Evaluation ofselected metals contents of*Brassica oleraceae*(cabbage), *Solanum tuberosum*(Irishpotatoes) and *Daucus carota* (carrot) grown at irrigation farms of Jos, Nigeria

ABSTRACT

Aim: This study assessed levels of Cd, Pb, Fe, Zn and As in *Brassica oleraceae* (cabbage), *Solanum tuberosum* (Irish potatoes) and *Daucus carota* (carrot). Six samples were collected from each of the two irrigation farms. Samples were analysed using inductively coupled plasma mass spectrometry (ICP-MS).

Study Design: Six (6) samples of each vegetables, *Brassica oleracea* (cabbage), *Solanum tuberosum* (Irish potatoes) and *Daucus carota* (carrot) were obtained from irrigation farms situated at *Lamingo* and *Russau.* On each farm, 2 samples were collected thereby giving a total sample size of 36.

Place and Duration of study: Department of Biochemistry Research Laboratory between February and April 2021.

Method: Turkey-Kramer multiple comparison tests in a one-way analysis of variance was used to analyse the data obtained.

Results:Samples of cabbage from ***Russau*** showed mean values for Zn, Pb and Fe levels thus: 32.80±0.62, 0.27±0.02, and 20±0.06 respectively. Samples from ***Lamingo*** had mean levels of Zn, Pb and Fe thus: 23.30±0.26, 0.07±0.02, and 0.08±0.03 in that order. The concentrations of Pb, Zn were above standard values *(P=0.05)* but Fe was not significant. The machine did not detect arsenic (As). Samples of cabbage from ***Russau*** contained higher levels of all the metals than those from ***Lamingo***. In Irish potatoes, samples from ***Lamingo*** had mean levels of Pb, Cd, Zn and Fe as follows: 0.188±0.017, 0.36±0.037 16.105±4.16 and 0.082 respectively. The levels were significantly above *(P=.05)* standard values. In Carrots, levels of the metals in samples from ***Lamingo*** were 0.15±0.015, 0.14±0.015 and 0.88±0.018 respectively for Fe, Cd and Pb. Similarly, samples from ***Russau*** had 0.14±0.016, 0.20±0.022 and 1.04±0.031 respectively for Fe, Cd and Pb.

Conclusion: Samples, especially those from *Russau,* contain zinc but they also contain cadmium, lead and iron which can predispose to their toxic effects.

Key Words: Cadmium, Lead, Zinc, Iron, Irrigation water, *Russau*, *Lamingo*

Introduction

“Heavy metal refers to metallic chemical element that has a relatively high density. Most heavy metals are toxic even at low concentrations” [1]. “Heavy metals are dangerous because they bio-accumulate in plants and water and hence get into humans through the food chain. The source of metals in aquatic environment is from natural or anthropogenic sources”[2].“Natural sources may include volcanic activity, continental weathering and forest fires, while anthropogenic sources may include industrial effluents, urban storm, water runoff, leaching of metals from garbage and solid dump, metal input from rural area. The contamination of soil, water and air with heavy metals even at low concentration are known to have potential impact on the environment and human health”[3]. This is because this ultimately gets inhaled or ingested in the form of food or drinking water.

“One of the major mechanisms associated with heavy metals toxicity has been attributed to generation of reactiveoxygen and nitrogen species, which develops imbalance between the prooxidant elements and the antioxidants in the body. In this process, a shift to the former is termed oxidative stress. Oxidative stress mediated toxicity of heavy metals involves damage primarily to liver (hepatotoxicity), central nervous system (neurotoxicity), DNA (genotoxicity), and kidney (nephrotoxicity) in animals and humans. They are reported to impact signaling cascade and associated factors leading to apoptosis”[4].

