

## Immobilization of Ni and Zn in Soil by Cow and Chicken Manure

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### Abstract

This study was performed to evaluate the effect of both cow and chicken manures application on the immobilization of Ni and Zn in soils. Effect of cow and chicken manures on the bulk density, pH and the distribution of Ni and Zn in various chemical fractions of an alkaline, sandy loam soil was explored in a PVC columns study. Cow and chicken manures were mixed at rates of 10, 20 and 30 g/kg of soil. The soil-manure mixture was incubated for two months at room temperature. Sequential extraction procedure was performed on all samples from each column to determine Zn and Ni in different fractions (soluble-exchangeable, organic, carbonates and residual). Results obtained showed a decrease in soil bulk density with an increase of 0.3 units in soil pH as compared to the control. After 60 days of incubation, Ni concentrations were found to be 28 and 34% of inorganic fraction, while the residual reaction accounted for 58 and 53% for cow and chicken manure respectively as compared to the corresponding control. In case of Zn, soil organic matter fraction accounted for 53-57% of the total Zn. The soluble and exchangeable fraction which, although, slightly increased with time remained very low (2-4%) for the two metals. Therefore, the addition of the manures resulted in improved soil bulk density and showed a good potential in immobilizing both two metals in the studied soil.

**Keywords:** Soil remediation; Immobilization; Organic manures; Metal fractionation; Heavy metals

### Introduction

In terrestrial ecosystems, the soil is the major sink of chemical contaminants. Anthropogenic activities, mainly resulting from industrial production, manufacturing and the disposal of domestic and industrial waste are the major sources of metal (loid)s input in soils. Unlike organic contaminants, metal (loid)s do not undergo microbial or chemical degradation, and their total concentration persists over extended periods after their incorporation in soils [1]. With greater public awareness of the implications of contaminated soils on human and animal health there has been increasing interest amongst the scientific community in the development of technologies to remediate contaminated sites [2].

Although the rehabilitation of the contaminated soils is imperative, the use of most conventional remediation strategies, such as soil removal through excavation and landfilling with clean soil are unfeasible economically apart from being environmentally disruptive. Keeping in view these concerns, the investigations have been made to explore other environmentally sustainable alternatives for soil remediation which should have better cost benefit ratio.

Among these technologies, in situ immobilization of metals has emerged as a promising option and it has been the focus of various research investigations for soil remediation [3,4]. By this approach metal immobilizing amendments have been employed to inactivate different metals thus alleviating the risk of groundwater contamination, plant uptake and exposure of other living organisms [5]. Application of organic amendments to agricultural soils can be beneficial because they can provide nitrogen, phosphorus, and other nutrients; improve the structure of degraded soils; beneficial organic matter is increased; and soil application offers a reasonable means for waste disposal. Organic amendments have been used as chemical barriers for heavy metal movement [6].

The mobility of trace metals, their bioavailability and related toxicity to plants strongly depend on their specific chemical forms or

the way that they are bound. Consequently, these factors, rather than the total elemental content, are the parameters that must be determined to assess toxic effects and study geochemical pathways [7,8].

Some organic materials traditionally used in agriculture, like animal manures, compost and peat, have been employed recently in different bioremediation experiments for soils contaminated with heavy metals [9,10]. These materials affect the speciation of metals through changes in soil chemical properties (pH, Eh, nutrient content, etc.) and by the metal chelating ability of their organic matter [11,12]. Also, the efficient use of these materials without damage to the environment is now considered to be of high priority. This is a way of recycling them apart from being precursors for improving soil fertility and its physical and chemical properties.

Addition of organic matter (OM) amendments, such as compost or manure, is an inexpensive and common practice to facilitate revegetation of contaminated soils. The effects of OM amendments on heavy metal bioavailability depend on the nature of the organic matter, and on the particular soil type and metals concerned [12], usually involving the formation of insoluble contaminant species, less likely to leach through the soil profile [13]. Organic amendments can decrease heavy metal bioavailability, shifting them from "plant available" forms to fractions associated with OM, carbonates or metal oxides [14], with consequent reductions in the metal uptake by the installed plants. When

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re-vegetation of contaminated soil is combined with addition of soil amendments such as organic manures, the mobility of contaminants in the soil can be further reduced [15,16].

