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Evaluation of concentration of heavy metals in leaf tissues of three improved varieties of *Manihot* esculenta crantz.

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The bioaccumulating ability of three varieties of cassava (*Manihot esculenta* Crantz) was studied under normal field conditions using perforated polythene bags. The varieties used in this study were TMS/30572, TMS/97/0505, TMS/96/0581. The heavy metals investigated were Cadmium (Cd), Chromium (Cr), Iron (Fe), Lead (Pb) and Zinc (Zn). Leave samples were collected from the plants twelve weeks after planting (12 WAP) prepared by dry ashing before analysis. The study revealed that heavy metals were significant (P<0.05) in the plant samples (TMS/30572, TMS/97/0505, TMS/96/0581). The least and highest concentrations recorded were <0.01 mg kg⁻¹ of Cd in TMS/30572 and 98.55 mg kg⁻¹ of Fe in the same variety. Generally, the heavy metals content of these plants is relatively high when compared with standard recommendations. The result of this work has practical applications in environmental science, health management and crop improvement.

Keywords: Manihot esculenta, heavy metal, soil, bioaccumulating.

INTRODUCTION

Cassava (Manihot esculenta Crantz) from the family Euphorbiaceae, being an all season crop in several parts of Africa (Nigeria inclusive), Asia and Latin America is well documented (Longe, 1980; Rosling, 1987; Bradbury et al., 1991). It is the third most important food source in the tropical world after rice, wheat and maize and provides calories for over 160 million people in Africa (Polsen and Spencer, 1991). Its food value is greatly compromised by the endogenous presence cyanogenic glycosides. It has been reported that apart from lower methionine, lysine and perhaps isoleucine content, the amino acid profile of cassava leaf proteins compares favorably with those of milk, cheese, soybean, fish and eqg (Rayindran, 1990; Wanapat, 2001). Tender leaves of cassava which could be harvested periodically throughout the growing season are utilized in some areas as relish, particularly during the dry season, when there are few leafy vegetables (IITA, 1990). In addition, leaf meal could also be prepared from cassava leaves as a component of livestock feed (Fasuyi, 2005).

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Nutritionally, the cassava leaf is rich in protein (14-40 %), potassium, iron, calcium, sodium, vitamin B1, B2, B6, C and carotenes (Eggum, 1970; Adewusi and Bradbury, 1993; Bokanga, 1994). Nonetheless, full exploitation of the nutritional, medicinal and fodder values of this crop is greatly compromised by the presence of substantial quantities of heavy metal in its tuber and above ground tissues especially when cultivated in a contaminated environment.

These metals contaminant make a significant contribution to environmental pollution and comes as a result of human activities such as electroplating, energy and fuel production, power transmission, intensive agriculture, sludge dumping and military operations, burning of fossil fuels, the mining and smelting of metalliferous ores, municipal wastes, fertilizers, pesticides and sewage (Nedelkoska and Doran, 2000).

Toxic metal contamination of soil, aqueous waste streams and groundwater pose a major environmental and human health problem, which is still in need of effective and affordable technological solutions (Kabata-Pendias and Pendias, 1989, Zeller and Feller, 1999). Heavy metals such as Cu and Zn are essential for normal plant growth and development since they are constituents

Table 1a: Physicochemical data obtained from analysis of experimental soil.

рН	%C	%N	P (mg kg ⁻¹)	Soil Textural class
6.51 <u>+</u> 0.03	1.98 <u>+</u> 0.23	0.06 <u>+</u> 1.34	39.99 <u>+</u> 1.40	Sandy Loam
Values are me	eans ± S.E.M.	of 3replicates.	pH = 1:1 (soil:	water)

Table 1b: Physico-chemical data obtained for analysis of experimental soil.

Na	K	Ca	Mg	H⁺	E.C.E.C	Cd	Fe	Pb	Zn	Cr	Ni
			(c mol kg ⁻¹)	(c mol kg ⁻¹)				(c mol kg ⁻¹)			
0.06 <u>+</u> 0.13	0.09 <u>+</u> 0.10	3.25 <u>+</u> 0.44	1.30 <u>+</u> 1.10	2.6 <u>+</u> 1.11	7.32 <u>+</u> 0.43	0.01 <u>+</u> 0.03	15.20 <u>+</u> 0.15	1.92 <u>+</u> 0.11	1.76 <u>+</u> 0.00	1.18 <u>+</u> 0.81	1.23 <u>+</u> 0.23

Values are means ± S.E.M. of 3replicates.

of many enzymes and other proteins. However, elevated concentrations of both essential and nonessential heavy metals in the soil can lead to toxicity symptoms and growth inhibition in most plants (Hall, 2002). Heavy metals are known to be non-biodegradable and persistent and may be deposited on the surfaces and then absorbed into the tissues of plants. Plants growing within the heavy metals contaminated areas usually take up heavy metals by absorbing minute deposits on the parts of the plants exposed to the air in the polluted environments and during nutrient uptake from contaminated soils (Zurayk *et al.*, 2001).