Background:“Heavy metals uptake in soils is of increasing concern due to food safety issues and potential health risks, as well as detrimental effects on soil ecosystems. Sources of these elements in soils mainly include natural occurrences derived from parent materials, volcanic eruptions, marine aerosols, forest fires, and human activities”[5]. “The anthropogenic sources of heavy metals include traffic emissions (vehicle exhaust particles, tire wear particles, weathered street surface particles, brake lining wear particles), industrial emissions (power plants, coal combustion, metallurgical industry, auto repair shops, chemical plants, etc.), domestic emissions, and weathering of building and pavement surfaces”[6][7]&[8]. “Heavy metal pollution of soil can result not only in decreased crop output and quality and hurt human health through the food chain, but also further deterioration of air and water environmental quality” [9][10] & [11].“Studies of heavy metal uptake by plants have often revealed their accumulation at a level toxic to humanhealth” [12]. “Generally, uptake is increased in plants that are grown in areas with increased soil contamination. Among the metals, Cd and Zn are fairly mobile and readily absorbed by plants” [13].

MATERIALS AND METHODS

Materials

Equipment, Chemicals and Reagents

Inductively coupled plasma mass spectrometry (ICP-MS), Trioxontrate (v) acid (HNO_3), hydrogen chloride **(**HCl), deionized and hot water. All reagents and chemicals used in this study were of analytical grade.

Methods

Collection of Samples

Vegetables used were *Brassica oleracea*(cabbage), *Solanum tuberosum* (Irish potatoes) and *Daucus carota* (carrot) which are commonly sold in local markets of the study areas. Vegetable samples were collected from two irrigation farms at ***Lamingo*** and ***Russau*** during period of harvest. They were randomly collected from two farms within each location. Samples obtained were wrapped in labeled polythene bags and immediately taken to the laboratory and washed with tap water to remove any soil particles and rinsed three times. The edible parts of the vegetable samples were air-dried for fifteen hours. Thereafter, they were dried in an oven set at 80^0C for two consecutive days until dry. The dried samples were then pulverised using pestle and mortar, sieved with muslin cloth and stored in air-tight transparent polythene and transported via DHL to ACME laboratory, Vancouver, Canada for analysis.

Digestion of samples

Plant samples were digested according to the method and protocol described by Acme laboratory. Pulverised samples were cold-leached with nitric acid and then digested on hot plate with intermittent addition of digestion mixture which contained HCl, $HNO_3$and deionised water in equal proportion.After obtaining a persistent white precipitate, samples were diluted to mark with deionised water and HCl in combination and then filtered. This digestion procedure was carried out in Acme laboratory, Vancouver, Canada.

Measurement of Heavy Metals

The heavy metals in the digested samples were determined using PerkinElmer ELAN 9000 Inductively Coupled Plasma Mass Spectrometry (ICP- MS)instrument in ACME Laboratory, Canada.

Statistics

Results obtained were analysed using inSta3 software. One Way ANOVA was used. $P = 0.05$ was considered significant.

RESULTS

Table 1: Mean concentration of selected heavy metals in ***Brassica oleracea* (cabbage)** obtained from irrigated farms at***Lamingo.***

	Metal concentration(mg/litre)			
Sample	Pb	Cd	Zn	Fe
Lamingo	0.07±0.02	0.12±0.02	23.30±0.26	0.08±0.03
STD V16 Observed	3.08	0.09	39.00	3.13
STD V16 Expected	2.80	0.089	39.20	4.04
%Recovery	110	104.65	99.49	77.48
WHO(2001)	0.30	0.10	20.00	0.30

Values are means of triplicate determinations; (±SEM). n=6

Table 1 shows the mean concentration of Pb, Cd, As, Zn and Fe.The certified reference material (CRM) used - STD V16, had % recovery greater than 86and hence the method used was efficient. Mean concentrations of zinc and cadmium, 23.3and 0.12 respectively were significantly *(P=.05)*above the [14] permissible limits.