Clemente et al. [12] reported that both fresh cow manure and compost having a high maturity degree amendments favored Zn and Pb fixation in soil from a former Pb–Zn mine area at La Union (Murcia, Spain). The main objective of the present research work was to explore the effect of organic matter on reducing the disruptive effect of heavy metal (loid)s, particularly Ni and Zn, on soil ecosystems.

Keeping in view the easy availability and abundance of chicken and cow manures in the region, it is interesting to study their effect on the fractionation of heavy metals such as Zn and Ni in soil. The aim of this work is to study the effect of cow and chicken manures on the bulk density, pH and the distribution of Ni and Zn in various chemical fractions of an alkaline, sandy loam soil. The specific objectives of this study were, therefore, to monitor changes, over a span of 60 days, in: 1) soil bulk density, and 2) soil pH and four chemical fractions of soil Zn and Ni as a function of time and manure source and thus assessing their ability to immobilize the metals under study in soil.

## Experimental

### Materials and methods

To fulfill the above objectives, outdoor incubation experiment was carried out in soil columns in split design with three replications. The main studied factors were two incubation times (IT) namely: one and two months. The sub-main included two types of manure (MT), cow and chicken manures. The sub-sub main were three application rates beside the control. The investigated application rates were 10, 20 and 30 g manure/kg soil. The Sandy loam soil was used in the experiment, chicken and cow manure samples were collected from the Experimental Research Station of King Abdulaziz University located at Hada El-Sham, Jeddah, Saudi Arabia. The sampled soil and manures were aurally dried, then grinded and passed through a 2 mm sieve to obtain homogenized particle size. 880 g of the above mentioned soil was thoroughly mixed with the cow manure and chicken manure according to the previous application rates. The soil-manure mixture was filled in PVC columns with a 10 cm radius and 35 cm length. The bulk density in the packed columns was adjusted to 1.5 g/cm<sup>3</sup>. The packed soil columns were flooded with tap water and then allowed to reach the field capacity by gravity. Then, moisture content was kept constant at this level by adding the deficient water every day during the course of the incubation period. Solution of 50 ml containing 10 mg kg<sup>-1</sup> Ni and 10 mg kg<sup>-1</sup> Zn in the form of NiCl<sub>2</sub>·6H<sub>2</sub>O and ZnCl<sub>2</sub>·X H<sub>2</sub>O respectively was applied to each column. The soil columns were placed at outdoors under the shade for incubation period of one and two months. The columns were sampled after the conclusion of first and second months respectively. Control soil columns were filled with the same soil and bulk density without the addition of organic manures.

### Soil analysis

**Soil bulk density:** At the end of each incubation period the weight, height and radius of the soil column were measured in order to calculate soil bulk density as follow:

$$V = \pi r^2 h$$

Where:-

$$V = \text{is volume in cm}^3,$$

$$r = \text{radius in cm}$$

h = height in cm

Soil bulk density (gcm<sup>-3</sup>) in each treatment was measured by dividing the dry mass of the soil sample by the volume obtained.

**Soil chemical analysis:** The soil was analyzed for general physical and chemical properties before the incubation period (Table 1). Soil pH and EC were measured in a 1:2 soil/water suspension using pH meter SensION PH3, Crison instruments, S.A and Jenway 4510 conductivity meter, Bibby scientific Ltd, respectively. Soil organic matter was analyzed by potassium dichromate oxidation and titration with ferrous ammonium sulphate. The hydrometer method was used for the particle size distribution. The soil samples were aurally dried at the end of first month and second month of incubation period. Sequential extraction procedure was performed on all samples from each column to measure Zn and Ni in different fractions after the application of cow and chicken manure according to McGrath and Cegarra (1992), which had the following steps:

1. 0.1 M CaCl<sub>2</sub> (1:10 w/v) for 16 h; metals in soil solution and in exchangeable forms.
2. 0.5 M NaOH (1:10 w/v) for 16 h followed by aqua regia digestion; metals associated with organic matter
3. 0.05 M Na<sub>2</sub>H<sub>2</sub>EDTA (1:10 w/v) for 1 h; metals mainly in the carbonate fraction.
4. Digestion with aqua regia; residual metals.

