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EFFECT OF GLUCOSE AND ACETIC ACID ON Ni, Pb AND Zn TRANSFORMATIONS IN CONTAMINATED SOIL

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Organic acids and sugars are essential constituents of root exudates and have the potential to alter solubility and bioavailability of trace metals. Present study was conducted to evaluate the effect of exogenous application of glucose and acetic acid on the extractability of nickel (Ni), zinc (Zn) and lead (Pb) from contaminated soil. Soil was contaminated by adding metals and then equilibrated at field capacity for 35 days. Thereafter soil was amended with glucose and acetic acid and incubated at room temperature and field capacity for 30 days. Soil samples were taken after 5, 10, 20 and 30 days and analyzed for AB-DTPA extractable metals. Results showed that glucose and acetic acid initially enhanced the AB-DTPA extractable Ni and Zn compared to that with unamended soil, but decreased with time. In case of Pb, glucose enhanced its extractability. On the other hand, at 5, 10, 20 and 30 days of incubation, extractability of Pb decreased with acetic acid application. It was concluded that easily decomposable organic acids may temporarily enhance the metal availability to plants whereas that of Pb could decrease through precipitation of Pb as sparingly soluble compounds like $PbSO_4$ and $PbCO_3$. However further research is needed to know the mechanism of Pb retention in calcareous soils upon acetic acid application.

Keywords: Contaminated soils, metals, transformations, glucose, acetic acid

INTRODUCTION

Concerns over the effects of heavy metals on ecosystem health have increased during the recent years. Soils are considered an important ultimate sink for metals due to retention capacities. Therefore, bioavailability and potential toxicity of heavy metals depends upon soil properties as well as metal speciation. A plethora of factors such as organic acids in soils can affect the metal release from soils and speciation in soil solution (Mench and Martin, 1991). Organic acids like oxalic, tartaric, citric and malic acid form soluble complexes with metal ions thereby affecting metal bioavailability. Acetic acid is one of the most common and important intermediates in anaerobic decomposition of organic matter. Other acids such as succinic, lactic, butyric and propionic acids are also produced during anaerobic decomposition of OM but their concentrations are much less than acetic acid (Lindsay, 1979; Gramss *et al.*, 2004). Organic ligands (citrate, malate, acetate) at relatively low concentration ($\leq 10^{-3}$ mol L^{-1}) inhibited Pb desorption but enhanced its desorption at higher concentrations (Yang *et al.*, 2006). Less Pb, Cu and Cd were desorbed with malic and acetic acid at pH 3.1 compared to that with citric acid at pH 7, indicating that pH was not dominant factor governing the release of physically adsorbed metals (Qin *et al.*, 2004).

Most of the research work done pertains to soils contaminated with the application of sewage sludge and soils affected by mining and smelting activities or sewage irrigation. Little information regarding bioavailability of metals in soils amended with glucose and acetic acid are available. Owing to contrasting chemical properties of organic acids and sugars, these were selected to evaluate their effect on the AB-DTPA extractability and thus bioavailability of Ni, Zn and Pb at different times of incubation from contaminated soils.

MATERIALS AND METHODS

The experiment was conducted in Soil and Water Chemistry Laboratory, Institute of Soil and Environmental Sciences, University of Agriculture, Faisalabad, Pakistan during 2006. Bulk soil samples were collected from plow layer of uncontaminated field receiving only canal water irrigation and rainfall. Soil was air-dried and ground to pass through 2 mm sieve. Soil pH_s, EC_e, and saturation percentage were determined by the method described by US Salinity Lab. Staff (1954). Calcium carbonate and organic matter were determined by the methods described by Moodie *et al.* (1959) and Walkley and Black (Jackson, 1962), respectively. For particle size analysis, hydrometer method was used (Bouyoucos, 1962). Soil was also analyzed for AB-DTPA extractable (Soltanpour, 1985) and total metals (Amachar, 1996). Soil was calcareous loamy with low organic matter content (Table 1). Metals viz. Ni, Zn and Pb were added to soil @ 0, 100, 200, 400 mg kg^{-1} of dry soil as their chloride salts.

