

Heavy Metals Accumulation In Amaranths (*Amaranthus Hybridus*) And Jews Mallow (*Corchorus Olitorius*) Grown on Soil Amended With Tannery Sludge From Challawa Industrial Area, Kano State Nigeria.

*Garba¹, M. D. Mohammed², M. I. and Kiyawa³, S. A.

¹Department of Chemistry, Federal College of Education (Tech), Bichi

²Department of Pure and industrial Chemistry, Bayero University, Kano

³Department of Chemistry, North-west University, Kano Nigeria

*Corresponding author: *Garba

Abstract: Tannery sludge is currently being used in the cultivation of vegetables. This study investigated the concentration of heavy metals in Amaranths (*Amaranthus hybridus*) and Jews mallow (*Corchorus olitorius*) grown on tannery amended soil in Pot. Heavy metals such as Fe, Ni, Co, and Cu were analyzed in the harvested vegetable tissues using Atomic Absorption Spectrophotometry. The result showed highest range (1247 – 2188.20mg/kg) of Iron in the roots, Nickel (13.80 – 17.50mg/kg) and Copper (15.50 – 20.70mg/kg) both in the leaves of *Corchorus olitorius*. The concentrations, except of Iron were lower than the FAO/WHO recommended limits. However, Transfer Factor for Copper in *Corchorus olitorius* tissues and *Amaranthus hybridus* leaves indicates higher potential of the vegetables for bioaccumulation. This implies continuous consumption of the vegetables may pose a significant threat and hazard effects to human and animals. Hence, farmers should be guided on the use of tannery sludge on agricultural land.

Keywords: Heavy metals, Vegetables, Tannery sludge, Amended soil.

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I. Introduction

The use of sludge for agricultural purposes has being a common practice because of its nutrients content, which improves the organic matter content and water-holding capacity of soil (Zoubi *et al.*, 2008). Numerous organic compounds are present in Tannery sludge and about 70% of the organic substances have applications as a high-quality fertilizer (Jamali *et al.*, 2006; Li, *et al.*, 2005). Thus, reduces fertilizer costs and improves soil fertility (Hough, *et al.*, 2003). Hence, its application on agricultural soil can be very effective in improving crop productivity. A number of experiments on the use of tannery sludge have demonstrated positive effects of the sludge, both raw and compost, on plant yield and soil properties (Gondek & Fillipek-mazur *et al.*, 2003; Mujacic, 2011). However, the presence of numerous metals in sludge has been severally reported (Andre *et al.*, 2012; Olowu *et al.*, 2012; Garba & Mohammed, 2017) and their concentration in soils persists for a very long time after land application (Ana, *et al.*, 2014). Heavy metals are naturally present in soil at varying levels, and originate mainly from several anthropogenic sources such as fertilizers applications, Animal manure, Sludge, or Atmospheric deposition. Regardless of the origin, high levels of metals can result in the degradation of soil fertility and poor quality of agricultural products. It could also pose a significant threat and hazard to human, animal and ecosystem (Rahman, *et al.*, 2013). Therefore, the use of sludge is often limited not only due to its pathogenic microorganisms, but mainly because of the large heavy metals contents. Their mobility and bioavailability to plants are influenced by several factors such as plant species, growth stage, types of soil and metals, soil conditions, weather, pH and the environment (Debrah, *et al.*, 2012). Therefore, agricultural utilization of tannery sludge implies knowing the metals concentration as well as its bioavailability in vegetables. In this study, leafy vegetables namely, Amaranth (*Amaranthus hybridus*) and Jews mallow (*Corchorus olitorius*), that are traditionally important across different cultures in Nigeria and other West African nations were used in the analysis. Because these vegetables serve as food sources and thus offer rapid and ideal means of providing adequate vitamins, mineral salts, trace essential elements and fibre (Fasola & Ogunsola, 2014). The study therefore evaluates the vegetables' ability in accumulating heavy metals such as Fe, Ni, Co and Cu in their tissues (roots, stems and leaves) and also determines their transfer factors and offer suggestion on the physiological fates of the metals in the environment.

II. Materials And Methods

All experiments were performed with analytical grade reagents and deionized water was used throughout the analysis. Heavy metals concentrations were determined with Variant spectra Atomic Absorption Spectrophotometer (Agilent Technology 240 model) equipped with a digital readout system. All measurements were carried out in triplicate.