All metal concentrations were determined in the extract through DR 6000 spectrophotometer (Hach-Lange Inc.).

**Statistical analysis:** The data was statistically analyzed as a split-split plot design with three replicates by the analysis of variance (ANOVA) procedure after application of ANOVA assumptions. Then the least significant difference (LSD) at P < 0.05 was used to compare the treatment means according to the SAS procedure.

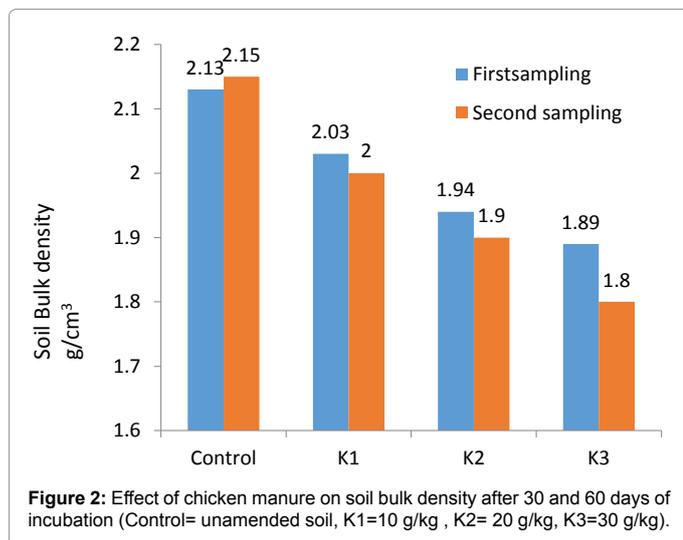
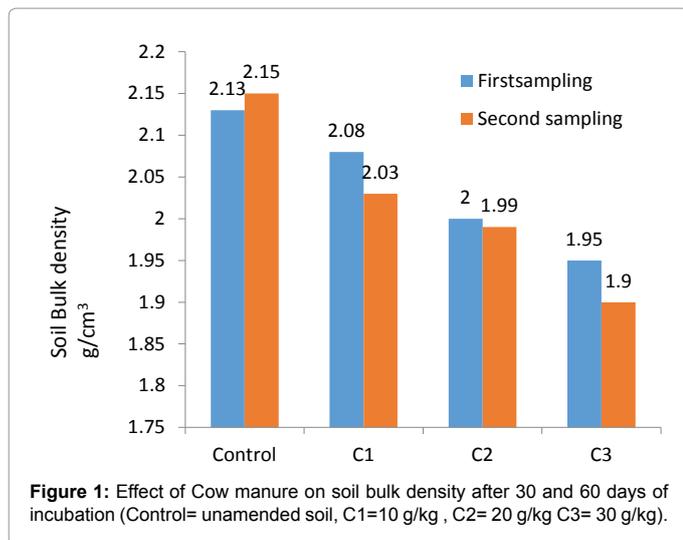
## Results and Discussion

### Soil bulk density and pH

The effect of two months of soil incubation with different levels of chicken and cow manures on the soil bulk density (SBD) has been presented in Figures 1 and 2. It can be seen that SBD decrease as compared to the control treatment. In particular, the treatment C3 caused a considerable decrease in soil bulk density by 0.18 gcm<sup>-3</sup> followed by 0.13 and 0.05 gcm<sup>-3</sup> for treatments C2 and C1 as compared to the corresponding control after one month of incubation, respectively. While after two months of incubation, the reduction in SBD in response to cow manures treatments C1, C2 and C3 was 0.25, 0.16 and 0.12 gcm<sup>-3</sup> respectively. In case of chicken manure the reduction after second month of incubation was 0.35, 0.25 and 0.15 gcm<sup>-3</sup>,

Properties	soil	Cow manure	Chicken manure
pH	7.5	8.0	7.9
EC	1.55 dSm <sup>-1</sup>	10.3 dSm <sup>-1</sup>	15.1 dSm <sup>-1</sup>
Organic matter	0.8 %	75.3 %	71.6 %
Clay	8 %	-	-
Silt	23 %	-	-
Sand	69 %	-	-
Total Zn	945 mgKg <sup>-1</sup>	576 mgKg <sup>-1</sup>	270 mgKg <sup>-1</sup>
Total Ni	522 mgKg <sup>-1</sup>	270 mgKg <sup>-1</sup>	133 mgKg <sup>-1</sup>

