DETERMINATION OF HEAVY METAL CONTAMINANTS IN LEAFY VEGETABLES CULTIVATED AND MARKETED IN ABA, NIGERIA

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ABSTRACT

Human food chain toxicity has been shown to be influence by application of fertilizers. This research studies the influence of fertilizer application on the uptake of heavy metals by *Telfaira occidentalis* and *Talinum triangulare* and the environmental health implication in Aba, Nigeria. The different fertilizer samples used were $N_{15}P_{15}K_{15}$, $N_{20}P_{10}K_{10}$ and $N_{27}P_{13}K_{13}$ with a soil treatment of 2.0g/kg. The heavy metal composition of the NPK fertilizer samples were studied using Atomic Absorption Spectrophotometer (AAS). Iron levels rated highest in the soil and the NPK fertilizer samples while V, As, Hg and Ag were not detected. The mean concentrations (mg/kg) of other metal followed the sequence Ni>Bo>Mg>Co>Zn>Mn>Pb>Mo>Cu>Cd. The potential toxic elements such as Cd and Pb in the fertilizers were 0.01 and 0.19mg/kg mean concentration respectively. The addition of the fertilizers showed an increase in heavy metal content of the vegetables. The result showed that the vegetables (*Talinum triangulare* and *Telfaira occidentalis*) accumulated significant amount of Iron and Zinc in their leaves than other heavy metals examined. *Talinum triangulare* accumulated about 12.29% of Pb and 55.50% of Cd from the treated soil while *Telfaira occidentalis* accumulated 4.09% of Pb and 13.88% of Cd. Heavy metal content of *Talinum triangulare* obtained from the cultivated and harvested samples were non-significantly higher than their corresponding concentrations obtained from the marketed samples, whereas *Telfaira occidentalis* showed the reverse in heavy metal concentrations. Their values however where within the recommended levels for vegetables nevertheless continuous consumption of these vegetables may lead to serious health challenges.

Keywords: NPK Fertilizers, Heavy metals, Talinum triangulare, Telfaira occidentalis.

INTRODUCTION

Most tropical countries of Africa depend on starch-based food as the main staple food for the supply of both energy and protein. Green leafy vegetables are key components of vitamins, minerals and antioxidants diet. The consumption is increasing gradually, particularly among the urban communities. This is due to increased awareness on their nutritive and herbal values, exposure to other cultures and acquiring proper education (Uwah *et al*., 2009). For instance, natural polyphenols from plant vegetable have been found to exert their beneficial effect by removing free radicals, chelating metal catalyst, activating antioxidant enzymes etc. Evidences from epidermiological studies suggest that high consumption of vegetables is linked to reduced risk of developing most oxidative stress- induced diseases (Dani *et al*., 2008; Wasson *et al*., 2008).

These vegetables are harvested at all stages of growth and fed either as processed, semi-processed or fresh to man while they are usually offered fresh to livestock (Aja *et al*., 2010). Consumers demand for better quality vegetables is also increasing (Itanna, 2003). The perceptions of what is regarded as a better quality are however subjective. Some consumers consider undamaged dark green and big leaves as characteristics of some quality leafy vegetables. However, the external morphology of vegetables cannot guarantee safety from contamination.

Heavy metals rank high amongst the major contaminants of leafy vegetables (Mapanda *et al*., 2005). Zheijazkov and Neilson (1996) found that the concentrations of heavy metals in vegetables per unit dry matter generally follow the order: leaves > fresh fruits > seeds. Although certain heavy metals (Cr, Mn, Ni, Cu and Fe) are essential components for various biological activities within the human body, elevated levels of them can cause numerous health consequences to mankind. In contrast, Pb, Cd, Hg and As are non- essential, toxic elements which are associated with many chronic diseases in human being.

