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Toxic Metals Uptake by Spinach (*Spinacea oleracea*) and Lettuce (*Lactuca sativa*) Cultivated in Sokoto: A Comparative Study

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Abstract: Potentially toxic metals (Pb, Fe, Mn, Cr, Cd, Zn and Cu) concentration in selected vegetables namely; lettuce *(Lactuca sativa)* and spinach (*Spinacea oleracea*) was investigated alongside their harvesting site soil samples using Atomic Absorption Spectroscopy (AAS). Some physicochemical parameters viz; pH, moisture and ash contents of the plant were also determined. The estimated metal concentrations for the vegetables ranges from 1.59-2.07 ug/g, 211-364 ug/g, 7.0-7.5 ug/g, 5.00-5.25 ug/g, 0.01 ug/g, 20.4-24.12 ug/g, 0.8-1.1 ug/g for Pb, Fe, Mn, Cr, Cd, Zn, Cu respectively. The result showed that soils in industrial site are higher in heavy metals and that vegetable grown in the industrial sites is considerably higher in metal content than those grown in normal agricultural soils. It is also observed that vegetables differ in their ability for heavy metal uptake. Highest metal concentrations were observed in lettuce and lowest in spinach.

Key words: Heavy metals, spinach, Lettuce, Kalambaina

INTRODUCTION

The presence of heavy metals in the environment has been the subject of public concern in the recent years. Some of these metals are present in low levels in plant and animal tissues where they play important roles. Such category of heavy metals is also called trace elements or the newer term ultra trace element. Human activities have altered the biochemical and geochemical cycle and level of most heavy metals. Since they are stable and cannot be degraded, they tend to build up in atmosphere, soil, sediments and water. Excessive levels of heavy metals can be harmful to living organisms, even when they are essential for good health (Ojeka and Ayodele, 1997).

Human exposures to heavy metals occur primarily through inhalation of air and ingestion of food and water. The concentration of these metals in the environment varies considerably depending upon factors such as difference in the soil concentration and proximity to sources of emission. The heavy metals of great concern for general population are those which are present almost everywhere in the environment e.g. lead, chromium, copper, iron, zinc manganese etc. (Kenneth and Harvey, 1979).

Vegetables are plants that are eaten raw or cooked. It is described as shrubs or herbaceous annual or perennial plants that are eaten by man (Longman, 2005). There are different kinds of vegetables ranging from edible roots, stems, leaves, fruits or seeds. Each group contributes to diet in its own way (Hanif *et al*., 2006). The utilization of vegetables is part of Africa's cultural heritage and they play important roles in the customs, traditions and food culture of the African household. Nigeria is endowed with variety of traditional vegetables and different types consumed by the various ethnic groups for different reasons (Mensah *et al*., 2008).

Lettuce (*Lactuca sativa*) is a temperate annual or biennial plant of the daisy family asteracease. It is most often grown as a leaf vegetable. In many countries, it is typically eaten cold and raw in salads, hamburgers, tacos and many other dishes. The lettuce plant has short stem initially (a rosette growth habit), but when it blooms the stem lengthens and branches and it produce many flower heads that look like those of dandelions, but smaller, which is called botting. When grown to eat, lettuce is harvested before it bolt.

Spinach (*Spinacia oleracea*) is an edible flowering plant that belongs to the members of the genus *Amaranthus* and the family of *Amaranthaceae*. It is native to central and southwestern Asia. It is an annual plant (rarely biennial), which grows to a height of up to 30 cm. Spinach may survive over winter in temperate regions. The leaves are alternate, simple, ovate to triangular-based, very variable in size from about 2-30 cm long and 1-15 cm broad, with larger leaves at the base of the plant and small leaves higher on the flowering stem.

Atomic Absorption Spectrophotometry is an analytical method that is based on the absorption of UV-visible radiation by free atoms in the gaseous state. The sample to be analyzed is normally digested and then dissolved in an aqueous solution. These samples solutions were neutralized into an air-acetylene flame, where they were heated to vapourized and atomized the metals. A beam of radiation was passed through the

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Fig. 1: Lettuce leaves

Fig. 2: Spinach leaves

atomized samples and absorption of radiation was measured at specific wavelength corresponding to the metals of interest. Information about the type and concentration of metals present was obtained by measuring the location and intensity of the peaks in the absorption spectra.

