EFFECT OF ORGANIC MANURE AMENDMENT SOILS ON UPTAKE OF Pb, Cd AND Zn IN SPINACH

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ABSTRACT
Organic amendment of soil is an effective pathway of remediation of heavy metals in polluted agricultural soils and it is mostly carried out in situ, hence, this study aimed was to re-established the remediation potentials of organic manure from different sources. Therefore, the study was conducted with spinach as the test crop to investigate the uptake of lead (Pb), cadmium (Cd) and zinc (Zn) from agricultural soils treated with poultry, cow and pig manure compost with different concentrations of the metals. The results showed that manure application decreased Cd content in the plant shoot. This indicated that application of manure compost reduced Cd concentration in the spinach. The lowest and highest available Pb content was found in soils treated with 30g and 10.71g of poultry droppings, respectively, this suggest low mobility of Pb in 30g compost manure compare to 10.71g compost manure. The lowest and highest available Zn content was found in soils treated with 30g and 7.14g of cow dung, respectively, although these materials contained the same concentration of Zn suggesting low mobility of Zn in soil with 30g cow dung compare with 7.14g cow dung. Generally, from the results, spinach showed its suitability for phytoextraction of Cd, Pb and Zn. However, application of organic manure could make spinach suitable for phytostabilization in a contaminated soil.

Keywords: Heavy Metals, Phytoremediation, Organic manure, Agricultural Soils

INTRODUCTION
Heavy metals are one of the most common and harmful pollutants in soils today. The availability of these metals poses risk to human and animal health as well as plant growth (Katoh et al., 2016). Due to the variable oxidation states, heavy metals can be transformed to become either less toxic, easily volatilized, more water soluble (and thus can be removed through leaching), less water soluble (which allows them to precipitate and become easily removed from the environment) or less bioavailable (Garbuis and Alkorta, 1997, 2003). Hence, these metals cannot be degraded during bioremediation but can only be transformed from one organic complex or oxidation state to another. Phytoremediation, an option in bioremediation, is defined as an emerging technology which uses selected plants to clean up contaminants from the environment in order to improve the quality of the environment (Tangahu et al., 2011). The use of soil amendment has also been described as a component of phytoremediation (Salt et al., 1995). Where the toxicity levels of pollutants do not encourage growth and survival of plants meant for remediation, the soil amendment is applied to encourage plant growth and survival. However, the application of soil amendment can decrease heavy metal bioavailability, thus shifting them from “plant available” forms to fractions associated with organic matter, carbonates or metal oxides (Walker et al., 2004) consequently resulting in the reduction of metals uptake by the installed plants.

However, in recent years, availability of Cd, Pb and Zn to agricultural soils and subsequently to plants through various sources has been reported in literature (Seaward and Richardson, 1990; Kabata-Pendias, 2001; Liu et al., 2005; Awode et al., 2008). This could be made available through irrigation, synthetic fertilizer application and direct dumping of refuse on the agricultural soils. Therefore, the aim of the present study was to determine the effect of organic manure (poultry, cow and pig manures) amendment soils on uptake of Pb, Cd and Zn in Spinach

MATERIALS AND METHODS
Sample collection
A laboratory experiment (in a pot) was conducted with spinach grown in soils collected from a Fadama site along PZ, Zaria, in Kaduna State, the farmland belongs to Mallam Adamu Sunusi. A total of ten (10) soil samples were collected from the farmland. This was achieved by dividing the plot of farm into four and one at the centre and two samples were collected at each location, then grab samples were homogenized together to give a composite sample. The soil samples were collected into polythene bags and transported to the laboratory. The organic matter...
namely; poultry and cow dung were collected from different farms in National and Production Research Institute (NAPRI), Zaria, Kaduna State. The pig slurry was collected from a pig farm in Mangu behind Dakace, Zaria. The soil sample collected from the farmland was divided into two; one part was used for physicochemical analysis of soil parameter and the other for the amended soil experiment. The soil for the physicochemical analysis was air dried and sieved with 0.2 mm mesh size and stored in a plastic bottle for further analysis.

**Determination of Soil Physicochemical Parameters**
The pH of soil samples was measured with a soil:water ratio of 1:2 (w/v) using Crison Micro pH 2000, pH meter (Herdeershot et al., 1993). Electric conductivity (EC) Electrical conductivity (EC) was determined in supernatant of 1:5 soil/water mixtures using Digital Conductivity meter model No PT360 (Herdeershot et al., 1993). Soil organic matter (SOM) was measured using dichromate oxidation (Nelson and Sommers, 1982). The exchangeable base was determined by the sum of exchangeable cations (Banerjee et al., 2004). Determination of total nitrogen and available phosphorus was carried according to Allen (1974).