Talinum triangulare (Portulaceae family) is popularly known as water leaf. It is a herb with fleshy green leaves, succulent stem and pink flower. It is well cultivated in West Africa from seeds or by vegetative propagation. Water leaf is employed as a laxative and purgative based on its preparation. *Telfairia occidentalis* (Cucurbtaceae family) is popularly known as fluted pumpkin. It is well cultivated in West Africa and the young shoots and leaves of the plant are the main ingredients of Nigerian soup. Oboh (2004) has reported that dietary intake of the leaf could prevent garlic-induced haemolytic anaemia in rats. The aqueous extracts had been reported to reduce blood glucose level in glucose induced hyperglycaemic Streptozotocin (STZ) induced diabetic mice, while it did not alter the glucose levels in normoglycaemic mice.

Several studies have shown that the main source of fertilizer derived heavy metals in soils is phosphate fertilizer, manufactured from phosphate rocks that contain various metals as minor constituents in the ores (Kpomblekou and Tabatabai, 1994). In Nigeria, NPK fertilizers are the main fertilizers used in the agricultural sector and the current application recommends 3-6 bags of NPK fertilizer application of nutrients (kg/Ha) (FMA and NRA, 1989, 1995). The maximum application rate of this management system is about 0.225g/kg Soil/gr for low fertility soil (where, 1 hectare ~ 2 X 10^6 kg/Soil). This study was designed to determine the heavy metal composition of these vegetables (*Talinum triangulare* and *Telfaira occidentalis*) cultivated and marketed in Aba, Nigeria.

MATERIALS AND METHODS
COLLECTION OF SAMPLES
SOIL SAMPLES

Soil samples were collected from different locations of the Abia State Polytechnic botanical garden Aba. The soil samples were collected by sterile method using plastic auger from a depth of 0-15cm. the auger was washed with water and 70% ethanol before use.

FERTILIZER SAMPLES

Inorganic fertilizers were purchased from different major companies, NPK fertilizer products of Federal fertilizer and chemical company limited, Abakaliki, and Golden Fertilizer Company limited, Kaduna.

The NPK percentage per weight of the inorganic fertilizers was $N_{15}P_{15}K_{15}$, $N_{20}P_{10}K_{10}$ and $N_{27}P_{13}K_{13}$ respectively. These inorganic fertilizers were taken from different parts of the fertilizer bags.

PLANT SEEDS

Seeds of the two vegetables- *Telfairia occidentalis* and *Talinum triangulare* were purchased from the market and authenticated by Mr. Ikeokwu Kingsley of the Ministry of Agriculture, Abia State and Ndukwe Okorie of Department of Biology, Abia State Polytechnic, Aba.

EXPERIMENTAL DESIGN

Two buckets was used to plant each of the vegetables (*T. occidentalis* and *T. triangulare*) on loamy soil treated with inorganic fertilizer at application rate of 0.225g/kg of soil.
The seeds of *T. occidentalis* and *T. triangulare* were planted and allowed to grow for a period of 12 weeks and then harvested.

SAMPLE PROCESSING
SOIL SAMPLES

All the soil samples underwent the pretreatment processes of drying, sorting and sieving in a 2mm mesh. The samples were spread out and air dried at room temperature.

PLANT SAMPLES

The vegetables were harvested at 12 weeks (84 days) after planting. The plugged leaves were washed with distilled water and deionized water, sun dried and ground into uniform powder using electrical blender and then stored in an air tight bottle till required for analysis within 7 days.

PHYSICAL ANALYSIS

Soil particle fractionation, moisture content and soil pH were determined by the method of Nwinuka *et al*. (2003).The determination of organic matter was carried out according to the method of Lu (2000).

CHEMICAL ANALYSIS
SOIL AND FERTILIZER SAMPLES

The total nitrogen, potassium, nitrate and sulphur were determined according to the method described by ASTM (1999). Phosphate was analyzed following the procedure of APHA (1998).