**Table 1:** Physical and chemical characteristics of incubated soil and organic manures.



respectively. Similar reduction in SBD as a result of manure addition has been reported by other authors. A decrease of  $0.20 \text{ g cm}^{-3}$  in bulk density of a clay loam soil as compared to control was reported by Celik et al. [16] after adding livestock manure at 25 t per hectare per year. Mossadeghi et al. [17] observed a decrease in SBD by  $0.12 \text{ g cm}^{-3}$  with a farmyard manure amendment at the rate of  $100 \text{ mg m}^{-3}$  to a silty clay loam soil. Studies [18,19] have shown that organic components have a dilution effect in lowering bulk density. The mixing of organic materials with more dense mineral fractions of soils causes a decrease in bulk density. The decrease in SBD can lead to favorable physical properties such as improving infiltration rate, increased porosity, better ion and gas exchange, enhanced root penetration and increased water holding capacity. Coarse textured soils as the one under current study, exhibit low water holding capacities. Manure addition to such soils could provide a medium with low drainage and thus potential for greater plant growth promoting immobilization of the soil heavy metals [20].

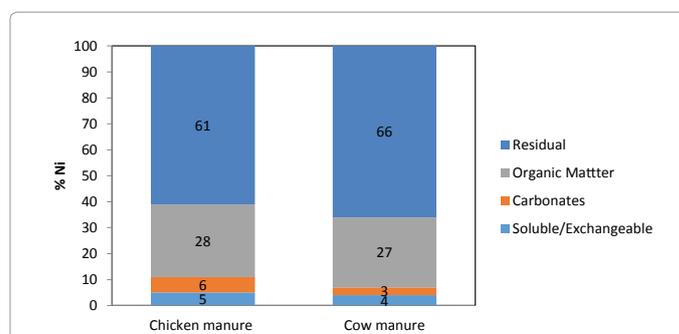
The soil pH after the incubation with the cow and chicken manure was 7.8 while in the control treatment it maintained to be 7.5, which means an increase of 0.3 units as compared to the control. Usman et al. [21] found that 1% poultry manure considerably increased soil pH from a minimum of 6.43 in soil samples without poultry manure to a

maximum of 7.12 in soil samples treated with poultry manure after a 90 days incubation period. This may be principally due to the alkaline pH of both the manures (Table 1) which, on incorporation to soil, increases the soil pH. Other possible reasons for the increase of the soil pH may be possibly due to the release of  $\text{NH}_4^+$  from organic N mineralization, the release of Ca and other base cations during mineralization of the organic manure into the soil [22,23] or the formation of organic aluminum complexes in soil solution [24]. Increase of soil pH may facilitate the adsorption of heavy metals on various soil binding sites, thus decreasing the partition of heavy metals to soil solution [25].

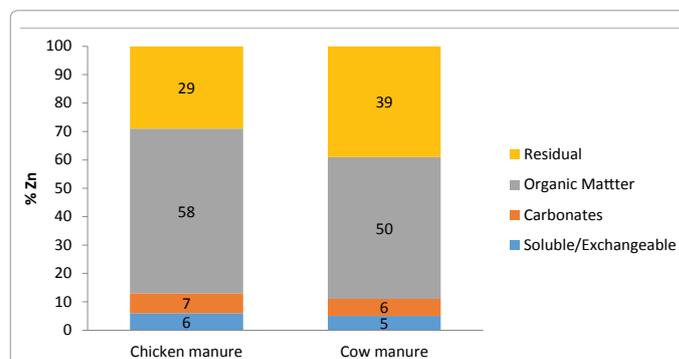
### Distribution of Ni and Zn in soil fractions

To elucidate the immobilization potential of cow and chicken manure in contaminated soil, four operationally defined fractions (soluble/exchangeable, carbonate, organic and residual) of Ni and Zn were monitored during the 60 days incubation period and the soil samplings were carried out at day 30 and day 60. The distribution of both the metals in amended soil for both sampling dates is reported in Figures 3-6. Both the metals showed a different patterns of distribution between fractions.