**Preparation of Stock Solution**
From the soil samples collected, 3 kg each of the soil was weighed into twenty eight (28) different pots for greenhouse pot experiment. Stock solution of Cd, Pb and Zn were prepared using standard methods, and 100 ml of tertiary mixture of stock solution of Cd, Pb and Zn were added to 1 kg of the dried soil to give 100 mg Cd, 400 mg Pb and 400 mg Zn. Therefore, in each 3 kg of soil, 300 mg of Cd, 1200 mg of Pb and 1200 mg of Zn were added and 300 ml of water was added to each pot to bring the soil to 80% of field capacity.

**Organic Amended Soil**
The experiment was performed using four levels of the composted manure: poultry, cow and pig manure: 30 g, 14.2 g, 10.7 g and 7.14 g in duplicate each with 3 kg of soil sample.

**Preparation of the Nursery, Transplantation and Harvest of Sample**
The nursery of spinach plant was prepared in Institute of Agricultural Research (IAR), Zaria in April, 2017 in the Department of Soil Science, Ahmadu Bello University, Zaria. After two weeks of plantation, it was transplanted into the pots (containing the compost manure and soils). The pots were taken to the greenhouse on the same day. The plants were harvested after 40 days of plantation. The plants were cut into two halves - the roots and shoots were dried in an oven and grinded using porcelain mortar and pestle.

**Digestion of Vegetable Samples**
Root and shoot of spinach samples of 0.2 g finely grinded were weighed separately into 30 ml porcelain crucible. Sample in crucibles was then ignited in a muffle furnace for an hour at a temperature of 600°C in which grayish white ash was indicated for complete ignition. 5 ml of 1N HNO₃ solution was then added to it and sample digested to dryness on a hot plate at low heat under ventilation. The sample was then returned to the furnace at a temperature of 400°C for 15 minutes until a perfectly grayish white ash is obtained. Then 10 ml of 1N HCl was added to each sample and the solution filtered into 50 ml volumetric flask. The crucible and the filter paper were then washed with 30 ml portion of 0.1 N HCl three times and make up to mark with 0.1N of HCl solution.

**RESULTS AND DISCUSSION**

**Soil physicochemical parameters**

The results of soil physicochemical parameters are presented in Table 1 below.

<table>
<thead>
<tr>
<th>Parameters</th>
<th>Mean</th>
</tr>
</thead>
<tbody>
<tr>
<td>pH_wat.</td>
<td>6.84</td>
</tr>
<tr>
<td>pH_{calcium chloride}</td>
<td>6.30</td>
</tr>
<tr>
<td>OM (%)</td>
<td>1.20</td>
</tr>
<tr>
<td>AP (mg/100g)</td>
<td>49.40</td>
</tr>
<tr>
<td>EC(µS/cm)</td>
<td>0.014</td>
</tr>
<tr>
<td>TN (%)</td>
<td>0.35</td>
</tr>
<tr>
<td>Exchangeable Base(cmolkg⁻¹)</td>
<td></td>
</tr>
<tr>
<td>Na</td>
<td>3.20</td>
</tr>
<tr>
<td>Mg + Ca</td>
<td>90.90</td>
</tr>
</tbody>
</table>

**Cd Uptake of Spinach**
The contents of available cadmium determined in the plant using different manure compost of different amount showed: 0.46 mg/kg (10.71 kg of poultry droppings) – 1.83 mg/kg (30 g of poultry droppings), 0.23 mg/kg (7.14 g of cow dung) – 1.01 mg/kg (30 g of cow dung) and 0.66 mg/kg (14.28 g of pig slurry) – 5.00 mg/kg (7.14 g of pig slurry) as shown in Figure 1. The lowest and highest available Cd content was found in soils treated with 7.14 g of cow dung and pig slurry, respectively, although these materials contained the same concentration of Cd suggesting low mobility of Cd in cow dung compare to pig slurry. Analysis of variance (ANOVA) of Cd in the plant grown in different soil compost showed significant variation (p = 0.026), this indicates that different manure compost effect varying binding capacity on the Cd.

It is noteworthy to indicate that decrease in rate of manure compost significantly suppressed the concentration of Cd in the plants with exception of...
pig slurry which indicates an increased in concentration of Cd in the plant. Also, comparison of the results to the control plants, application of manure compost at 30g increased the availability of Cd 213, 74 and 84% for poultry droppings, cow dung and pig slurry respectively. At 14.28g manure compost soil, the uptake of Cd was suppressed at the rate of 3.4, 31.0 and 13.7% for poultry droppings, cow dung and pig slurry respectively. Similarly, at 10.71g manure compost soil, the uptake of Cd was suppressed at the rate of 20.7, 55.2 and 89.7% for poultry droppings, cow dung and pig slurry respectively.

Furthermore, at 7.14g manure compost soil, the uptake of Cd was suppressed using cow dung (60.3%) manure and increased using pig slurry manure at the rate of 762.1%. The results is contrary to report by Tlustoš et al. (1997) who found a decrease of the cadmium content in oat biomass after manure application into the experimental soil. Also, the results showed contrary trend to report that addition of organic materials into soil can build up Cd stable complexes which are not able to enter plant cells (Hanc et al., 2008). However, the results of Shuman et al. (2002) showed that a decrease in phytotoxicity was attributed to redistribution of Cd from the water soluble and exchangeable fractions to the organic fraction, which decreased the plant availability and Cd uptake. According to Stewart et al. (2000), application of manure compost improves nutrient cycling. Manure is a more easily mineralizable organic matter source whose nutrients are more readily available to plants.