Pot experiment:

A pot experiment was conducted using surface soil (0 – 20cm depth) obtained from a farm at the new site of Bayero University Kano and the tannery sludge sample used was from Challawa industrial area, Kano Nigeria. Tannery sludge amended soil samples were formed by mixing each sludge sample with soil in 1:10 ratio. Each homogenous mixture was transferred into a polythene pot measuring 16.5cm × 12cm × 14.5cm and was replicated three times. The control Pot contained only the soil sample (uncontaminated). Seeds of Amaranths (*Amaranthus hybridus*) and *Corchorus olitorius* (Jute or Jews mallow) were sown in each Pot separately. The Pots were arranged in a complete block design, in eleventh columns and three rows. Each row contained a particular vegetable species and tenth columns (each contained particular amended soil). The eleventh column contained Pots with only soil serving as the control. Two weeks after planting, vegetables were thinned and left to grow under natural conditions with watering at regular interval in the Botanical garden of Bayero University. After eight weeks, the plants were harvested by uprooting (when soil was moist) using plastic hand trowel and gently removed (Ogunkunle, et al., 2013). The vegetables were then wrapped, labeled and transported to the laboratory.

Preparation and analysis of Soil samples:

Samples of the background (uncontaminated) and amended soil used for the experiment were air-dried under room temperature, grounded to fine particles using mortar and pestle. The grounded particles were sieved through 2mm sieve (Ogunkunle, et al., 2013). One gram each of soil samples was placed in 250cm³ Pyrex beaker and 20cm³ of aqua-regia (mixture of HCl and HNO₃ in 3:1 ratio) was added. 10cm³ of 30% hydrogen peroxide was then added slowly without allowing any losses. The beaker was covered with a watch glass and heated for 2 hours at 90°C. The digest obtained was then filtered through number 42 Whatman filter paper and diluted to 100cm³ using a flask with deionized water (Association of Official Analytical Chemist; AOAC, 1995). pH and conductivity were separately determined by dissolving 10g of the amended soil in 50cm³ deionized water (1:5w/v) and shaken until homogeneity was reached. A glass electrode pH meter (Jenway 3510 model) and conductivity meter (Jenway 4010 model) were used to measure pH and conductivity respectively (Rayment & Higginson, 1992). Soil organic matter content was determined using the modified Walkley – Black method as described by (Bruce & Schelte, 2009). The same was repeated for the control soil.

Preparation and analysis of plant samples:

The harvested vegetables were carefully chopped into smaller portions (root, stem and leaf) with a clean stainless steel knife. The chopped vegetable samples were air dried and reduced in size by grinding to a fine powder using cleaned mortar and pestle. Each sample was sieved using 2mm plastic sieve and stored in a labeled container.

The vegetable tissues (roots, stems and leaves) were measured one gram separately into porcelain crucibles. The crucibles were placed in a muffle furnace and heated to 500°C for 8 hours. The completely ashed (clean white ash) sample was allowed to cool and then removed from the furnace. The ash was dissolved with 5cm³ of 6M hydrochloric acid, warmed and then filtered through number 42 whatman filter paper into 100cm³ volumetric flask. The crucible was washed as well as the filter paper several times and the solution was then made up to the mark. The procedure was repeated to all the samples including the control.

Determination of Transfer factor:

Transfer factors (TF) also bioconcentration factor is an index demonstrating the potential of whole vegetable or its tissues to accumulate metal from soil. It is a parameter used to describe the transfer of elements from soil to plant's edible parts or tissues. It is calculated as the ratio between the concentration of heavy metals in vegetables and that in the corresponding soil all based on dry weight for each vegetable separately (Shukla, et al., 2013). $TF = C_{\text{tissue}} / C_{\text{soil}}$ Where C_{tissue} is the concentration of a metal in the root, stem or leaves (dry weight basis) and C_{soil} is the total concentration of the same metal in the soil (dry weight basis) where the plant was grown. The higher the value of the TF, the more mobile/ available the metal is. High TF value indicates suitability of the plant or its tissue for phytoextraction (Rangnekar, et al., 2013).

III. Results

Concentration of heavy metals in the Background and amended Soil samples:

The results indicated that the pH of the amended soil was slightly alkaline with an average pH of 7.40 ± 0.19 and higher than ($\text{pH} = 6.87 \pm 0.03$) found in the background (control) soil and was within the allowable limits (6 – 9) for appropriate growth and efficient uptake of nutrients materials from soil (Jadia & Fulekar, 2008). The EC and OM content were also higher in the amended soil compared to the control soil. The concentration of heavy metals in the amended soil were $6061.64 \pm 101.43 \text{mg/kg}$ Fe, $19.40 \pm 1.52 \text{mg/kg}$ Ni and $13.50 \pm 8.72 \text{mg/kg}$ Cu, while $481.00 \pm 0.99 \text{mg/kg}$ Fe, $12.90 \pm 0.07 \text{mg/kg}$ Ni and $2.00 \pm 0.36 \text{mg/kg}$ Cu concentrations were observed in the control soil. Cobalt was not detected in any of the sample analyzed (Table 1) and all the heavy metals analysed in the soil were below the World Health Organisation permissible limits (FAO/WHO, 2007).