Table 1. Physical and chemical characteristics of soil used in the study

Soil characteristic	Unit	Value
pH_s	-	7.72
EC_e	$dS\ m^{-1}$	2.41
Organic matter	%	0.58
$CaCO_3$	%	5.47
Clay	%	24.0
Saturation	%	34.4
Textural class	-	Loam
AB-DTPA extractable metals		
Ni	$mg\ kg^{-1}$	0.33
Zn	"	3.01
Pb	"	2.91
Total metals		
Ni	$mg\ kg^{-1}$	49.88
Zn	"	56.01
Pb	"	32.69

After mixing of metal salts, soil was placed in plastic jars of one kg capacity. Soil was equilibrated in greenhouse at field capacity (moisture content approximately equivalent to 50% of the saturation percentage of soil) for 35 days. At the expiry of equilibration period, soil was air-dried and sieved again.

Analytical grade glucose and acetic acid amendments were added @ 2.40 and 40.0 g glucose per kg soil, while concentrated acetic acid was used @ 10 mL kg^{-1} of soil. Glucose and acetic acid were added to soil in solution form. A similar set of treatments but without amendments was kept for comparison. Pots were arranged in completely randomized design and lids of pots were kept loose to allow gaseous exchange and avoid build up of CO_2 in pots.

The pots were incubated at room temperature (≈25 °C) and moisture contents were maintained at field capacity throughout the experimentation and adjusted gravimetrically through the addition of distilled water. About 30 g of soil samples were taken at 5, 10, 20 and 30 days. These samples were extracted with AB-DTPA (Soltanpour, 1985) and analyzed with the help of atomic absorption spectrophotometer (Thermo S-series) for metal concentration. Data pertaining to AB-DTPA extractable metals were subject to statistical analysis by computer based statistical package MSTATC.