Similarly researches have been conducted on the heavy metal contents of vegetables in many parts of the world such as "Heavy metal contents of vegetables irrigated by sewage/tube well water" by Lone *et al.* (2003). Similar works have been conducted within the country such as "Levels of selected heavy metals in some Nigerian vegetables" by Anthony *et al.* (2007). The variation in copper, zinc, manganese and lead content of raw and processed tomato samples from Bauchi, Kaduna and Kano factories by Ejeh and Maikai (2002) all revealing Pb and Fe concentrations to be higher than their permissible limits. Similarly Audu and Lawal (2006) analyzed the heavy metal contents in onions, spinach, tomatoes, lettuce, cabbage and carrot in Kano metropolis in which onions recorded the highest mean levels of Fe, Cu, Zn and Co; while spinach recorded the highest Pb content. This present research is aimed at comparing heavy metal uptake in vegetables Spinach (*Spinacea oleracea*) and lettuce (*Lactuca sativa*).

MATERIALS AND METHODS

The sampling was carried out in the month of April, 2009. Samples were collected from each sample location (Kalambaina) using clean plastic material. 2 kg of each of the plants (*Lactuca sativa)* and spinach (*Spinacea oleracea*), 1 kg of their soils were collected from 6 different points with 100 m radius. Each collected sample was immediately transported in cleaned polythene bags to the laboratory. Prior to analyses, the samples were identified and authenticated by Botanical unit of the Usmanu Danfodiyo University Sokoto.

The soils samples were collected at 15 cm depth around the sampled plant (Amusan *et al.*, 2005). These were thoroughly mixed and transferred into clean and labeled envelopes.

Sample treatment: The fresh lettuce (*Lactuca sativa*) and spinach (*Spinacea oleracea*) leaves were washed in a 1% detergent solution and then rinsed with distilled water in order to remove contaminant (Wallace *et al.*, 1982). The samples were oven dried at 105°C to constant weight (Hassan *et al.*, 2005). The plant material was then ground using pestle and motar in order to obtain a homogeneous sample from which representative samples were taken. The milled plant materials were passed through a 20 µm sieve (Aiboni, 2001).

The soil samples were oven dried at 105°C to constant weight for 6 h, crushed and sieved through a 20 µm mesh (Aiboni, 2001). Analytical grade reagents were procured and used. All the glassware used in the preparations were carefully washed with detergent, soaked in (1:4) nitric acid and then rinsed with distilled water before drying in the oven.

Methods earlier described by AOAC (1990) was adopted for moisture and Ash Content Determinations. pH determination was carried out while AAS analysis prior to a wet digestion method as proposed by Uba and Uzairu (2008) was adopted. In order to investigate the ratio of the concentration of heavy metal in a plant to the concentration of heavy metal in soil, the transfer factors for each heavy metal were computed based on the method described by Uwah *et al.* (2009), according to the formula:

$$TF = Ps\ (\mu g/g)/St\ (\mu g/g)$$

Where Ps is the concentration of heavy metal in plant and St is the total metal contents in the soil. Generally, the collected data were subjected to statistical tests of significance using student t-test ($p<0.05$) to assess results in the spinach and lettuce samples.

RESULTS AND DISCUSSION

The results in this analysis revealed a moisture content of 90% for lettuce and 81.75% for spinach. The values are within the range of some common Nigeria vegetable. Generally, the moisture content of vegetable as reported by Hassan *et al.* (2002) is very high ranging from 72% in cassava leaves to 92.93% in Indian spinach and water leaves respectively. The ash content indicates

Table 1: Moisture content, ash content and heavy metal compositions in (µg/g) dry weight of the vegetable samples

Sample	Metals concentration (µg/g) Pb	Fe	Mn	Cr	Cd	Zn	Cu	Moisture content (%)	Ash content (%)
Spinach	$2.07^{a}\pm0.07$	$211^{b}\pm23$	$7.0^{a}\pm0.5$	$5.00^{a}\pm2.80$	0.01	$20.40^{b}\pm0.25$	$1.1^{a}\pm0.1$	81.75±0.4	16.24
Lettuce	$1.59^{b}\pm0.08$	$364^{a}\pm32$	$7.5^{a}\pm2.5$	$5.25^{a}\pm0.35$	0.01	$24.12^{a}\pm0.80$	$0.8^{a}\pm0.2$	90.00±0.5	22.50