Table 1: Some physicochemical properties of the Background and amended soil

Parameter	Background Soil used	Amended soil	Safe limits
Ph	6.87 ± 0.38	7.40 ± 0.20	-
Conductivity (mS/cm)	0.42 ± 0.68	1.12 ± 0.27	-
Organic matter (%)	0.90 ± 0.23	1.83 ± 0.47	-
Iron (mg/kg)	481.00 ± 0.99	6061.64 ± 101.43	50000^*
Nickel (mg/kg)	12.90 ± 0.07	19.40 ± 1.52	$75 - 150^*$
Cobalt (mg/kg)	ND	ND	50^*
Copper (mg/kg)	2.00 ± 0.36	13.50 ± 8.72	$135 - 270^*$

Source: [FAO/WHO, 2007*; Chiroma et al., 2014^x]

Heavy metals concentration in vegetables:

The range and mean concentration of heavy metals in the vegetables (*Amaranthus hybridus* and *Corchorus olitorius*) tissues are presented in Table 2. In the roots, the concentrations ranged from $617.76 - 2188.20 \text{mg/kg}$ for Fe, $4.00 - 16.80 \text{mg/kg}$ for Ni and $9.60 - 18.80 \text{mg/kg}$ for Cu. In stem, the metals ranged between $224.70 - 1475.30 \text{mg/kg}$ for Fe, $2.80 - 15.40 \text{mg/kg}$ for Ni and $5.50 - 17.20 \text{mg/kg}$ for Cu. In the leaves, it ranged between $256.20 - 1540.70 \text{mg/kg}$ for Fe, $8.60 - 17.50 \text{mg/kg}$ for Ni and $10.70 - 20.70 \text{mg/kg}$ for Cu and Co was not detected (ND). The accumulation of the metals revealed higher concentration in the root tissues significantly compared to the control. The highest concentration of Fe ($1681.82 \pm 258.86 \text{mg/kg}$) was found in the roots of *Corchorus olitorius* and the lowest $417.44 \pm 151.87 \text{mg/kg}$ in *Amaranthus hybridus* stem. The trend of Fe accumulation was in the order of root > leaves > stem in the vegetables. In contrast, the leaves accumulated relatively higher concentration of Ni and Cu in the vegetables. The highest concentration of Ni in *Amaranthus hybridus* and *Corchorus olitorius* were $8.46 \pm 1.18 \text{mg/kg}$ and $15.73 \pm 1.17 \text{mg/kg}$ respectively.

Transfer Factor:

In the study, the transfer factor which indicates the potential of the vegetable or its tissues to accumulate metal from soil revealed values varied in the range of 0.07 to 1.31 (Table 3). The values in *Corchorus olitorius* were detected to be 1.13, 1.12, and 1.31 in the roots, stem and leaves respectively. While only leaves of *Amaranthus hybridus* showed TF index greater than 1, demonstrating its potential to accumulate Copper.

IV. Discussion

Heavy metals analyzed showed varied concentrations in the vegetable tissues with highest concentration of iron ($1681.82 \pm 258.86 \text{mg/kg}$) in the roots of *Corchorus olitorius*. While Nickel and Copper were both higher in the leaves of the vegetables (Table 2). The high concentration of Fe in the species tissues could be attributed to the microbial consortium which excretes organic acids that facilitate the absorption and accumulation of Fe in the roots (Debrah, et al., 2012). In support of this Crowley, et al., (1991) reported that some microorganisms excrete organic compound which increase bioavailability and enhance root absorption of essential metals including Fe. The level in the vegetables analyzed were above the prescribed safe limits (425.50mg/kg) set by Food and Agriculture Organisation and World Health Organisation; FAO/WHO. But transfer factor index indicates the vegetables are not accumulators of the metal. But, since the species are edible, the elevated Fe levels call for concern as it can cause some health hazards to the consumers at chronic levels. Therefore, periodic monitoring of contamination and consumption rates is necessary to assess the overall exposure level in the consumer's community. The highest concentration of Ni ($15.73 \pm 1.17 \text{mg/kg}$) in

Corchorus olitorius leaves, indicates the ability of the vegetable for the metal. The value showed that greater proportion of the metal absorbed in the leaves. This indicates that Corchours olitorius had greater ability to transfer Ni to the aerial parts than did Amaranthus hybridus, shown by relatively higher transfer factor index.