Table 2: Mean concentration of selected heavy metals in ***Brassica oleracea* (cabbage)** obtained from irrigated farms at***Russau.***

Metal concentration(mg/litre)

Sample	Pb	Cd	Zn	Fe
Russau	0.27±0.02	0.10±0.04	32.80±0.62	0.20±0.06
STD CDV-1 Observed	1.04	0.04	24.00	0.254
STD CDV-1 Expected	1.00	0.04	23.30	0.256
%Recovery	104.00	100.00	103.00	99.23
FAO/WHO(2001)	0.30	0.10	20.00	0.30

Values are means of triplicate determinations (±SEM); n= 6.

The mean concentrations of metals in cabbage obtained from Russau are presented in Table 2. Concentration of Zn was significantly *(P=.05)*above standard values; Cd concentration in samples was similar with the standard values. Levels of Pb and Fe were below the reference values.

Table 3: Mean concentration of selected heavy metals in ***Solanum tuberosum*** **(Irish potatoes)** samples obtained from irrigated farms at ***Lamingo.***

	Heavy Metals Concentration (mg/litre)			
Samples	Pb	Cd	Zn	Fe
Lamingo	0.188±0.017	0.357±0.037	16.105±4.158	0.082±0.003
STD CDV-1 Observed	1.04	0.04	24	0.254
STD CDV-1 Expected	1	0.04	23.3	0.256
%Recovery	104	100	103.00	99.21
WHO	0.10	0.10	20.00	0.30

Values are means of triplicate determinations (±SEM); n= 6.

Levels of Cd *(P=.05)* and Pb were above the reference values. Arsenic was not detected by the equipment. The levels of Zn and Fe were below standard values.

Table 4: Mean concentration of selected metals in ***Solanum tuberosum*** **(Irish potatoes)**obtained fromirrigated farms at***Russau***.

Heavy Metals Concentration (mg/litre)

Samples	Pb	Cd	Zn	Fe
Russau	0.88±0.017	0.14±0.037	16.105±4.158	0.082±0.003
STD CDV-1 Observed	1.04	0.04	24	0.254
STD CDV-1 Expected	1	0.04	23.3	0.256
%Recovery	104	100	103.00	99.21
WHO	0.10	0.10	20.00	0.30

Values are means of triplicate determinations (±SEM); n= 6.

Both Pb and Cd levels were above *(P=.05)* safe limits. Levels of zinc and Fe were each below reference values. Arsenic was not detected by machine used.

Table 5: Mean concentration of selected metals in ***Daucus carota*** **(carrot)** samplesobtained from irrigated farm at ***Lamingo***

	Pb	Cd	Fe
Sample	Metal concentration in Mg/litre		
Lamingo	0.88±0.180.1±.15		0.15±0.015
STD. CDV-1 Observed	1.04	0.04	0.254
STD. CDV-1 Expected	1	0.04	0.256
% RECOVERY	84.62	100	99.2
WHO	0.10	0.14	0.15

Values are means of triplicate determinations (±SEM); n= 6.

It is seen that Pb had values above *(P=.05)* the safe permissible limits, while Fe and Cd had values that were not significantly different from the permissible safe limits.

Table 6: Mean concentration of selected metals in ***Daucus carota*** **(carrot)** samples obtained from irrigated farms at ***Russau.***

	Metal concentration in Mg/litre		
SAMPLE	Pb	Cd	Fe
Russau	1.04±0.03	0.20±.02	0.14±.06
STD. CDV-1 Observed	3.08	0 .09	0.313
STD. CDV-16 Expected	2.8	0.09	0.40
%RECOVERY	110	104.7	77.50
WHO	0.1	0.1	0.15

Values are means of triplicate determinations (±SEM); n= 6.

Levels of Cd and Pb were above *(P=.05)* reference values whereas Fe was below the reference values. Arsenic was not detected by the equipment used.

