.

Sequential fractionation techniques are being used increasingly to provide more useful assessments of soil heavy metal contamination than is possible with single extractions or total metal concentrations alone. In addition, heavy metals fractionation is a fairly widely used technique for understanding the mechanisms of heavy metals distribution in soils and to help assess bioavailability of trace metals in soils.

In pure system, [9] studied Pb distribution in three Egyptian soils to understand Pb reactions as affected by their chemical characteristics and to predict the bioavailability of Pb in theses soils. The data indicated that distribution of PTE's drastically influenced by soil type, type of clay minerals, the presence of CaCo3 and the concentrations of the PTE's in soil system. However, metals added as a salt do not undergo the same processes as those metals already present in the sewage effluents entering the treatment plant. For this reason the studies exists with simple metal salts is controversial, since there is no certainty that the added metals will be present in the same forms as those metals already present in the sludge. This is important because there is evidence that the bioavailability of metals added to soil as metal salts may differ from the bioavailability of metals established from sewage effluents affected soils [10,11].

Nickel distribution after phytoremediation and remediative materials applied

In this part, because the wide variations in concentration between different treatments, we used percent expression in comparing between different phytoremediation plants used to absorb PTE's and different remediative materials applied to minimize hazards of PTE's in soil system or to enhance phytoremediation technique applied. In evaluation the effect of individual phytoremediation plants used on distribution of Nickel in soil, data showed that after canola plantation, the readily available form RAF (the sum of available and exchangeable forms) value was 55% of total concentration found in soil, decreased to 19 and 22 % in mustard and black nightshade. This result perhaps could refer to root exudates characterization of this plant compared to others tested which directly led to increase the available form of studied pollutant in soil system. In contrast, the moderately available form (MAF) represented by the sum of carbonate and Fe-Mn oxide values were 14, 43 and 29% after canola, Mustard and black nightshade respectively. This result may declare the effect of root exudates in availability situation of different plant used the preferability of using canola in sewaged soils. The respective values of hardly available form (HAF) represented by the sum of organic and residual forms were 31, 38 and 49% of total concentrations found in treated soil. However, it should be mention that the organic form values in soil system after harvesting of different plant species were 9, 24 and 31% which represents the ability of canola to absorb organic form followed Indian mustard and for less extent black nightshade.

Using of AM inoculated to tested plants, led to increase the RAF in all plants used compared to control treatments (only phytoremediation plants), for example, AM treatment increased RAF from 19 to 21% in mustard and from 22 to 25% in Black nightshade, a reverse trend however was observed in canola. Application of AM increased the MAF value from 14% to 36% and from 29 to 32 after canola and black nightshade, meanwhile it decreased from 43 to 38% in mustard. In other plants used as a phytoremediation technique however, inculcation of used plants increased all forms compared to control treatment (only phytoremediation plants). Inoculation of Black nightshade with AM decreased the HAF from 49% to 43%, a reverse trend, however, was observed in Canola and Mustard plants.

Data depicted in the same figure showed that application of thiobasillus applied in soil system almost take the same trend of AM. In Mustard Plant, application of thiobasillus increased the HAF from 38% in control to 42 % in treated soil. The same trend was observed in RAF by increasing the value from 19 to 21%. In Canola, data showed that MAF and HAF percentage significantly increased over control; meanwhile RAF was decreased to 29% compared to 55% in control. Thiobasillus assumed to be a material used as an oxidant material used for oxidate sulfur found in sewaged soil. The direct effect of such material declares through the decreasing the soil ph, according to the obtained results this trend was varied according to type of plant used through the increasing the RAF in Mustard and Black nightshade, however, a reverse trend observed in Canola.

The effect of probentonite and the mixture of all treatments showed the best managements in minimizing the availability of Ni in sewaged soils. Data showed that application of probentonite decreased the RAF in Canola to 9% against 55% in control. The respective values were 3 and 4% in Mustard and Black nightshade respectively. The effect of probentonite on Ni availability directly led to increase the hardly available form to 62% Canola against 31% in control and the same trend was observed in other hyperaccumulator plants used in this study. For the mixture treatment applied in this work, the best treatment, data showed that most of the studied pollutant retained in HAF regardless the type of Plant used. Different mechanisms take place will be documented in the discussion of this work.

Copper distribution after phytoremediation and remediative materials applied

Like Zn, distribution of Cu varied according to type of remediation used i.e. in phytoremediation plants used individually in sewaged soils, data in figure 6 showed that Mustard absorb the highest amounts of Cu compared to other plants used, this results showed through the decreasing percentage of RAF (sum of WS and Exchangeable forms). Application of probentonite and mixture treatments significantly decreased the RAF and increased of HAF especially residual one. For example, under mustard plantation data showed that application of mixture of all treatments decreased the RAF form 14% to 5%, meanwhile, the residual form increased from 48% to 66%, the same trend was observed in both application of probentonite under Mustard and other plants used.