Results are presented as mean±standard deviation. Mean values with superscript alphabets, ^{a} and ^{b} are statistically different ($p<0.05$)

that the leaves are rich in mineral elements. The values obtained are higher compared to 1.8 of sweet potatoes leaves (Asibey-Berko and Tayie, 1999). 19.61% in Amaranthus hybridus leaves (Nwaogu *et al*., 2000) 10.83% in water leaves and spinach leaves (Umar *et al*., 2007) and 18.00 in Balsam apple leaves (Hassan and Umar, 2006).

Heavy metal content of the vegetables: The result shows that the lead concentrations in the vegetable samples are 2.07 µg/g and 1.59 µg/g. The values are lower than 10.13 µg/g earlier reported by Anthony *et al.* (2007) and 13.99 µg/g, 17.03 µg/g respectively in the leaves of cocoyam and pawpaw leaves as observed by Amusan *et al.* (2005). The values compared well with the 2.00 µg/g value set as the critical level for edible portions of vegetables by WHO.

The Cadmium concentration in the samples is 0.01 µg/g. These values are lower than the 3.20 µg/g for spinach irrigated with sewage reported by Lone *et al.* (2003) and much lower than the range of 1.34-14 5 µg/g reported for vegetables grown near smelters in New South Wales of Australia by Anthony and Balwart (2009).

The iron concentrations are 364.00 µg/g for lettuce and spinach 211 µg/g. Spinach showed the lower Fe concentration when compared with the analysis reported by Yahaya (2009) 1075.79 µg/g. The concentration of lettuce is almost the same with that of cocoyam leaves (211.64 µg/g) as reported by Amusan *et al.* (2005). The values are within 100-500 µg/g ranges recommended as the normal Fe concentration in plants by ICAR (2006). There were significant differences between the two samples at ($p<0.05$).

The result revealed concentrations of copper in the vegetables showed that there is no significant difference ($p<0.05$) between spinach and lettuce. The concentrations are 1.1 µg/g (spinach), 0.8 µg/g (lettuce). The Cu concentrations in the samples were observed to be lower the values 12.2 µg/g (spinach) by Miller-Ihli and Baker (2000). The values are low when compared with the 10 µg/g value set as the critical level for edible portions of vegetables by WHO (1996).

The result revealed the concentration of 7.0 µg/g and 7.5 µg/g for manganese in spinach and lettuce respectively. The value for spinach is lower than the 165 µg/g reported for spinach leaves by Miller-Ihli and Baker (2000). Similarly, the values are lower than 10.0 ug/g observed in spinach and Indian standard by Kashif *et al.* (2009). Generally, the values are lower than the range of 25-500 µg/g dry mass reported as the normal Mn concentration in plants (ICAR, 2006). The samples showed no significant difference ($p<0.05$).

The zinc concentrations in the vegetables are found to be 20.4 µg/g spinach and 24.12 µg/g for lettuce. There is significant between ($p<0.05$) between the two samples. The value for spinach is lower when compared with the spinach irrigated with sewage water than the 26.169 µg/g reported by Lone *et al.* (2003), the values are low when compared with the 50.00 µg/g reported by Miller-Ihli and Baker (2000). Generally the values were within the range of 20-150 µg/g reported as the normal Zn concentration in plants (Audu and Lawal, 2006).

Cr concentration in the spinach and lettuce ranges between 5.00-5.251 µg/g. The values are significantly in accordance with the research conducted by Saggo and Grewal (2003) that show the uptake of Cr in amended soil is relatively less when compared to other metals and very lower when compared 90.5-93.5 µg/g reported by Uwah *et al.* (2009). The values are higher than value set as the critical level for edible portions of vegetables by WHO (1996). There is no significant difference at ($p<0.05$).

Heavy metal content of the soil: Table 2 shows the pH and concentration of heavy metal in (µg/g) of 0-15 cm top soil. The soil pH ranged from 7.01-7.5 indicating that the soil is almost neutral and therefore very suitable for growing crops (Aiboni, 2001).