Table 2: Heavy metal concentrations (mg/kg) in vegetables: Mean and (Range)

metals	Sample	Amaranthus hybridus			Corchorous olitorius		
		Root	Stem	Leaf	Root	Stem	Leaf
Iron	Control	327.00	215.40	309.00	661.20	177.80	594.40
	Amendment	1256.64 (617.76 - 1940.70)	417.44 (224.7 - 590.80)	691.59 (256.20 - 951.00)	1681.82 (1247.00 - 2188.20)	829.64 (556.10 - 1475.30)	1049.37 (915.90 - 1540.70)
Nickel	Control	2.60	1.60	1.25	13.00	11.60	12.75
	Amendment	6.15 (4.0 - 9.0)	4.38 (2.80 - 7.00)	8.46 (8.60 - 10.40)	15.41 (11.70 - 16.80)	14.10 (12.50 - 15.40)	15.73 (13.80 - 17.50)
Cobalt	Control	ND	ND	ND	ND	ND	ND
	Amendment	ND	ND	ND	ND	ND	ND
Copper	Control	2.60	2.00	3.70	3.50	2.28	4.50
	Amendment	11.08 (9.60 - 12.80)	7.50 (5.50 - 8.80)	14.14 (10.70 - 18.60)	15.26 (13.10 - 18.80)	15.11 (11.00 - 17.20)	17.63 (15.50 - 20.70)

Table 3: Transfer factor of heavy metals through different vegetable tissues

Metals	A. hybridus			C. olitorius		
	Root	Stem	Leaf	Root	Stem	Leaf
Iron	0.21	0.07	0.11	0.28	0.14	0.17
Nickel	0.32	0.23	0.44	0.79	0.73	0.81
Cobalt	ND	ND	ND	ND	ND	ND
Copper	0.82	0.56	1.05	1.13	1.12	1.31

It seems that Amaranthus has a mechanism to sequester/detoxify excess Ni in the vacuoles of root cells, whereas Corchorus lacks this mechanism. Thus, concentrations of Ni in the tissues were all below the WHO/FAO permissible limit of 67.90 mg/kg recommended in vegetables (WHO, 2007). Therefore, consumption of the vegetables may assist in preventing the adverse effect of Ni deficiency, which results in retard growth, because of its role in nucleic acid metabolism and protein synthesis (Cempal & Nikel, 2006). Similarly, the trend of Ni where higher concentrations were noticed in the leaves of the vegetables was also observed for Cu. The maximum concentration of Cu (17.63 ± 1.61mg/kg) was observed in the leaves of Corchorus olitorius followed Amaranthus hybridus (14.14 ± 2.48mg/kg). The order of Cu accumulation was leaves > roots > stem. The trend was also observed in the control and the concentrations were below the recommended limits (73.30mg/kg) set by FAO/WHO. However, transfer factor of Cu showed greater affinity for the metal by Corchorus olitorius, possibly due the physical and chemical characteristics of the rhizosphere, which changes the characteristics of trace metals in plants (Wang, et al., 2008). Hence, indicated high amounts of Cu accumulation (TF > 1) compared to Amaranthus, charactering the vegetables as metal accumulators and on may be a candidate plant in remediation of soil contaminated with metal. Copper being an essential micronutrient element that is vital to the health of living things, it plays important roles in various oxidations-reduction reactions. It serves as an essential co-factor for several oxidative stress-related enzymes including catalase, peroxidase, etc (Paul, et al., 2012). But, it's also toxic to man at a concentration of 250mg/day, causing anemia, kidney damage, intestinal irritation, hypertension, sporadic fever and coma, its acceptable limit for human consumption is 10ppm (Nair et al., 1992). Thus, higher dietary intake can result in a number of adverse health effects.

V. Conclusion

Tannery sludge though has high nutrients content for plants, but heavy metal is one of the limiting factors against its use in agriculture. High concentration of Fe was detected in all the vegetables tissues studied. The potential to transfer Cu to the *Corchorus olitorius* tissues and the aerial parts (leaves) of *Amaranthus hybridus* leaves were also identified in the study. Therefore, consumption of the vegetables may pose adverse health effect. Measures should be taken on the industries to treat their effluents to conform regulated standards before discharge into the environment.

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