The percentage of moderately available forms (MAF) represented in the same figures indicated that all treatments applied under Canola plant led to significant increase of these forms. Data showed that in probentonite treatment, the percentage of MAF was 37%, the respective value for sewaged soil planted with canola only was 16%, and worth to mention that all other treatments took the same trend. On the other hand, for the same plant led to decrease RAF and increased the HAF. The mixture treatment, for example, the RAF decreased from 55% to 23%, in contrast the HAF increased from 29 to 49%. Except bentonite in mustard and probentonite and mixture treatments in Black nightshade, the same trend was found.

In general it could be seen more clearly that Copper distribution after applying treatments depend on type of plants used as phytoremediation treatment, type of chemical treatment represented by probentonite or biological treatment represented by thiobasillus and Mycorohyza (AM) treatments applied, in all cases, however, of all treatments the mixture one was the best in having highest residual form, the aim of applying the remediative materials.

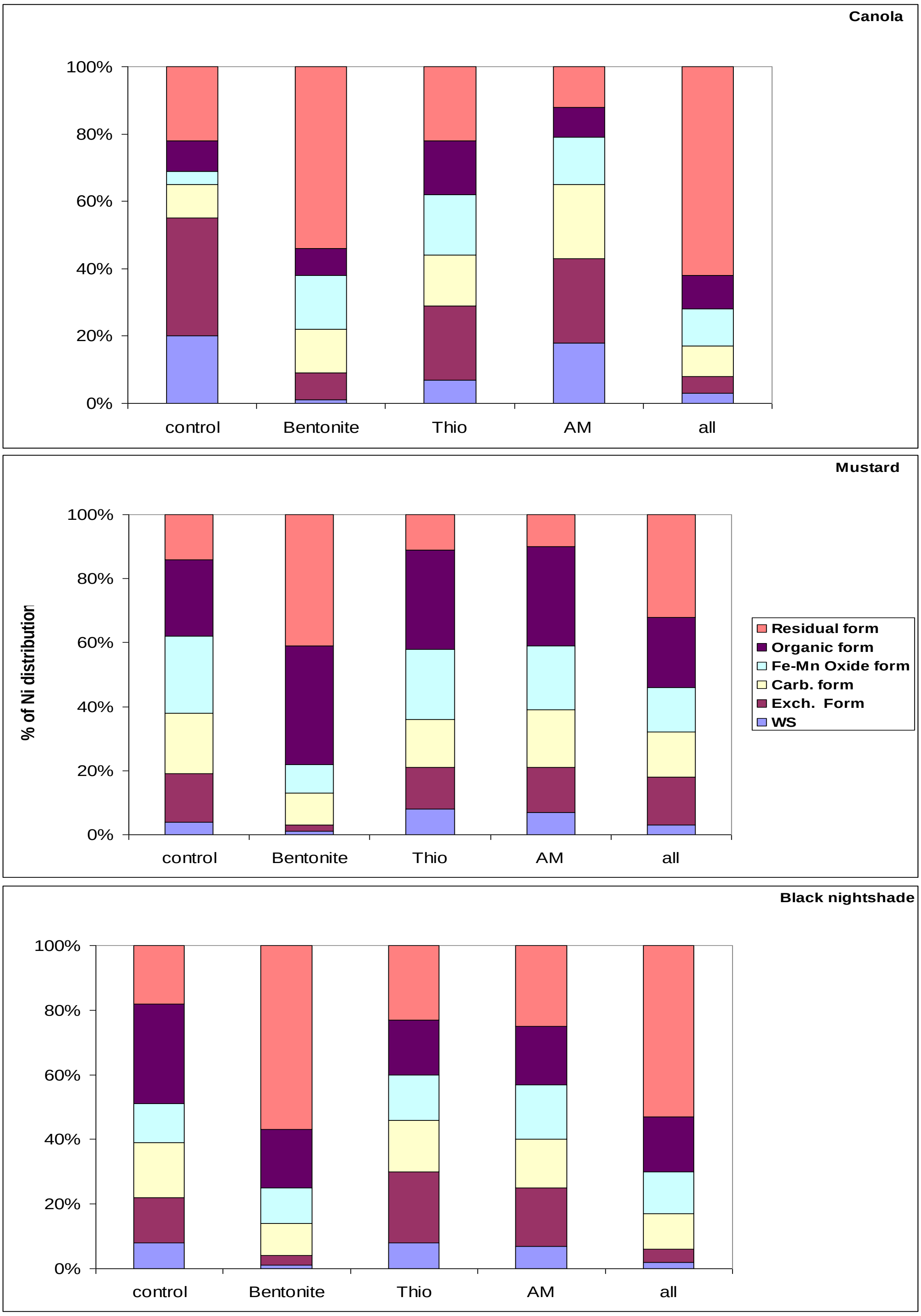

Figure 5. Nickel distribution in remediated soils as affected by type of phytoremediation plants used and remediation materials applied in sewage soils.