**First sampling:** After 30 days of incubation with cow and chicken manure, the soluble/exchangeable fraction of Ni and Zn was higher in manure treatments than the non-amended control. The maximum Ni concentration was  $29.16$  and  $30.82 \text{ mg kg}^{-1}$  for C3 and K3 respectively while for Zn it was  $78.69$  and  $70.23 \text{ mg kg}^{-1}$  in response to C3 and K3 treatments respectively. In case of carbonate fraction, the maximum Ni and Zn concentration was  $41.5$  and  $88.92 \text{ mg kg}^{-1}$  for K3 and C3 treatments. Ni associated to organic fraction was  $204.67$  and  $185.41 \text{ mg kg}^{-1}$  for C3 and K3 while in case of Zn the concentrations were



**Figure 3:** Effect of manure treatments (K3, C3) on % distribution of Ni in various soil fractions after 30 days of incubation.



**Figure 4:** Effect of manure treatments (K3, C3) on % distribution of Zn in various soil fractions after 30 days of incubation.

750.12 and 710.05 mgkg<sup>-1</sup> for C3 and K3 respectively. The residual metals formed the major portion of the total metal in soil and organic manures. The percent distribution of Ni in various fractions after 30 days of incubation (Figure 3) shows that 27 and 28 % of the metal was associated to organic fraction as compared to 4 and 5 % in soluble/exchangeable fraction and 3 and 6% as carbonate fraction in cow and chicken manure treatment respectively. This distribution clearly indicates that after residual fraction (61-66%), the bulk of the metal was bound in organic fraction. In case of Zn (Figure 4) the organic fraction constituted 50 and 58 % of the total Zn in cow and chicken manure respectively, being the biggest fraction showed a good potential of the manures to bind the metal in organic form. In this case chicken manure performed better than cow manure.

**Second sampling:** Similar, but more pronounced trend was observed in both the metals after 60 days of incubation. In case of Ni (Figure 5), 28 and 34% was found in organic fraction, while the residual reaction accounted for 58 and 53% for cow and chicken manure respectively. On the other hand, Zn-organic matter association was considerably high, as compared to other soil fractions for the second sampling. As depicted in Figure 6, it was mainly associated with soil organic matter fraction (53-57%) of total metal (soil+ manure) as compared to non-amended control immobilizing the metal and thus rendering it less available for plant uptake. The soluble and exchangeable fraction which, although, slightly increased with time remained very low (2-4%) for the two metals. The carbonate fraction made up to 11 and 12% of the total Ni and 9 and 8% of Zn in chicken and cow manure treatments respectively.

Various researchers have reported the potential use of manures as soil amendments in immobilization of heavy metals in soil. Park et al. [26] reported that chicken manure and green waste biochars immobilized soil Cd, Cu and Pb due to partitioning of metal(loid)s

from the exchangeable to organic fraction. Chamon et al. [27] found a reduced Ni, Cr and Mn uptake by the application of organic manures in soil from metal contaminated sites. Application of poultry manure compost transformed 47.8%-69.8% of soluble/exchangeable Cd to the organic bound fraction and consequently decreased Cd uptake of plants by 56.2-62.5% compared to control [28].

It is well documented that the addition of organic amendments to soils increases the immobilization of metal(loid)s through adsorption reactions. The organic amendment-induced retention of metal(loid)s is attributed to an increase in surface charge and the presence of metal(loid) binding compounds [29]. The main intent of the incorporation of amendments into contaminated soils is not to alter the total metal concentration but to impair the mobility and toxicity of metals by accelerating key immobilizing processes such as (ad)sorption, precipitation, complexation and redox reactions. In our study the possible mechanisms for the observed immobilization of Ni and Zn seem to be increase in soil pH and the organic matter. This is due to the presence of chicken and cow manure and its clear positive effect to decrease soil bulk density (Figures 1 and 2). Keeping in view the abundance and easy access to these manures, they can be utilized in Zn and Ni contaminated soils. However, continuous monitoring of the heavy metal pools in the soil is essential so as to prevent their transformation into mobile fractions which entails the risk of consequent plant uptake.