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DISCUSSION

This work sought to determine the levels ofPb, Cd, As, Zn and Fe) in *Brassica oleracea*(cabbage), *Solanum tuberosum* (Irish potatoes) and *Daucus carota* (carrot) cultivated at *Russau*and*Lamingo* by irrigation. Levels ofall the heavy metals were found to be above *(P=.05)* reference values with Pb being the least in magnitude; Arsenic was not detected. In order of magnitude, the mean concentration of the metalsin cabbage from *Lamingo* was Zn>Cd>Fe>Pb; in samples from *Russau,* the trend was: Zn>Pb>Fe,>Cd.Samples of Irish potatoes from *Lamingo* had this trend: Zn>Cd>Pb>Fe, those from *Russau* had Zn>Pb>Fe>Cd. As for carrots, samples from*Lamingo*had Pb>Fe>Cd whereas trend for samples from *Russau*had Pb>Cd>Fe.

"The human body contains 2–3 g (mmol) zinc, and nearly 90% is found in muscle and bone"[15]. "Other organs containing estimable concentrations of zinc include prostate, liver, the gastrointestinal tract, kidney, skin, lung, brain, heart, and pancreas"[16] [17] & [18]. "Oral uptake of zinc leads to absorption throughout the small intestine and distribution subsequently occurs via the serum, where it predominately exist bound to several proteins such as albumin, α-micro globulin, and transferrin"[19]. "At the cellular level, 30–40% of zinc is localized in the nucleus, 50% in the cytosol and the remaining part is associated with membranes"[20]. "Cellular zinc underlies an efficient homeostatic control that avoids accumulation of zinc in excess. The cellular homeostasis of zinc is mediated by two protein families; the zinc-importer (Zip; Zrt-, Irt-like proteins) family, containing 14 proteins that transport zinc into the cytosol, and the zinc transporter (ZnT) family, comprising 10 proteins transporting zinc out of the cytosol"[21].

From tables 1 & 2, mean level of zinc (ppm) in *Brassica oleracea*(cabbage) ranged from 23.30 (*Lamingo*) to 32.80 (*Russau*). The area of study is a plateau; therefore since *Lamingo* is situated uphill, waste water, spillage of fuel and oil from mechanic sites, lead from battery works and sludge (waste) from industries and other sources readily flow downhill. The irrigation farms are situated downhill. Hence, zinc content in samples from *Russau* is higher due to usage of contaminated water that flows downhill from *Lamingo* (uphill). "The human body contains 2 g of zinc. It is available within cells in contrast to iron, which is contained in defined cellular components and has defined physiological roles. The role of zinc in biology can be grouped into three general functional classes, namely catalytic, structural and regulatory functions"[22]. "Zinc absorption is concentration-dependent and increases with increasing dietary zinc up to a maximum rate. In addition, zinc status may influence zinc absorption. Zinc-deprived humans absorb this element with increased efficiency, whereas humans on a high-zinc diet show a reduced efficiency of absorption.It is an important cofactor in the body and is essential for normal function; however, increased levels can become toxic through inhalation, oral, and dermal. Most cases are acute toxicity"[23].

"The trend for iron was similar with content of 0.08 ppm (*Lamingo*) relative the content of 0.20ppm (*Russau*).Iron is a most crucial element for growth and survival of almost all living organisms"[24]. "It is one of the vital components of organisms like algae and of enzymes such as cytochromes and catalase, as well as of oxygen transporting proteins, such as hemoglobin and myoglobin"[25]. "Iron is an attractive transition metal for various biological redox processes due to its inter-conversion between ferrous (Fe^{2+}) and ferric (Fe^{3+}) ions"[26]. "The source of iron in surface water is anthropogenic and is related to mining activities. The production of sulphuric acid and the discharge of ferrous (Fe^{2+}) take place dueto oxidation of iron pyrites (FeS_2) that are common in coal seams"[24].

In the case of Cd, levels were however almost the same for both locations. But in the case of Pb, levels in samples from *Russau* (downhill) were higher than those from *Lamingo*(uphill). The concentration of arsenate in samples was not detected by the machine. Cadmium is very toxic finding its way through the food chain. Application of pesticides and phosphate fertilizers leads to dispersion of Cd [27]. It presents a serious hazard to public health and is a threat to most life forms [28]. WHO standards for Cd in vegetables are 0.1 mg/kg [14]. Metal trace elements, including Cd, cause severe damage at each level in living organisms, from populations and communities to cell elements [29]. In this study, Cd has been found in all the sampling sites (Table 1 and 2), it could be due to use of chicken manure, phosphate fertilizer and metals-based pesticides.