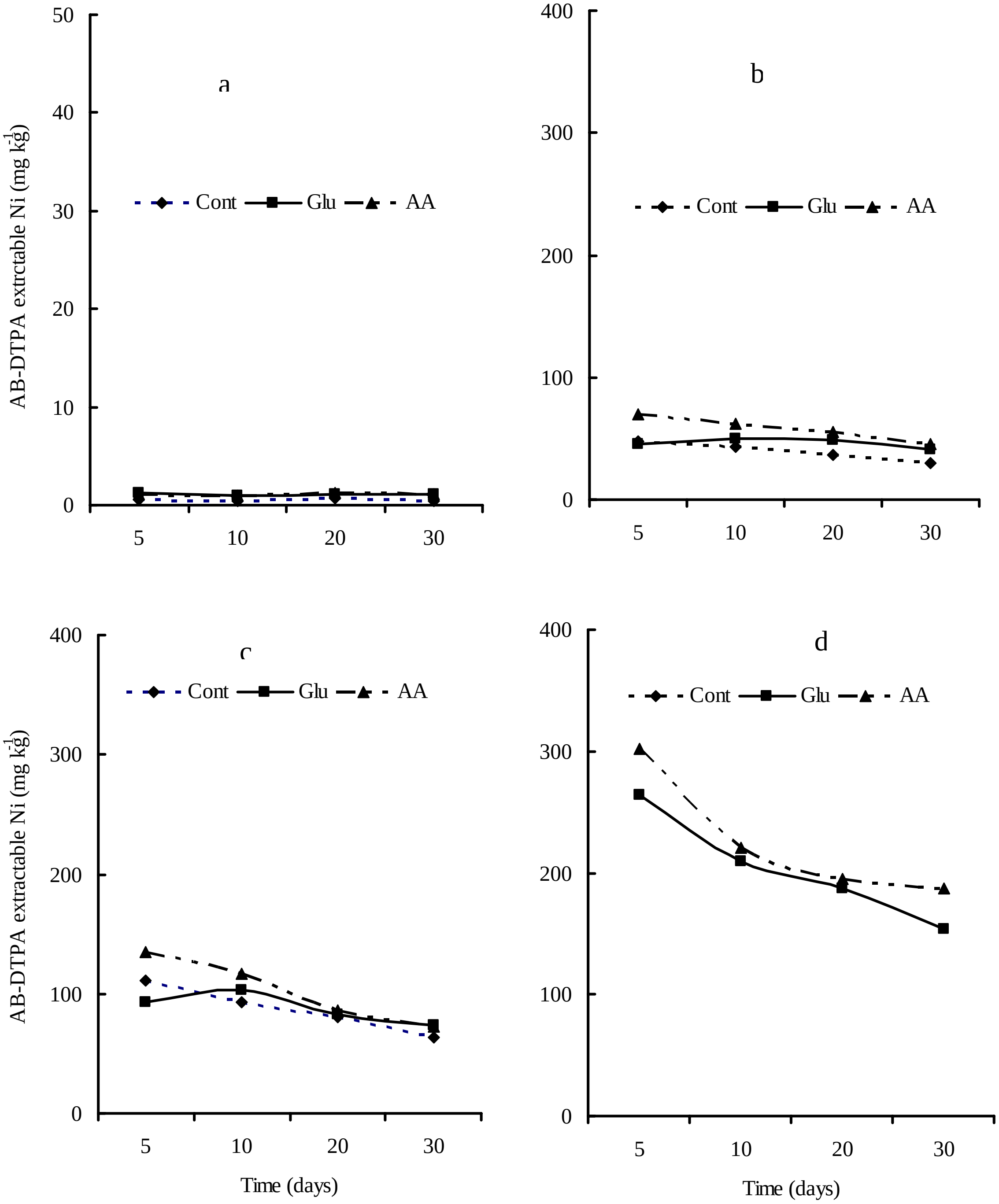
RESULTS AND DISCUSSION

At low concentration of Ni in soil (native level), the effects of amendments was not contrasting (Fig. 1). At 100, 200 and 400 mg Ni kg^{-1} soil, effect of amendments is obvious. In contaminated soil without any amendment, Ni gradually decreased from day 5 to day 30 and maximum decrease (29.5%) was recorded at the lowest Ni application level of 100 mg kg^{-1}. In soil amended with glucose, available Ni increased from day 5 to day 10 in soil contaminated with 100 and 200 mg Ni kg^{-1}; thereafter decreased at days 20 and 30. This initial increase in Ni availability might be due to microbial conversion of glucose into organic acids particularly lactic, acetic and n-butyric acid, which possibly caused fluctuation in pH as reported by Gramss *et al.* (2003) and thus available Ni. Decrease in Ni at days 20 to 30 might be the result of formation of stable metal compounds and decomposition of organic acids affecting an increase in soil pH, which in turn enhanced metal retention in soil. Misra and Pande (1974) observed that out of the added 100 mg Ni kg^{-1}, only 14.1 mg Ni kg^{-1} was recovered with AB-DTPA extraction after 80 days from glucose amended soil.

Glucose can undergo both oxidative and reductive reactions and has tremendous potential for CH_3COO^- (acetate) formation. As there is no transfer of electrons in changing glucose to acetate, the Redox Potential does not affect this transformation. However, $C_2O_4^{2-}$ (oxalate) and $CHOO^-$ (formate) are products of glucose oxidation. Other oxidation reaction products include carbon dioxide, oxalate, formate and carbon monoxide (Lindsay, 1979) which would decrease pH to alter the availability of metals including Ni.

In contrast to un-amended and glucose amended soil, Ni was higher throughout the study period with the addition of 10 mL kg^{-1} of acetic acid. Presence of similar level of extractable Ni in glucose and acetic acid amended soil at day 30 may be the result of elimination of pH effect through reaction with lime and formation of stable compounds like nickel carbonates and co-precipitation of Ni with Fe and Mn oxides (Kabata-Pendias and Pendias, 2001).

Different processes operate at higher level of metal contamination for the conversion of AB-DTPA extractable metals into non-recoverable forms, but precipitation reactions generally dominate over sorption. One reason could be the toxicity of applied metals to microbes resultantly slower and less conversion of organic amendment into stable organic products (Vasquez-Murrieta *et al.*, 2006). Precipitation is another mechanism of sequestering Ni if the precipitated phase is relatively stable (Brown Jr. *et al.*, 1999). Calcite deposition could also result in co-precipitation of Ni. Moreover, reactions of Ni with clay particles and hydroxide may increase with an increase in pH_s of acetic acid amended soil.



(Incubation time)

Fig. 1. Extractability of Ni as affected by time and application of amendments (Cont. – control , Glu – Glucose, AA – acetic acid).
a. control; b. Ni @ 100 mg kg^{-1}; c. Ni @ 200 mg kg^{-1}; d. Ni @ 400 mg kg^{-1}.