HEAVY METAL DETERMINATION

The samples (soil, fertilizers and the leaves) were digested separately using the method of American Standard for testing materials (ASTM, 1999) as described in Ebong *et al*. (2007) and Ebong *et al*. (2008). Each of the samples (2g) was weighed into a beaker and digested with 20ml of conc. HNO_3 and 10ml of conc. $HClO_4$ on a hot plate with gentle boiling. The

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digested sample was evaporated to dryness and the residue mixed with 10ml of 2M HCl, filtered into a 100ml standard flask using Whatman No 1 filter paper.

Several digested samples were analyzed for the heavy metals of interest-Mn, Bo, Cu, Fe, Mo, Ni, Zn, Mg, Ca, V, As, Cd, Hg, Co, Cr, Ag using Atomic Absorption Spectrophotometer equipped with a computer printout. The samples were analyzed in triplicates. The effectiveness of the methods used was tested by using spiked samples that were later used as reference samples.

RESULTS

TABLE 1: SOIL ANALYSIS

PARAMETERS 1	PARAMETERS 2 (mg/kg)	VALUES
pH		5.90 ± 0.05
Moisture (%)		24.9 ± 0.11
Silt (%)		8.00 ± 0.03
Clay (%)		16.00 ± 0.06
Organic matter		1.76 ± 0.02
	Phosphate (P_2O_5)	300.00 ± 0.32
	Sulphate (SO_4^{2})	20.00 ± 0.15
	Nitrate (NO_4^{3})	93.00 ± 0.24
	Total nitrogen	621.00 ± 0.30
	Potassium (K)	39.00 ± 0.04
	Manganese (Mn)	5.08 ± 0.09
	Boron (Bo)	21.09 ± 0.10
	Copper (Cu)	1.5 ± 0.01
	Iron (Fe)	250.00 ± 0.07
	Molybdenum (Mo)	1.580 ± 1.20
	Nickel (Ni)	22.10 ± 0.08
	Zinc (Zn)	6.66 ± 0.10
	Magnesium (Mg)	20.80 ± 0.01
	Cadmium (Cd)	0.35 ± 0.02
	Lead (Pb)	2.24 ± 0.14
	Cobalt (Co)	8.59 ± 0.05
	Vanadium (V)	BDL
	Arsenic (As)	BDL
	Mercury (Hg)	BDL
	Chromium (Cr)	BDL
	Silver (Ag)	BDL

TABLE 2: FERTILIZER ANALYSIS
HEAVY METAL COMPOSITION OF FERTILIZERS USED IN THIS STUDY

PARAMETERS mg/kg	CANADIAN STANDARD mg/kg	$N_{15}P_{15}K_{15}$	$N_{20}P_{10}K_{10}$	$N_{27}P_{13}K_{13}$
Fe	BDL	11.12± 0.83	19.14 ± 1.61	22.41 ± 3.12
Zn	1850	0.01± 0.01	0.01± 0.01	1.12 ± 0.01
Ni	100	0.01± 0.01	0.01± 0.01	0.01± 0.01
Mn	BDL	1.14 ± 0.10	1.58 ± 0.12	0.01± 0.01
Mo	20	0.01± 0.01	0.01± 0.01	0.01± 0.01
Cd	20	0.01± 0.01	0.01± 0.01	0.01± 0.01
Pb	500	0.20 ± 0.01	0.27 ± 0.08	0.12 ± 0.01
Cr	BDL	0.01± 0.01	0.01± 0.01	0.17 ± 0.02
Co	BDL	0.18 ± 0.01	0.14 ± 0.10	0.22 ± 0.01
Cu	BDL	2.56 ± 0.09	0.01± 0.01	0.12 ± 0.01
Hg	BDL	BDL	BDL	BDL
Ag	BDL	BDL	BDL	BDL
V	BDL	BDL	BDL	BDL
As	BDL	BDL	BDL	BDL

TABLE 3: HEAVY METAL CONTENTS OF *Telfairia occidentalis* HARVESTED VEGETABLE LEAVES (0.225g/kg/Yr Recommendation)