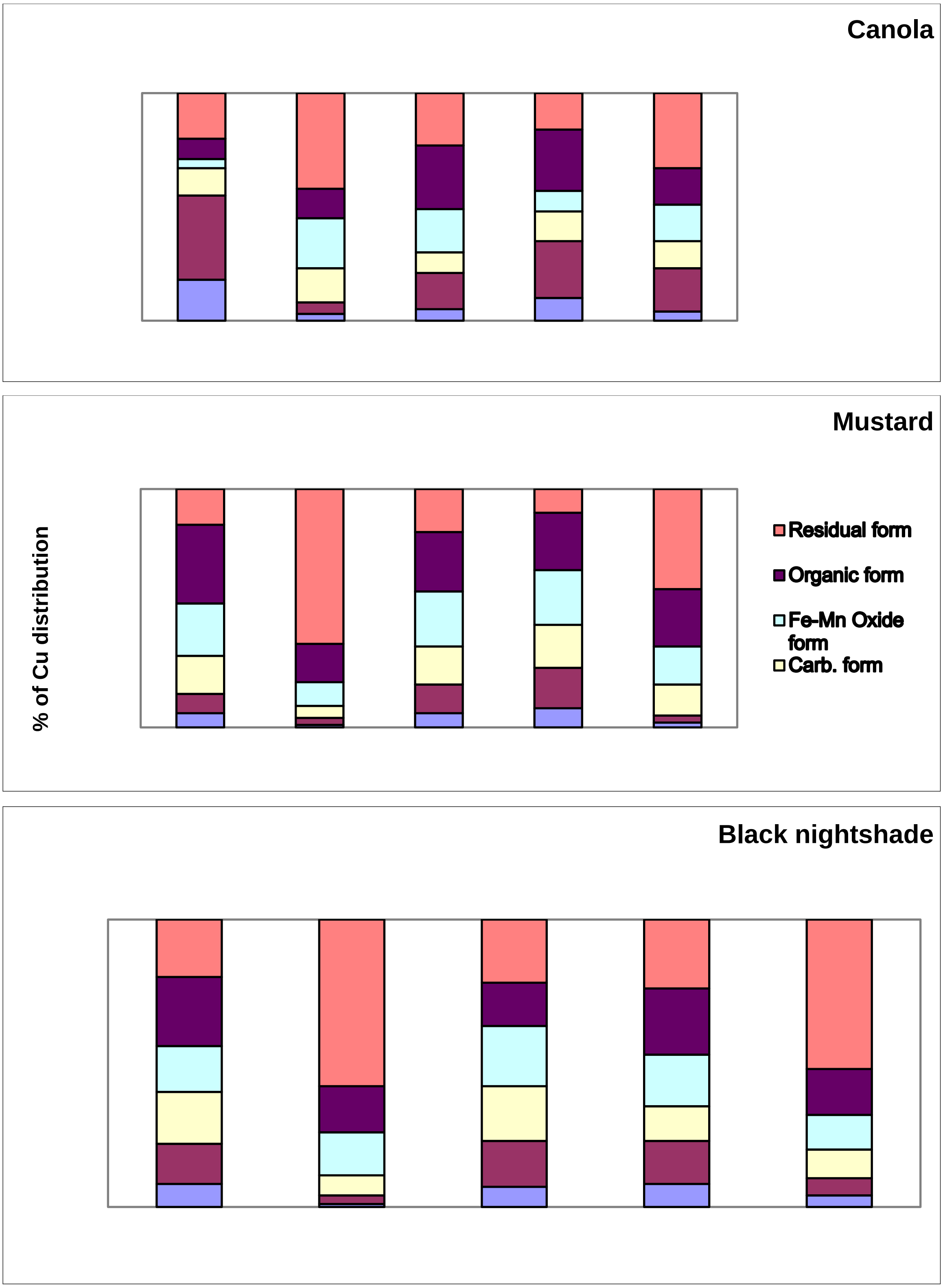

Figure 6. Copper distribution as affected by type of type of phytoremediation plants used and remediation materials applied in sewage soils.