It appears leafy vegetables (spinach, broad leaved mustard), have the better absorption capacity to heavy metal Cd [30]. The kidney is the main organ affected by chronic Cd exposure and toxicity. Cd accumulates in the kidney as a result of its preferential uptake by receptor-mediated endocytosis of freely filtered and metallothionein bound Cd (Cd-MT) in the renal proximal tubule. Internalized Cd-MT is degraded in endosomes and lysosomes, releasing free Cd into the cytosol, where it can generate reactive oxygen species (ROS) and activate cell death pathways.

From Table 2, it can be seen that *Brassica oleracea*(cabbage) from *Russau* tend to accumulate heavy metals exceeding or equaling the standard values of [14].Lead was found elevated in all the study areas. Lead is a leading toxic element in that it is a multi-target toxicant, causing effects in the gastrointestinal tract, hematopoietic system, cardiovascular system, central and peripheral nervous systems, kidneys, immune system, and reproductive system [31]. [32] reported that 3-15 fold higher concentrations of Pb in wastewater irrigated area of Kanpur and Varanasi as compared to those reported in the present study. A maximum Pb limit for human health has been established for edible parts of vegetables at 0.3 mg/kg [14] but this limit in China is 0.2 mg/kg. It is only 10% less than the limit; this is already a potential signal to impending danger because lead is a multi-target toxicant, causing effects in the gastrointestinal tract, hematopoietic system, cardiovascular system, central and peripheral nervous systems, kidneys, immune system, and reproductive system [30]. Zinc was found to exceed the limits by 64% this can impact toxic effect to consumers especially zinc induced copper deficiency. Zinc is an essential element in humans however elevated levels in the body can lead to toxicity.

Lead and cadmium unlike zinc and iron are non-essential to the human body therefore even small quantities in the body can pose toxic effects this is because they lack any biological function and the body is not able to excrete them leading to bioaccumulation. The higher contamination of heavy metals in cabbage from Russau as compared to those of *Lamingo* could be attributed to the fact that Russau is located where there is high anthropogenic activities such as industrial runoffs and effluents, waste from mechanic workshops and so on whose activity we believe has great impact on the contamination of the water source in *Russau* and as such the bioaccumulation of heavy metals in vegetables.

In tables 3 & 4,*Solanum tuberosum* (Irish potatoes) levels of Pb in samples from *Russau* were higher than those from *Lamingo*with levels in samples from both locations having values higher *(P=.05)* values relative value of 0.10ppm by WHO. As for Zn, Fe and Cd, levels in all samples from *Lamingo* were however lower than those from *Russau.*

Results in tables 5 & 6, were those of *Daucus carota* (carrot). Concentration of Pb and Cd in samples from both *Lamingo* and *Russau* indicated higher levels of both metals in samples from *Lamingo* to be higher *(P=.05)* relative WHO standards. The results revealed that the vegetables contained significant concentrations of Cd, Pb. They also contain Zn and Fe. This study shows that cabbage is a very efficient bioaccumulator of all the metals analysed which enter the body of eaters through the food chain. Further, of three vegetables, the most commonly eaten is cabbage. Its level in the samples was significant. Magnitude of the metal concentrations appeared to be location-dependent.

Conclusion

The variation of these metals in the vegetables from these areas may be attributed to anthropogenic activities. Dietary intake of foods with high concentration of heavy metals results in long-term low level bioaccumulation and the detrimental impact becomes apparent only after several years of exposure. Thus regular monitoring and screening of these vegetables for toxic heavy metals is imperative to prevent their excessive build-up in the food chain. The government may wish to ban irrigation activities downhill (*Russau*) due to alarming levels of heavy metals in samples from there.

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