PARAMETERS mg/kg	RMLV	CONTROL SOIL	FERTILIZED SOIL at 2.0g/kg
Fe	425.5	92.0 ± 2.0	154.0 ± 2.64
Zn	99.40	2.60 ± 3.00	5.50 ± 0.59
Cu	73.30	1.22 ± 0.20	1.68 ± 0.81
Ni	67.90	0.05 ± 0.01	1.24 ± 0.15
Cd	0.20	0.02 ± 0.01	0.05 ± 0.02
Pb	0.30	0.06 ± 0.02	0.10 ± 0.03
Mo	BDL	0.10 ± 0.03	0.99 ± 0.11
Co	50.0	0.14 ± 0.03	0.19 ± 0.02
Cr	2.30	BDL	BDL
Hg	BDL	BDL	BDL
Ag	BDL	BDL	BDL
V	BDL	BDL	BDL
As	BDL	BDL	BDL

TABLE 4: HEAVY METAL CONTENT OF *Talinum triangulare* HARVESTED FROM CONTROL AND FERTILIZED SOILS

PARAMETERS mg/kg	CONTROL SOIL	FERTILIZED SOIL
Fe	86.0 ± 1.92	143.33 ± 1.34
Zn	3.16 ± 0.87	9.28 ± 2.01
Cu	1.14± 0.08	1.28 ± 0.99
Ni	0.05 ± 0.01	1.28 ± 0.01
Cd	0.02 ± 0.01	0.20 ± 0.02
Pb	0.07 ± 0.02	0.30 ± 0.02
Mo	0.10 ± 0.01	0.10 ± 0.03
Co	0.12 ± 0.03	0.20 ± 0.01
Cr	BDL	0.10 ± 0.02

TABLE 5: HEAVY METAL CONTENT OF MARKETED VEGETABLE LEAVES

	NEW MARKET		**UMUNGASI MARKET**		
	Telfaira occidentalis	***Talinum triangulare***	***Telfaira occidentalis***	***Talinum triangulare***	**RMLV**
Fe	110 ± 0.17	108 ± 0.02	119 ± 0.14	102 ± 0.25	425.5
Zn	76.1 ± 0.02	26.0 ± 0.77	11.5 ± 0.44	58.8 ± 0.26	99.40
Cu	7.05	9.02	8.86	8.92	73.30
Ni	0.71	1.12	0.98	1.20	67.90
Cd	0.07	0.09	0.8	0.10	0.20
Pb	0.18	0.15	0.08	0.23	0.30
Mo	0.06	0.05	0.07	0.08	BDL
Co	0.10	0.12	0.08	0.18	50.0
Cr	1.28	1.28	0.34	1.62	2.30
Hg	BDL	BDL	BDL	BDL	BDL
Ag	BDL	BDL	BDL	BDL	BDL
V	BDL	BDL	BDL	BDL	BDL
As	BDL	BDL	BDL	BDL	BDL

DISCUSSION

Elemental composition of the soil is shown in Table 1 which reports on the variation of the total heavy metal contents on the top soil (0-15cm) used for the soil treatment study. The data revealed Fe (250.00 ± 0.07) mg/kg as the highest value while V, As, Hg, Cr and Ag were not detected by our analytical method. The mean concentrations (mg/kg) of other metals followed the sequence Ni>Bo>Mg>Co>Zn>Mn>Pb>Mo>Cu>Cd. The soil showed heavy metal contents below the threshold values reported in literature for agricultural soils, therefore establishing the suitability of the soil for planting (Ebong *et al*., 2007).

Elemental composition of the three chemical fertilizers ($N_{15}P_{15}K_{15}$, $N_{20}P_{10}K_{10}$, $N_{27}P_{13}K_{13}$) are given in table 2. The data showed that the concentrations of heavy metals varied sligtly but where within the Canadian maximum acceptable concentrations (Heekmah, 2006). Fe level was the highest whereas V, As, Hg and Ag were below the detection limits of our analytical method. There was no significance difference obtained in the heavy metal values of the three NPK fertilizers. Cd and Pb were detected in the fertilizer samples though at a very low concentrations. Their phyto-availability have urged many investigators to give these toxic elements more attention (McLaughlin *et al*., 1996). Kongshaug *et al* (1992) and Finck (1992) compiled the average heavy metal concentrations in phosphate rocks in different parts of the world. They deduced that the main source of fertilizer derived heavy metals in soils is inorganic fertilizer manufactured from phosphate rocks that contain various metals as minor constituents in the ores.