### Statistical analysis

**Soluble / exchangeable fraction:** Results presented in Tables 2 and 3 showed significant reduction in soluble/exchangeable fraction of Ni and Zn by increasing incubation time from one month to two month. The soluble/exchangeable Ni and Zn was reduced by about 53% and 30% in two month incubation compared with 1 month incubation respectively. Significant reduction was found in soluble/exchangeable Ni when using cow manure compared with chicken manure while a reverse behaviour was found in soluble/exchangeable Zn. Increasing rate of manure application (RM) resulted in a gradual increase in soluble/exchangeable Ni and Zn. The highest soluble/exchangeable value for Ni and Zn was recorded in RM3 followed by RM2, RM1 and control respectively. Soluble/exchangeable Ni and Zn was significantly affected by the second and the third level interactions (IT \* MT, IT \* RM, MT \* RM and IT \* MT \* RM) except for IT \* MT for Zn where the interaction was not significant.

**Carbonate and organic matter (OM) fractions:** Results of carbonate and OM fractions presented in Table 2 for Ni and Table 3 for Zn indicated that, increasing incubation time from 1 month to 2 months significantly increased the adsorption of Ni and Zn on both fractions. Using cow manure increased the adsorbed Ni and Zn on both fractions compared with chicken manure. Similar as in soluble /exchangeable fraction, increasing rate of manure application (RM) significantly increased the adsorbed amount of Ni and Zn on carbonate and OM fractions where the highest value of adsorbed Ni and Zn was recorded in RM3 and gradually reduced to reach the least in control treatment. Results also clearly revealed that, the adsorbed Ni and Zn on OM fraction were higher than that in carbonate fraction. Carbonate and organic matter Ni and Zn fractions were significantly affected by the second and third level interactions (IT \* MT, IT \* RM, MT \* RM and IT \* MT \* RM) except for IT \* MT for Ni in organic matter fraction where the interaction was not significant (Table 2).

**Residual fraction:** Results of residual fraction indicated that increasing time of incubation significantly reduced the Ni and Zn on residual

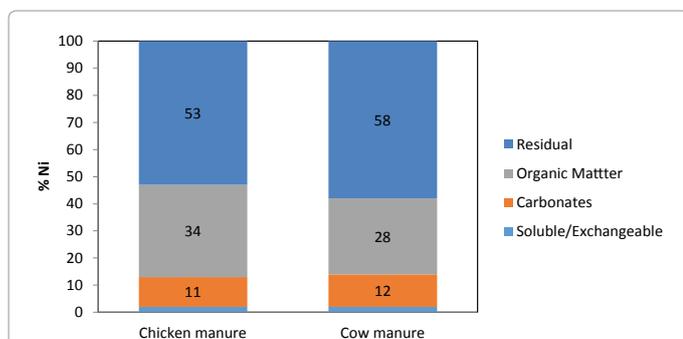


Figure 5: Effect of manure treatments (K3, C3) on % distribution of Ni in various soil fractions after 60 days of incubation.

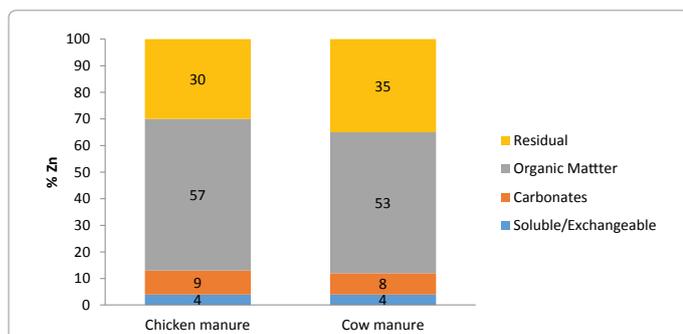


Figure 6: Effect of manure treatments (K3, C3) on % distribution of Zn in various soil fractions after 60 days of incubation.