The Cr concentrations are 38.67 µg/g (soil samples for spinach) and 38.36 µg/g (soil samples for lettuce) respectively. The values are lower than the values for soils in Alau dam, Gongulon in Maiduguri reported by Uwah *et al.* (2009).

The concentration of the Fe in the soil samples ranged from 416.7 (soils for spinach) to 407 µg/g (soils for lettuce). The values obtained were higher than those reported by Amusan *et al.* (2005) but lower when compared with the values reported by Uwah *et al.* (2009) and so also 75.7-34.1 µg/g by Kashif *et al.* (2009), The values of 12.5 µg/g for the lettuce soil and 41.67 µg/g for the spinach soil were obtained for manganese. The cadmium concentration in 0.5 µg/g of lettuce and spinach soils are 0.5 µg/g. The values generally were observed to be higher than the values of 0.32 µg/g reported by Kashif *et al.* (2009), but lower than the values reported by Uwah *et al.* (2009) and also higher than 0.032-0.21 µg/g normal range for Australian soils

Table 2: pH and heavy metal composition in (µg/g) of the spinach *(Spinacea oleracea)* and lettuce *(Lactuca sativa)* cultivated soils

	Heavy metals (µg/g)							
Sample	Pb	Fe	Mn	Cr	Cd	Zn	Cu	pH
Spinach	11.75±0.11	416.7±35	41.67±2.40	38.67±0.9	0.5	30.10±0.40	4.1±0.26	7.6±0.04
Lettuce	12.07±0.05	407.0±50	12.50±2.04	38.36±0.9	0.5	48.31±9.03	3.5±0.08	7.45±0.0

Results are presented as mean±standard deviation

Table 3: Transfer factor for each metal from soil to lettuce (*Lactuca sativa*) and spinach (*Spinacea oleracea*)

Sample	Pb	Fe	Mn	Cr	Cd	Zn	Cu
Lettuce	0.17	0.9	0.60	0.14	0.02	0.23	0.5
Spinach	0.13	0.5	0.17	0.13	0.02	0.30	0.6

(Kachenko and Singh, 2009). The values fall within the range of 0.001-0.7 µg/g reported for world soils (Kachenko and Singh, 2009). The concentration of Copper in the soil samples are 4.1 µg/g spinach soil and 3.5 µg/g for lettuce soil respectively. Generally, the values are low when compared to 72.99-28.30 µg/g reported by Amusan *et al*. (2005). Lead concentration is 12.07 µg/g for lettuce soil and 11.75 µg/g for spinach soil. The value are low when compared with 418.58 µg/g by Amusan *et al*. (2005).

The transfer factors for heavy metals from soils to lettuce and spinach: Table 3 shows the transfer factors for heavy metals from soils to lettuce and spinach. All transfer factors are below 1, the value for Pb compared well with 0.1 obtain by Kashif *et al*. (2009) and lower than 1.10 reported by Uwah *et al*. (2009). Fe transfer factor is 0.5-0.9 and higher than that reported by Uwah *et al*. (2009). The value for Cu is also high when compared with 0.2 observed by Kashif *et al*. (2009). The transfer factor for Mn, Cr, Cd and Zn are 0.6-0.17, 0.14, 0.02, 0.23-0.3, 0.5-0.6 respectively. Cd values are low than all transfer factors obtained by Kashif *et al*. (2009) and Zn is lower than 0.4 for spinach and for Cr compare well with low values reported by Kashif *et al*. (2009). Transfer factor is one of the key components of human exposure to metals through food chain.

Transfer factors were computed for the heavy metals to quantify the relative differences in bioavailability of metals to plants or to identify the efficiency of a plant species to accumulate a given heavy metal. The factors were based on the root uptake of the metals and discount the foliar absorption of atmospheric metal deposits.

Conclusion: Highlights from this research showed the availability of levels of some heavy metals pollutant (Pb, Fe, Mn, Cr, Cd, Zn and Cu) in the soils with their resultant availability in lettuce and spinach harvested from the same sites as linked to industrial activities, use of fertilizers, manures, pesticides, herbicides and other agro chemicals as well as the use of wastewater by irrigating the soil can caused the increase in the heavy metal contents. The results also revealed that while the levels of some metals in vegetables were within recommended range in plants, others were above the permissible levels recommended by the United States Environmental Protection Agency and World Health Organization.

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