It has been reported that some plant species (hyperaccumulators) can absorb very high concentrations of toxic metals upto levels which far exceed that of the soil (Baker and Brooks, 1989). However, excessive accumulation of these heavy metals in plant tissues can be toxic to plants and other organisms. Also, the ability to tolerate heavy metals vary with plant species and varieties (Ernst *et al.*, 1992). This present work aims to study the bioaccumulating potential of three (3) varieties of *Manihot esculenta* with regard to the six (6) heavy metals studied, using their leaf tissues.

MATERIALS AND METHODS

Sites Description and Samples Collection

The soil used in the study was collected as a composite sample of top soil (0-20 cm depth), in the Department of Botany and Ecological Studies' University of Uyo Postgraduate Research Farm, Uyo. Data of the physicochemical properties of this soil is presented in Table 1(*a* and *b*).

The cassava leaves were harvested from the three varieties and grouped separatly. From each variety, 18 young fresh leaves were selected for heavy metals analysis. Three groups based on the variety i.e. Group1= harvested leaves of TMS/30572, Group 2 = harvested leaves of TMS/97/0505 and Group 3 = harvested leaves

of TMS/98/0581. Each group consisted of 18 leaves, which were randomly divided into three subgroups so as to give three determinations per group, during the analysis.

Sample Preparation

The leaf samples in each of the three groups were air dried to remove the moisture then kept in an oven at 65 °C to get a constant weight, and pulverized to fine powder using a laboratory grinder. 3.0 g of each sample was carefully weighed into clean platinum crucible and ashed at 450 - 500 °C then cooled to room temperature in a desiccator. The ash was dissolved in 5 ml of 20 % hydrochloric acid and the solution was carefully transferred into a 100ml volumetric flask. The crucible was well rinsed with distilled water and transferred to the flask and made up to the mark with distilled water and shaken to mix well. The resulting sample solutions were then taken for the determination of the heavy metal concentrations. The samples from each group were analyzed in three replicates.

Sample Analysis

The determination of the heavy metals (Pb, Ni, Cr, Fe, Cd, and Zn) content and physico-chemical properties (pH, E.C.E.C., Na, K, Ca, Mg, H⁺, %C and %N etc.) of the plant and soil samples respectively was carried out according to the standard procedures of the AOAC (2005) on dry samples. The pH of the soil was measured using pH meter.

Statistical Analysis

Analysis using two-way analysis of variance (ANOVA) test was carried out to examine the statistical significance of difference in the mean concentration of metals

Table 2: Mean concentration of heavy metals in leaves of three varieties of Cassava (mg kg⁻¹).

Group	Cd	Cr	Fe	Ni	Pb	Zn
TMS/30572	<0.01 <u>+</u> 0.23	0.30 <u>+</u> 0.11	98.55 <u>+</u> 0.21	6.39 <u>+</u> 0.31	8.36 <u>+</u> 0.11	18.24 <u>+</u> 0.33
TMS/97/0505	0.01 <u>+</u> 0.03	0.18 <u>+</u> 0.52	79.46 <u>+</u> 0.11	4.87 <u>+</u> 0.22	9.73 <u>+</u> 0.51	15.92 <u>+</u> 0.42
TMS/98/0581	0.03 <u>+</u> 0.20	0.23 <u>+</u> 0.01	81.57 <u>+</u> 0.22	6.22 <u>+</u> 0.04	6.98 <u>+</u> 0.71	15.84 <u>+</u> 0.81

Values are means ± S.E.M. of 3replicates.

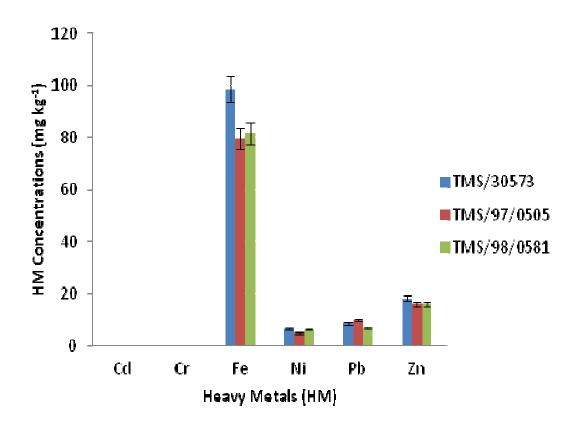


Figure 1: Mean concentration of heavy metals in leaves of three varieties of Cassava (mg kg⁻¹).

between varieties of *Manihot esculenta* using SPSS, version 11. A probability level of P<0.05 was considered statistically significant.