Treatments	Soluble / Exchangeable fraction	Carbonate fraction	Organic matter fraction	Residual Fraction
Incubation time (IT)				
One Month	16.36	21.77	112.0	500.2
Two Months	7.62	53.62	131.7	473.3
<i>F test</i>	***	***	**	***
Manure Type (MT)				
Cow manure (C)	11.2	37.8	124.7	530.2
Chicken manure (K)	12.7	37.5	119.1	443.2
<i>F test</i>	***	*	**	***
Rate of manure (RM)				
Control	4.5 <sup>d</sup>	14.7 <sup>d</sup>	5.8 <sup>d</sup>	479.1 <sup>c</sup>
RM1	9.2 <sup>c</sup>	35.5 <sup>c</sup>	121.0 <sup>c</sup>	544.4 <sup>a</sup>
RM2	13.2 <sup>b</sup>	44.3 <sup>b</sup>	151.3 <sup>b</sup>	496.8 <sup>b</sup>
RM3	21.0 <sup>a</sup>	56.3 <sup>a</sup>	209.5 <sup>a</sup>	426.7 <sup>d</sup>
<i>F test</i>	***	***	***	***
<i>L.S.D (0.05)</i>	0.51	0.40	1.25	6.81
Interactions ( <i>F test</i> )				
<i>IT * MT</i>	***	***	NS	***
<i>IT * RM</i>	***	***	***	***
<i>MT * RM</i>	***	***	***	***
<i>IT * MT * RM</i>	***	***	***	***

**Table 2:** Means of Ni distribution in various soil fractions (g/kg) as affected by incubation time, manure type and rate of application. Means within each column followed by the same letter are not significantly different at level P = 0.05. (NS), not significant at level P = 0.05; (\*), (\*\*), (\*\*\*) significant at P = 0.05, 0.01 and 0.001 respectively. Letters a, b, c, d is an indication for significance in each column same letters means significantly similar, but different letters mean significantly different.

Treatments	Soluble / Exchangeable fraction	Carbonate fraction	Organic matter fraction	Residual Fraction
Incubation time (IT)				
One Month	50.2	79.1	506.6	612.5
Two Months	35.6	96.0	526.4	601.5
<i>F test</i>	***	***	**	*
Manure Type (MT)				
Cow manure (C)	45.8	89.9	538.7	692.7
Chicken manure (K)	40.0	85.3	494.2	521.2
<i>F test</i>	***	***	***	***
Rate Of manure (RM)				
Control	19.0 <sup>d</sup>	60.5 <sup>d</sup>	21.5 <sup>d</sup>	828.2 <sup>a</sup>
RM1	38.0 <sup>c</sup>	92.2 <sup>c</sup>	641.0 <sup>c</sup>	589.7 <sup>b</sup>
RM2	52.6 <sup>b</sup>	97.8 <sup>b</sup>	664.5 <sup>b</sup>	549.7 <sup>c</sup>
RM3	62.0 <sup>a</sup>	99.7 <sup>a</sup>	739.0 <sup>a</sup>	460.3 <sup>d</sup>
<i>F test</i>	***	***	***	***
<i>L.S.D (0.05)</i>	0.68	0.95	2.58	2.30
Interactions ( <i>F test</i> )				
<i>IT * MT</i>	NS	***	***	***
<i>IT * RM</i>	***	***	***	***
<i>MT * RM</i>	***	***	***	***
<i>IT * MT * RM</i>	***	***	***	***

**Table 3:** Means of Zn distribution in various soil fractions (g/kg) as affected by incubation time, manure type and rate of application. Means within each column followed by the same letter are not significantly different at level P = 0.05. (NS), not significant at level P = 0.05; (\*), (\*\*), (\*\*\*) significant at P = 0.05, 0.01 and 0.001 respectively. Letters a, b, c, d is an indication for significance in each column same letters means significantly similar, but different letters mean significantly different.

fraction (Tables 2 and 3). The amount of Ni and Zn presented in residual fraction were higher in cow manure application compared with chicken manure.

Effect of rate of manure application (RM) on Ni and Zn in residual fractions was varied. The highest significant Ni-residual fraction was recorded in RM1 followed by RM2 and control respectively. The least Ni-residual fraction was recorded in RM3 (Table 2). Gradual significant reduction in Zn-residual fraction was recorded by increasing the rate

of manure application. The least Zn-residual fraction was recorded in RM3 followed by RM2 and RM1 respectively. The highest Zn-residual fraction was found in control treatment. Ni and Zn residual fraction was significantly affected by the second and third level interactions (Tables 2 and 3).