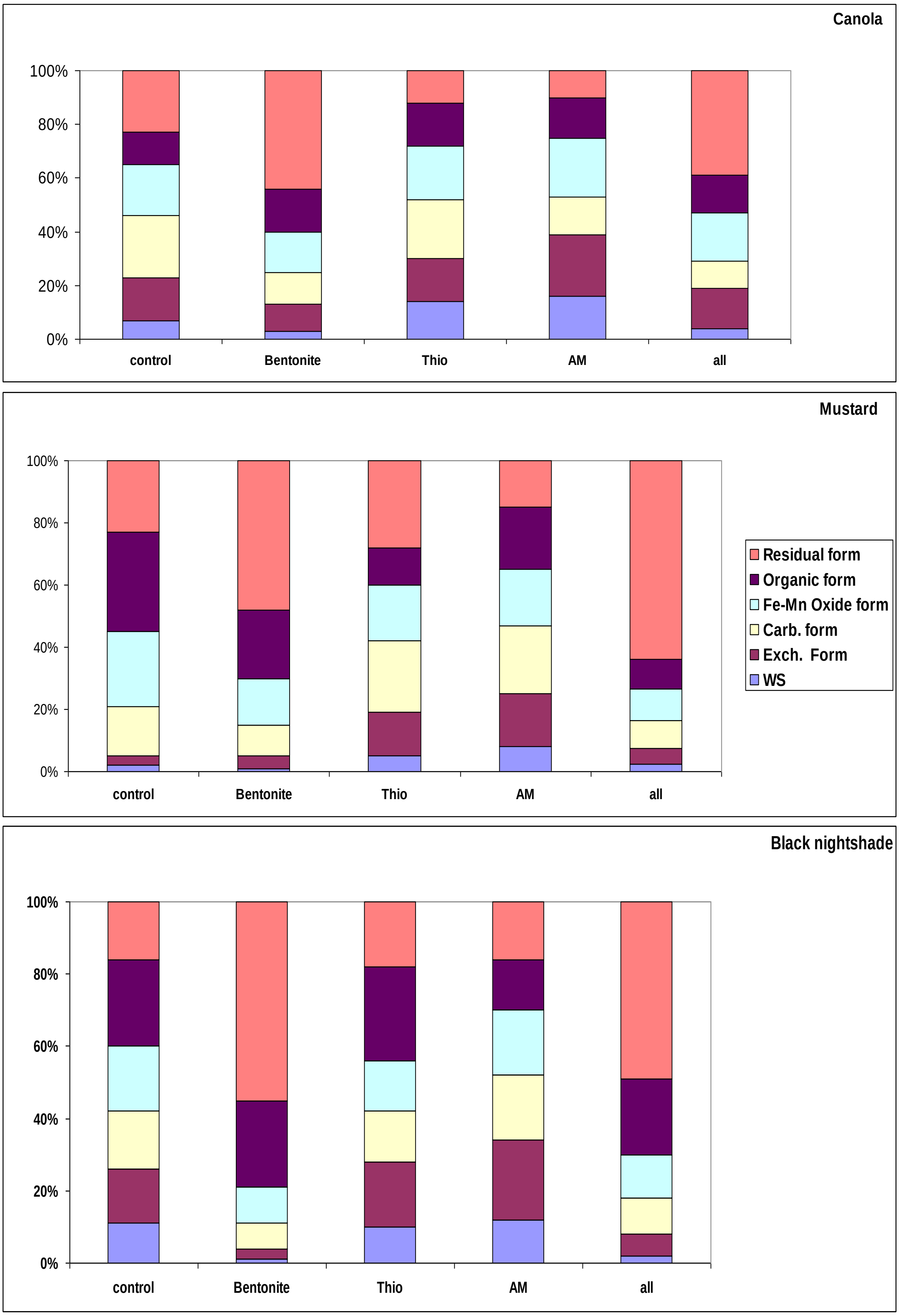

Figure 7. Zinc distribution as affected by integrated management applied in sewage soils.

Zinc distribution in sewaged soil as affected by remediative materials.

Zinc was fractionated in different gateways according to type of treatment applied. Data in figure 7 showed that the individual application of phytoremediation plant led to increase MAF in canola to reach 42%, the respective values for HAF and RAF were 35 and 23%. The application of AM and thiobasillus almost gave the same trend of having increase in this form. The application of probentonite and mixture of all treatment only decreased this form i.e. MFA against the increase of HAF to reach 60 and 53 % for probentonite and mixture treatment.

Bioremediation strategy applied in this work was set in three main associative actions, starting with sulfuric acid release in the soil ecosystem as a result of the action of inoculated *Thiobacillus sp* on added elemental sulfur in the presence of special remediation solution, followed by lowering the availability of PTE's in the soil ecosystem by proprobentonite which made complexes compounds with pollutants studied next to the action of phosphate released from rock phosphate by phosphate dissolving bacteria, AM and sulfuric acid. All Remediative treatments significantly influenced pollutants distribution in the soil ecosystem. For example, probentonite significantly increased the residual form, while application of *Thiobacillus sp* enhanced and increase the readily available forms of pollutants, In addition, results imply that cultivation of canoloa plants in contaminated soil showed high capability to minimize readily available forms (water soluble and exchangeable forms) compared to moderately or hardly available forms.

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