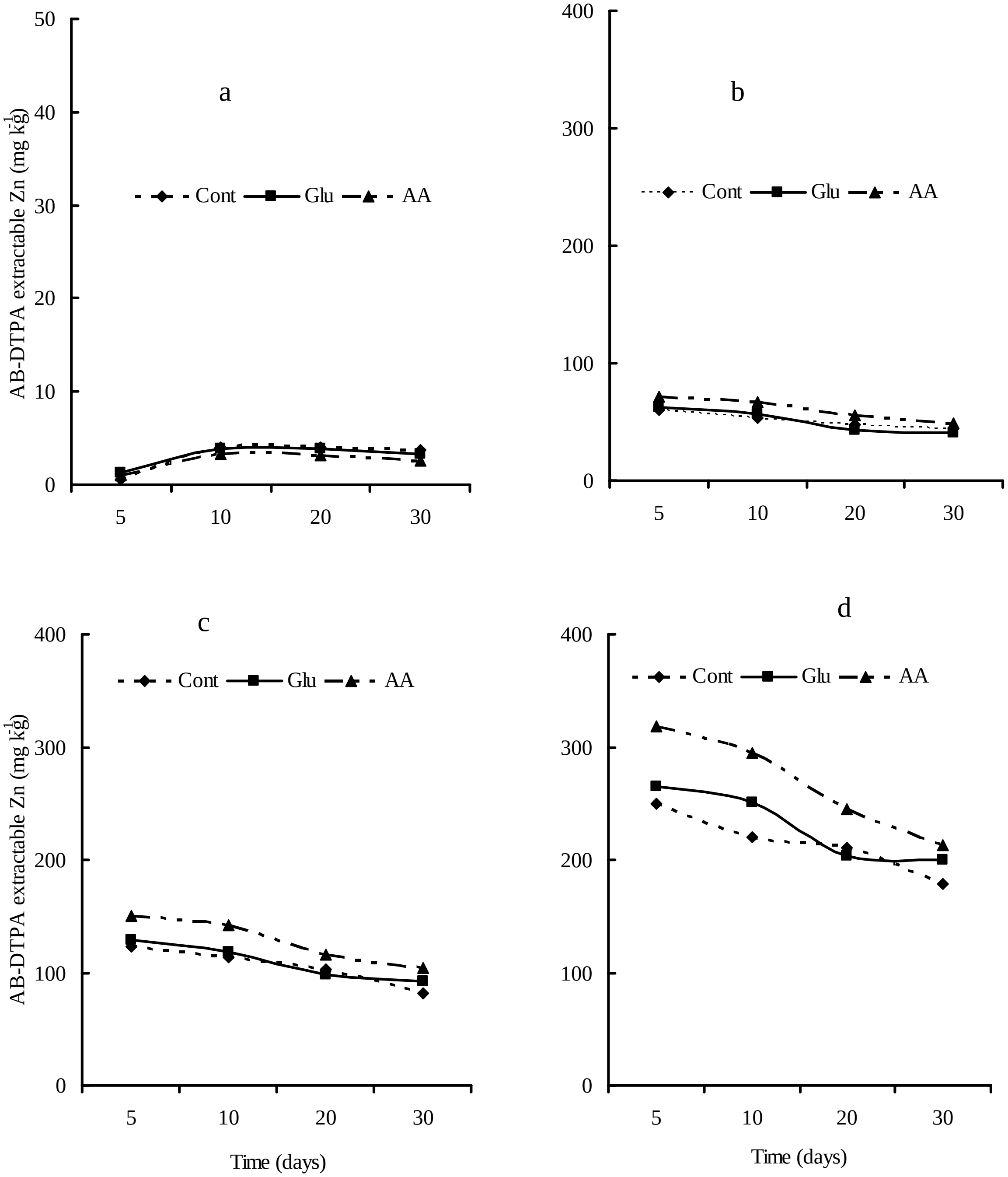


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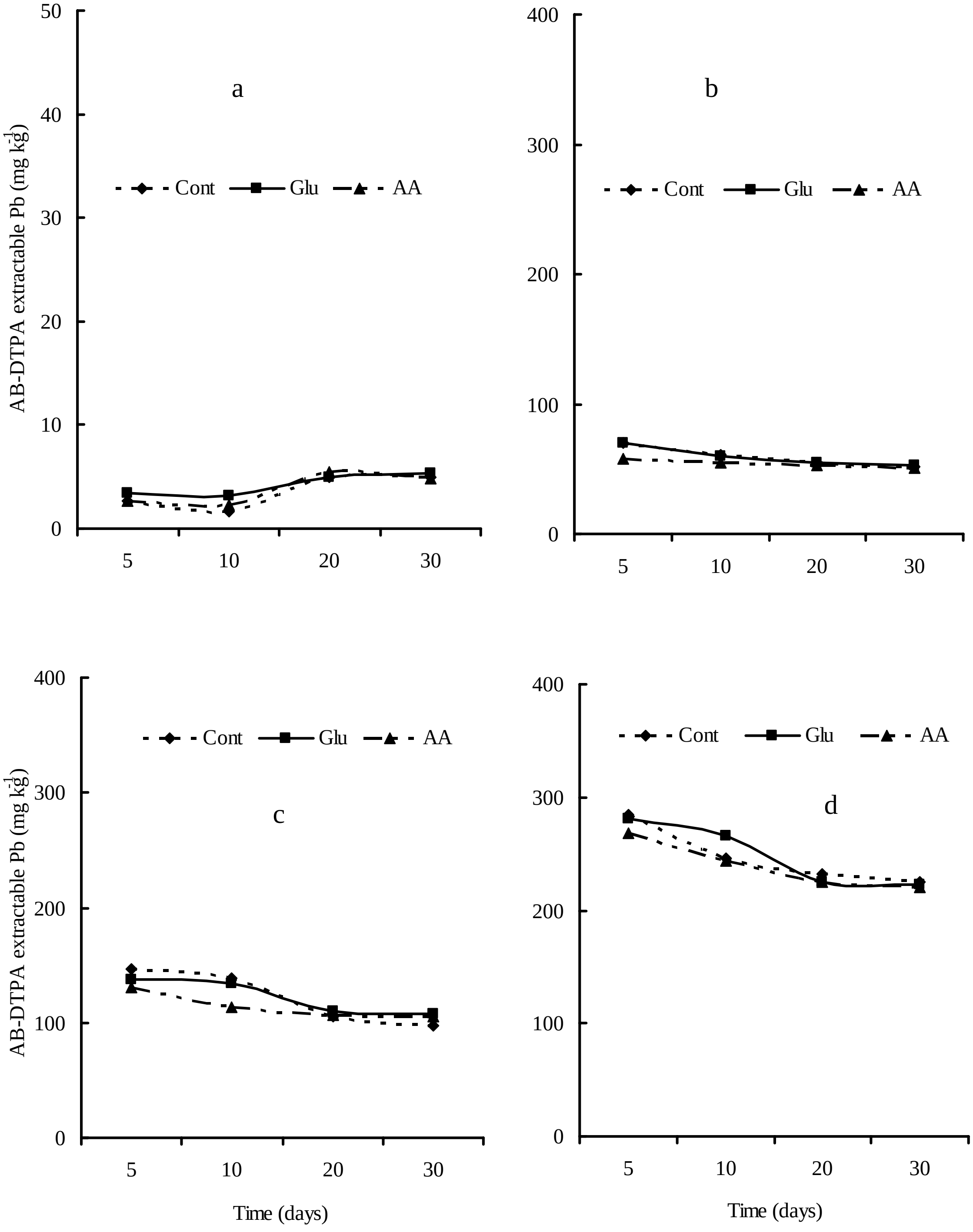
Fig. 2. Extractability of Zn as affected by time and application of amendments (Cont. – control , Glu – Glucose, AA – acetic acid)
a. control; b. Zn @ 100 mg kg^{-1}; c. Zn @ 200 mg kg^{-1}; d. Zn @ 400 mg kg^{-1}.



(Incubation time)

Fig. 3. Extractability of Pb as affected by time and application of amendments (Cont. – control, Glu – Glucose, AA – acetic acid).
a. control; b. Pb @ 100 mg kg^{-1}; c. Pb @ 200 mg kg^{-1}; d. Pbi @ 400 mg kg^{-1}.