**Pb Uptake of Spinach**

From the results shown in Figure 2, the contents of available Pb determined in the spinach using different manure compost of different amount showed: 31.28 mg/kg (30 g of poultry droppings) – 92.64 mg/kg (10.71g of poultry droppings), 37.39 mg/kg (10.71g of cow dung) – 41.81 mg/kg (14.28g of cow dung) and 33.09 mg/kg (30g of pig slurry) – 74.28 mg/kg (14.28g of pig slurry). The lowest and highest available Pb content was found in soils treated with 30g and 10.71g of poultry droppings, respectively, this suggest low mobility of Pb in 30g compost manure compare to 10.71g compost manure. Analysis of variance (ANOVA) of Pb in the spinach grown in different soil compost showed no significant variation (p > 0.05), this indicates that different manure compost does not affect significant changes in mobility of Pb to the spinach.

**Fig. 1: Mean concentration of Cd in Spinach grown in different compost manure soils**

Furthermore, at 7.14g manure compost soil, the uptake of Cd was suppressed using cow dung (60.3%) manure and increased using pig slurry manure at the rate of 762.1%. The results is contrary to report by Tlustoš et al. (1997) who found a decrease of the cadmium content in oat biomass after manure application into the experimental soil.
Fig. 2: Mean concentration of Pb in spinach grown in different compost manure soils

Similar to Cd, the check on percentage availability of Pb to the spinach indicated that decrease in rate of manure compost significantly increased the concentration of Pb in the plants for poultry droppings, while cow dung and pig slurry increased the concentration of Pb at 14.28g compost manure soils and decrease the availability of Pb as the rate of manure compost decreases. From the results, it showed that composition and application rate of composts play an important role in the availability of Pb sorption.

Comparison of Pb uptake by the spinach grown in the manure compost soil to the control soil showed that application of manure compost at 30g increased the availability of Pb 7.2% cow dung. At 14.28g manure compost soil, the uptake of Pb was increased at the rate of 58.2, 19.7 and 112.7 % for poultry droppings, cow dung and pig slurry respectively. Similarly, at 10.71g manure compost soil, the uptake of Pb was increased at the rate of 165.3, 7.1 and 6.3% for poultry droppings, cow dung and pig slurry, respectively.

Zn Uptake of Spinach

The contents of available Zinc determined in the plant using different manure compost of different amount showed: 0.51mg/kg (10.71kg of poultry droppings) – 1.78mg/kg (30g of poultry droppings), 0.29mg/kg (30g of cow dung) – 1.96 mg/kg (7.14g of cow dung) and 0.69mg/kg (30g of pig slurry) – 1.78mg/kg (10.71g of pig slurry) as shown in Figure 3. The lowest and highest available Zn content was found in soils treated with 30g and 7.14g of cow dung, respectively, although these materials contained the same concentration of Zn suggesting low mobility of Zn in soil with 30g cow dung compare to one with 7.14g cow dung.. Analysis of variance (ANOVA) of Zn in the plant grown in different soil compost showed significant variation (p = 0.020), this indicates that different manure compost effect varying binding capacity on the Zn.
Heavy metal immobilization by organic amendments has been attributed to a number of reasons; Clemente et al. (2005) and Erinle et al. (2017) reported that manure provides binding sites for metals thus reducing their availability to plant roots, Cao et al. (2011), metal immobilization was due to its conjugation with the manure-containing phosphorus to form insoluble hydroxyppromorphite. According to Walker et al. (2004) and Bolan et al. (2014), manure-containing humic acids have immobilizing effects on metalloids. Likewise, manure application has been reported to increase soil organic matter, electrical conductivity (EC) as well as pH (Zhao et al., 2014). Hence, increase in these soil properties with the application of manure could have consequently caused a decrease in Zn mobility for easy uptake by the plant shoot.

CONCLUSION

From the results presented in the study, the measured heavy metals showed contrasting responses to the applied manure compost. Firstly, manure application decreased Cd content in the plant shoot. This indicated that application of manure compost reduced Cd concentration in the spinach. The lowest and highest available Pb content was found in soils treated with 30g and 10.71g of poultry droppings, respectively, this suggest low mobility of Pb in 30g compost manure compare to 10.71g compost manure. The lowest and highest available Zn content was found in soils treated with 30g and 7.14g of cow dung, respectively, although these materials contained the same concentration of Zn suggesting low mobility of Zn in soil with 30g cow dung compare to one with 7.14g cow dung. Also, generally manure application showed suppressive effect on heavy metal uptake into above ground portion, as presented in this study. Spinach might be suitable for phytoextraction of Cd, Pb and Zn but application of manure could make spinach suitable for phytostabilization in a contaminated soil.

REFERENCES