**Effects of the triple interaction on Ni and Zn distribution in various soil fractions:** The triple interaction is considered the most important

effect because it shows the complete effect of all investigated variables on Ni and Zn immobilization in soil. Results of the triple interaction are presented in Table 4 for Ni and in Table 5 for Zn. Results obtained indicate that Soluble/ Exchangeable fractions after one month of incubation time are higher than that of two month with both cow and chicken manures. On another hand, using two months incubation period with either cow or chicken manure with high application rate immobilized heavy metal in soil, however cow manure was better than chicken manure. The best combination which optimized the immobilization of Ni and Zn in soil was RM3 of cow manure for two month incubation period followed by the same combination for chicken manure. Because these both combinations fixed a large portion of Ni and Zn on carbonate and organic matter fraction, while reduced the residual fraction to its minimum. In spite of the increase in soluble/ exchangeable fraction in these combination for Ni and Zn, their values were still within the save range.

## Conclusions

The incorporation of organic manures of cow and chicken improved the soil physical properties such as soil bulk density and reduced the concentration of soil Ni and Zn in soluble/exchangeable and carbonate fractions. The concentrations in organic and residual fractions increased which indicates the potential of these readily available manures to immobilize these heavy metals in soil. Increasing rate of manure application (RM) resulted in a gradual increase in soluble/exchangeable Ni and Zn. The highest soluble/exchangeable value for Ni and Zn was recorded in RM3 followed by RM2, RM1 and control respectively. Carbonate and organic matter Ni and Zn fractions were significantly affected by the second and third level interactions. Increasing time of incubation significantly reduced the Ni and Zn on residual fraction. The best combination which optimized the immobilization of Ni and Zn in soil was RM3 of cow manure for two month incubation period followed by the same combination for chicken manure.

Incubation time	Manure type	Rates g/kg soil	Soluble / Exchangeable fraction	Carbonate fraction	Organic matter fraction	Residual Fraction
One month	Cow manure (C)	Control	2.9	16.0	5.1	464.9
		10	13.9	17.9	119.9	602.9
		20	14.1	22.1	132.1	580.2
		30	29.0	24.0	205.0	508.0
	Chicken manure (K)	Control	5.0	14.0	7.0	481.0
		10	9.9	17.9	103.9	517.4
		20	24.9	20.1	138.1	449.9
		30	31.1	42.1	185.1	397.1
Two month	Cow manure (C)	Control	5.0	13.0	5.0	490.0
		10	7.0	51.0	142.0	587.0
		20	5.0	70.0	164.0	549.0
		30	13.0	89.0	225.0	460.0
	Chicken manure (K)	Control	4.9	15.9	5.9	480.4
		10	6.1	55.1	118.1	470.1
		20	9.0	65.0	171.0	408.0
		30	10.9	69.9	222.9	341.9
L.S.D (0.05)			0.85	0.67	2.0	11.3

**Table 4:** Effect of the interaction among incubation time, manure type and rate of application on Ni distribution in various soil fractions (g/kg).

Incubation time	Manure type	Rates g/kg soil	Soluble / Exchangeable fraction	Carbonate fraction	Organic matter fraction	Residual Fraction
One month	Cow manure (C)	Control	16.9	57.9	18.9	803.9
		10	48.9	82.9	640.9	745.9
		20	66.4	90.2	677.1	685.1
		30	79.0	89.0	750.0	584.0
	Chicken manure (K)	Control	20.3	69.1	23.0	812.0
		10	44.9	80.9	602.9	469.9
		20	54.9	82.9	629.9	445.9
		30	70.1	80.1	710.1	353.1
Two month	Cow manure (C)	Control	18.0	50.0	24.0	854.0
		10	30.0	112.0	686.0	690.0
		20	51.0	116.0	710.0	640.0
		30	56.0	121.0	803.0	539.0
	Chicken manure (K)	Control	20.9	64.9	19.9	842.9
		10	28.1	93.1	634.1	453.1
		20	38.0	102.0	641.0	428.0
		30	42.9	108.9	692.9	364.9
L.S.D (0.05)			1.13	1.57	4.28	3.82

**Table 5:** Effect of the interaction among incubation time, manure type and rate of application on Zn distribution in various soil fractions (g/kg).

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