As soil was calcareous ($CaCO_3$ = 5.47%), there is strong possibility of dissolution of native lime upon the addition of acetic acid along with production of organic acids upon glucose decomposition. The sorption kinetics and the adsorption-desorption isotherms has indicated that in a calcareous soil, Ni was retained by carbonates (Businelli *et al.*, 2004). Along with insolublization process as hydroxide salts caused by high pH induced retention through formation of strong complexes, there could be Ni coprecipitation involving formation of Ni-Ca carbonate double salt or mixed Ni/Al hydroxides and carbonate formation (Businelli *et al.*, 2004). The Ni retention in soil could be through the formation of inner sphere complexes while clay sorption mechanism is totally overshadowed by the double effect of Ni precipitation and complex formation caused by $CaCO_3$.

Available Zn increased from day 5 to day 10 in uncontaminated soil with or without amendment, which decreased slightly at day 20 to day 30, but concentration remained higher than that at day 5 (Fig. 2). In soil contaminated with 100 and 200 mg Zn kg^{-1}, available Zn was almost similar in control and glucose amended soil. At 400 mg kg^{-1} Zn addition, decrease in Zn from day 20 to day 30 was less in glucose-amended soil. In soil contaminated with 100 mg kg^{-1}, there was less Zn at day 30 compared to that for control and acetic acid application. It was higher at higher rates of Zn application of 200 and 400 mg kg^{-1}. With acetic acid, Zn concentration was higher than that for the control and glucose at all the sampling times and Zn application levels. However, Zn decreased with time in acetic acid amended soil. Redistribution of Zn among different soil solid phases because of more solubilization and subsequent re-precipitation may be responsible for the observed decrease in Zn. Overall, Zn remained generally higher with acetic acid and glucose amendments than un-amended contaminated soils.

Two processes could be responsible for mobilization and higher extractability of Zn especially that of native Zn (Fig. 2a) upon glucose and acetic acid application. First, water was added to dry soil, it resulted in the solubilization and mobilization of the added Zn. Secondly, added amendments generated acidity to help bring more Zn into labile form. Addition of 10% sucrose (w/w) and casein 15.6% (w/w) produced a suite of organic acids (lactic, acetic and n-butyric acids, etc.) resulting in a decrease in soil pH and thus mobilization of metals including Zn (Gramss *et al.*, 2003). However, the effect of pH could be considered temporary in alkaline soils because as the decomposition of low molecular weight organic acids proceeds, soil could return to original pH (Gramss *et al.*, 2004). Alkalinity could, therefore result from H^+ consuming decarboxylation of organic acids, if these are present as carboxylates instead of undissociated acids (Barekzai and Mengel, 1993). Moreover, degradation of dissociated carboxylic acids by oxidants is a source of OH^- as per the following reaction (Gramss *et al.*, 2004):

$$CH_2(COO^-) + 2O_2 \xrightarrow{\qquad\qquad} 3CO_2 + 2OH$$

Gramss *et al.* (2003) noticed that during the phase of acid formation, solution concentration of Zn increased 57-fold owing to a decrease in pH.

At higher rate of Zn application, lack of any influence of added glucose could be the result of Zn toxicity to microbes decomposing organic materials (Rost *et al.*, 2001). Zinc in soils at elevated levels is considered more toxic to microbes than other metals (Moffett *et al.*, 2003). Muhlbachova (2002) found maximum increase in DTPA-extractable Zn in arable soil two days after the addition of glucose equivalent to 1000 µg C g^{-1} soil and thereafter Zn decreased. In 48 days incubation period, transformation of applied Zn to Fe- and Mn-oxides and organically bound forms was enhanced with the application of 1% starch (Phogat *et al.*, 1994).

Extractability of native Pb increased with time both with and without amendments (Fig. 3). At day 20, native Pb was higher with acetic acid while at day 30 with glucose. However, in Pb receiving soil, available Pb concentration gradually decreased from day 5 to day 30. Maximum reduction was recorded at the lowest Pb application rate of 100 mg kg^{-1}. Acetic acid immobilized Pb more than that with glucose and extractability of Pb was the highest in un-amended soil at all the Pb application rates (100, 200 and 400 mg kg^{-1}). At the end of experiment, Pb concentration was the highest in soil amended with acetic acid @ 100 and 400 mg kg^{-1} of Pb, whereas its concentration was the lowest in un-amended soil at 100 mg Pb kg^{-1} level.