Heavy metal concentrations in the leaves of vegetables grown with the chemical fertilizer ($N_{15}P_{15}K_{15}$) are shown in Table 3 and 4. The addition of the fertilizers showed an increase in heavy metal contents of the vegetables. This is in agreement with Ayari *et al* (2010) and Yoshide *et al* (2003) who showed that heavy metals and other potentially toxic element contents in soil greatly increase after fertilizer application. The result showed that the vegetables (*Talinum triangulare* and *Telfaira occidentalis*) accumulated significant amount of Iron and Zinc in their leaves than other heavy metals examined. However, Iron and Zinc concentrations (425.50mg/kg and 99.40mg/kg) observed in all the leaf tissues were below the recommended maximum level for vegetables as described by Wiegert (1991). *Telfaira occidentalis* accumulated more Iron in its leaf tissues than in the leaf tissues of *Talinum triangulare*. This could account for the therapeutic relevance of *Telfaira occidentalis* in the treatment of anaemia as reported by Opabode and Adeboye (2005). Subsequently, the accumulation of the non-essential / toxic heavy metals varied among the plant species examined. For example, *Talinum triangulare* accumulated relatively greater amount of Pb and Cd in its leaves than *Telfaira occidentalis*. *Talinum triangulare* accumulated about 12.29% of Pb and 55.50% of Cd from the treated soil while *Telfaira occidentalis* accumulated 4.09% of Pb and 13.88% of Cd. Relatively, the amount of Cd absorbed by the two vegetables was greater than the amount of Pb absorbed. In view of this property *Talinum triangulare* could be recommended for phytoremediation application especially of Cd. Secondly, the relative lack of selectively in transmembrane ion transport may also explain why non-essential heavy metals can enter cells, even against a concentration gradient. This observation is not surprising since tight binding of Pb to soils and plant materials, are at least partially explains the relatively low mobility of this metal (Alloway and Ayres, 1999). The heavy metal composition of the vegetables purchased from the markets was shown in Table 5. Iron and Zinc concentrations were higher than other heavy metals examined. Heavy metal content of *Talinum triangulare* obtained from the cultivated and harvested samples were non-significantly higher than their corresponding concentrations obtained from the market purchased samples, whereas *Telfaira occidentalis* showed higher concentration in the purchased samples than the cultivated and harvested samples. This result may suggest that *Telfaira occidentalis* purchased from the market probably was planted in a heavy metal contaminated soil presenting higher potential public health hazard.

The practice of growing vegetables using fertilizers especially inorganic fertilizers is aimed at increasing agricultural yield. This practice may not be safe and sustainable in long-term if abused coupled with the influx of heavy metals from other sources. The health implications however depend on the dietary pattern of the consumers. The average amount of vegetables consumed per day by a person in most of the developing countries in Africa is less than the international daily average of 50g for leafy vegetables (Itanna, 2003). It is because of this that the intake of heavy metals from plant food constitutes much less than the theoretical maximum daily intakes which are used to express the exposure of consumers and associated health risk.

CONCLUSION

The practice of growing crops using fertilizers especially inorganic fertilizers is aimed at increasing agricultural yields. Although these applications are of great socio-economic benefits, the practice may not be safe and sustainable in the long-term if abused.

RECOMMENDATION

From this research, *Talinum triangulare* was able to extract significantly ($p<0.05$) Cd from the treated soils. In view of this, improvement of its phytoremediation quality should be the major target of biotechnology. Furthermore, the evaluation of total dietary consumption rate for urban and rural dwellers is strongly recommended in future research for human toxicity assessment.

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