RESULTS

The physicochemical properties of the experimental soil have been stated on Table 1a and b. Mean concentrations of heavy metals (Pb, Ni, Cr, Mn, Cd, and Zn) obtained for the cassava leaf samples from the three common varieties are summarized in Figure 1, Table 2. The bioconcentration values of these heavy metals in the three species varied from trace to high. The variety TMS/30752 had the lowest concentration of Cd (<0.01

mg kg⁻¹) and it also accumulated the highest concentration of 98.55 mg kg⁻¹ of Fe. **Lead (Pb)**

The concentration value of lead in the plant samples obtained from the three varieties ranged between 6.98 mg kg⁻¹ in TMS/98/0581, 8.36 mg kg⁻¹ in TMS/30752 and 9.73 mg kg⁻¹ in TMS/97/0505.

Zinc (Zn)

Zinc is one of the heavy metals which is highly accumulated in this study. The concentration values ranged between 15.84 mg kg⁻¹ in TMS/98/0581, 15.92

mg kg⁻¹ in TMS/97/0505 and 18.24 mg kg⁻¹ in TMS/30572.

Iron (Fe)

The phytoavailability of Iron in the samples studied was higher than that of the other five (5) heavy metals. The accumulation was up to 79.46 mg kg⁻¹ in TMS/97/0505, 81.57 mg kg⁻¹ in TMS/98/0581 and 98.55 mg kg⁻¹ in TMS/30572.

Nickel (Ni)

From the least Nickel accumulator to the highest, the concentration ranged from 4.87 mg kg⁻¹ in TMS/97/0505, 6.22 mg kg⁻¹ in TMS/98/0581 and 6.39 mg kg⁻¹ in TMS/30572.

Chromium (Cr)

The Chromium content of leaf samples of the varieties was low compared to that of Iron and Zinc. The bioconcentration values of this trace element ranged between 0.18 mg kg⁻¹ in TMS/97/0505, 0.23 mg kg⁻¹ in TMS/98/0581 and 0.30 mg kg⁻¹ in TMS/30572.

Cadmium (Cd)

Cadmium was sparingly present in the samples studied. Its phytoavailability ranged between <0.01 mg kg⁻¹ in TMS/30572, 0.01 mg kg⁻¹ in TMS/97/0505 and 0.03 mg kg⁻¹ in TMS/98/30572.

DISCUSSION

The three varieties studied slightly differ in their accumulation of heavy metals thus verifying the reports of Zurayk (2001) that species or varieties of a species differ in their metal tolerance and accumulation. Generally, the levels of these metals present in the plant leaves, apart from those of Cadmium were observed to be significantly higher than the permissible levels given by the FAO and WHO. The results indicate the fact that the plants studied hyper-accumulators since the heavy concentrations in the plant is greater than that in the soil. This work supports the report that plants (especially those from urban districts) have the potential of absorbing metal contaminants from their environment (i.e. not only soil, but also from air and water) and concentrating them in their above ground tissue to the extent that the tissue concentration far exceeds that of the soil (Kabata-Pendias and Pendias, 1989). This has serious ecological and health implications. This is in line with the results of

the studies carried out by Yusuf et al. (2002) on vegetables from industrial areas of Lagos City, Nigeria, Othman et al. (2002) on edible portions of five varieties of green vegetables collected from several areas in Dar Es Salaam, Africa, as well as Nirmal Kumar et al. (2004) on vegetable plants and its parts collected from organic farms and village agriculture fields around Anand province, Gujarat. Also, Jassir et al. (2005) have reported elevated levels of heavy metals in vegetables sold in the markets at Riyadh city in Saudi Arabia due to atmospheric deposition. And recently, Sharma et al. (2008a, b) and Zurayk, (2001) reported that atmospheric depositions could significantly elevate the levels of heavy metals contamination in plants. With this in mind, it is worthy to note that prolonged consumption of unsafe concentrations of heavy metals through food may lead to their chronic accumulation which hinder proper the functioning of the kidney and liver of humans thereby causing disruption of numerous biochemical processes. leading to cardiovascular, nervous, kidney and bone diseases (Jarup, 2003, WHO, 1995, Steenland and Bufetta, 2000, Radwan and Salama, 2006). Zn acts as micronutrient for the growth of animals and human beings when present in trace quantities, whereas others such as Cd and Cr act as carcinogens (Feig et al., 1994 and Trichoupulos, 1997.) and Pb is associated with the development of abnormalities in children (Hartwig, 1998) and Saplakog¢lu and Iscan, 1997).

The results of this study imply that large daily intake or usage of medicine, fodder or food material made from tissues of varieties such as these, grown under similar environmental conditions, might constitute health hazard to the consumer(s). Therefore, it is recommended that, varieties such as TMS/30572, TMS/97/0505 and TMS/98/0581 should not be cultivated near refuge dumpsites, traffic congestions, or other pollution prone areas in order to reduce incidence of heavy metal poisoning and its complications.

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