Generally, precipitation reactions could effectively remove metals from soil solution through the formation of new solid phases, usually in association with anions or cations already present in soil solution. On the other hand, changes in soil moisture contents (indicating the Eh) may bring metals from solid to solution phase as has been observed in case of Pb in uncontaminated soil where Pb increased with incubation time (Fig. 3a). Muhlbachova (2002) reported that in glucose amended soil, DTPA extractable Pb was higher at day 2 than that at day 10. Initial increase in Pb could be due to formation of low molecular weight organic acids from glucose and a decrease in pH. As the decomposition advanced, it might caused elimination of acids from soil and stabilize soil pH (Charlatchka and Cambier, 2000), resulting in decreased availability of Pb.

Although precipitation and adsorption could exhibit time-dependent reaction rates, yet, precipitation reactions could become more important at higher Pb application level. The Pb availability could decrease as a result of dissolution and re-synthesis of solid phase, thereby removing Pb from the bio-available pool (McLaughlin, 2001). In many cases, Pb-contaminated soils contain minerals like anglesite ($PbSO_4$), cerrussite ($PbCO_3$) and various Pb-oxides as well as Pb(II) adsorbed onto oxides of Fe and Mn (Traina and Laperche, 1999). Ghafoor *et al.* (1999) reported formation of $PbCO_3$ and $Pb(OH)_2$ from added Pb into soil having pH 7.58 and 2.60% lime. Lindsay (1979) reported that $PbSO_4$ (anglesite) is the most stable compound below pH 6, whereas $PbCO_3$ (cerrussite) is the most stable at alkaline pH. Lu *et al.* (2005) reported that in alkaline soils, most of the applied Pb is usually retained in Fe-Mn oxide fraction of soil. With the passage of time, more of the Pb was transformed from exchangeable fraction to Fe/Mn oxide and organic matter fraction and less of the applied Pb was retained in carbonate fraction of soil.

At all the levels of Pb addition, decrease in extractable Pb with acetic acid compared to that for the control may be the result of modification of soil sorption characteristics. As demonstrated by Gramss *et al.* (2004), citric acid application to calcareous soil yielded alkalinity upon the decomposition of acid to CO_2. Under well-oxidized conditions, acetate was converted to CO_2 through other intermediates like $C_2O_4^-$, $HCOO^-$ and CO (Lindsay, 1979). Qin *et al.* (2004) reported that amount of Pb desorbed from soil upon the addition of acetic acid was many times less than that with citric or malic acid at pH 7, but was more than that with $CaCl_2$ or $NaNO_3$. Garcia-Delgado *et al.* (1996) reported that in soils with high lime contents, the added acetic acid may react with calcium carbonate and little of Pb is mobilized from soil. It was observed that acetic acid of pH 4.0 did not result in a significant Pb mobilization and pH also remained the same for the duration of experiment (24 hour). However, in our study, pH of original soil was 7.72 and after 24 hour of acetic acid addition, it decreased to 5.60 and on the fifth day again increased to 6.42. As this pH was below the initial soil pH, yet less amount of Pb extracted from acetic acid receiving soil than the unamended Pb receiving soil. The actual mechanism of this increased lead retention in acetic acid amended soil is not yet known. This aspect needs further investigation to elucidate the mechanism responsible for the present results.

CONCLUSIONS

In uncontaminated calcareous alkaline soil, addition of glucose or acetic acid resulted in mobilization of native Ni, Zn and Pb. At day 30, concentration of AB-DTPA extractable metals (Ni, Zn, Pb) was higher than the respective initial levels in soil. In contaminated soil, application of acetic acid resulted in increased metal extractability with AB-DTPA. Behavior of glucose and acetic acid was also similar, i.e. temporary mobilization of these metals. In contaminated soil without amendment, there was a gradual decrease in available Ni, Zn and Pb. Acetic acid application resulted in decreased extractability compared to un-amended or glucose amended soil. It is very difficult to predict the behavior of organic amendments with respect to Ni, Zn and Pb dynamics; as it is affected by the nature of metals, metal contamination level, composition of soil and organic amendment itself. Multiple metal contamination, as is the case under field conditions, these amendments may have different role in affecting metal bioavailability. Although it is hypothesized that acetic acid induced $CaCO_3$ dissolution may have resulted in precipitation of Pb into the forms like $PbCO_3$, but further research is needed to establish the mechanism of decrease in Pb extractability upon acetic acid addition.

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