COMPARISON OF HEAVY METAL CONTENT OF SELECTED VEGETABLES GROWN WITH ORGANIC AND INORGANIC FERTILIZERS

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ABSTRACT
Background: Leafy vegetables are good sources of micronutrients but are also bio-accumulators of heavy metals.
Objective: The study compared the heavy metal content of selected vegetables grown with organic and inorganic fertilizer in Odeda Local Government Area (LGA), Ogun State, Nigeria.
Methods: *Celosia argentea, Telfairia occidentalis and Corchorus olitorius* leaves were collected from four randomly selected farmlands. Soup recipes from a recipe book were standardized and used to prepare soups the samples. The vegetables and soups were analyzed in duplicate for lead (Pb), arsenic (As), cadmium (Cd), chromium (Cr), nickel (Ni) and zinc (Zn) using the atomic absorption spectrophotometer. Data obtained were analyzed using SPSS version 20.
Results: Arsenic, Pb and Cd were not detected in both *Telfairia occidentalis* and *Corchorus olitorius* samples, Similarly, As, Pb, Cr were not detected in the *Celosia argentea* and its soup. The Ni and Zn in the *Corchorus olitorius* samples ranged from 0.03mg - 0.6mg/100g and 0.22mg - 0.44mg/100g, respectively. There was not significantly ($P > 0.05$) different between the organic and inorganic and between the raw and cooked samples. The Ni in *Telfairia occidental*, ranged from 0.04mg - 0.06mg/100g, zinc from 0.34mg - 0.47mg/100g in both raw and soup samples. In *Celosia argentea*, Cd ranged from 0.09mg - 0.65mg/100g and was not significantly ($P > 0.05$) different between the organic and inorganic and between the raw and the cooked samples. Heavy metal in the samples did not exceed FAO/WHO limit for vegetables.
Conclusion: The heavy metal content of the organically grown vegetables was not significantly different from the inorganically grown.
Keywords: heavy metals; vegetables; organic, inorganic and fertilizers

Introduction

Vegetables constitute an important part of the human diet since they are rich in carbohydrates, proteins as well as vitamins, minerals and trace elements (1). Vegetables are part of daily diet in many households, making an important source of micronutrients required for health (2). However, they are bio-accumulators of heavy metals (3), they easily absorb heavy metals and it is known that heavy metal accumulation in the body could have adverse effects on human health (4). Although fertilizer is added to soil to supply one or more plant nutrient essential to the growth of plants (5, 6), the presence of metals in some agricultural fertilizers has raised fears that continued use could lead to accumulation of heavy metals to toxic levels in the soil (7) and consequently, in the vegetables planted on the soil.

The organic materials most commonly used to improve soil conditions and fertility include farmyard manure (FYM), animal wastes, crop residues, urban organic wastes (either as such or composted), green manures, biogas spent slurry, microbial preparations, vermin-composting and biodynamic preparations. Sewage sludge and some of the industrial wastes are also find application in agriculture (8). Heavy metals are individual metals and metal compounds that can impact human health. Common examples are arsenic, barium, cadmium, chromium, lead, mercury, selenium, and silver. These are all naturally occurring substances which are often present in the environment at low levels but in larger amounts, they can be dangerous. Generally, humans are exposed to these metals by ingestion or inhalation (9). This study therefore compared the heavy metal content of selected vegetables grown with organic and inorganic fertilizer.

Materials and methods

Sample collection

Three leafy vegetables - *Celosia argentea*, *Telfairia occidentalis*, *Corchorus olitorius* were collected from four randomly selected farmlands (two farms practicing organic and two practicing inorganic agriculture) in Odeda LGA. The organic and inorganic farms were selected by referrals from the farmer's association (A farm was practicing organic or inorganic farming if it had done so consecutively for five years). The collected vegetables were rinsed with water and each sample divided into two and properly labelled. A portion of each was transported to the laboratory for heavy metal analysis while the other portion was taken to the food preparatory laboratory.

Collection and standardization of recipe
Three soup recipes collected from a recipe book (10) were standardized. In the first stage of standardization, the recipe was prepared, and yield was verified. The time of preparation and quantity of ingredients were noted. Afterwards, the recipe was reviewed and prepared and changes made to the recipe was recorded. Exact ingredient by form were recorded (diced, chopped, blended) and every preparation step were also recorded. Detailed procedure, cooking time and recipe yield was recorded. The quantities of ingredients were adjusted, and recipe were prepared

Recipe of the selected vegetables
Two soups were prepared from both organic and inorganic vegetables giving a total 6 soup samples.

Recipe for *Celosia argentea* soup
Ingredients: Palm oil (30ml), 200g fresh tomatoes, 50g fresh red pepper, 10g locust beans, 25g onions, salt to taste, 50g animal skin (*ponmo*), 18g dried fish, 9g red crayfish, 9g small crayfish, seasoning1 cube, 25g onion, 400ml water and 150g *Celosia argentea* leaf.

Method: The dried fish was cleaned, deboned, separated into pieces and the leaves plucked from the stem, rinsed and shredded. Some of the onion was sliced, some blended with pepper using the Kenwood BL 490 model into a puree and the small crayfish ground. The palm oil was heated for 1 minute, the puree, *ponmo*, salt and sliced onions were added, and later the locust beans was added and fried for 10 minutes on a gas cooker in the food preparation laboratory. The red crayfish, dried fish, seasoning and water were added, stirred and allowed to simmer for 5 minutes. The vegetable was shredded and added. The soup cooked for 3 minutes. It was stirred and removed from heat. The yield was 430g.

Recipe for *Corchorus olitorius* soup
Ingredients: 2g potash, 100g *Corchorus olitorius* leaves, 10g locust beans a pinch of salt, 1 cube seasoning and 100ml water
Method: Edible portion of the *Corchorus olitorius* leaves was plucked from the stem and washed properly with a lot of water to remove any sand left on it. The potash was added to boiling water and 200g of *Corchorus olitorius* leaves and locust beans were added to the water and allowed to boil for 10 minutes. A pinch of salt and seasoning for taste were added and it was whisked with a local broom when soft. The yield was 280g

Recipe for *Telfairia occidentalis* soup
Ingredients: Palm oil-30ml, fresh tomatoes-200g, fresh red pepper-50g, locust beans-10g, onions-25g, salt to taste, animal skin (ponmo) -50g dried fish (peeled and deboned)-18g, red crayfish-9g, small crayfish-9g, seasoning 1 cube, water 400ml, *Telfairia occidentalis* leaves-120g
Method: The dried fish was peeled, deboned, separated into pieces and the leaves plucked from the stem, rinsed and shredded. Some of the onion was sliced, some blended with pepper using the Kenwood BL 490 model into a puree and crayfish (small) ground. The palm oil was heated for 1 minute, the puree, *ponmo*, salt and sliced onions were added, and later the locust beans was added and fried for 10 minutes on a gas cooker in the food preparation laboratory. The red crayfish, dried fish, seasoning and water were added, stirred and allowed to simmer for 4 minutes. The vegetable was shredded and added. The soup cooked for 4 minutes. It was stirred and removed from heat. The yield was 370g.

Chemical analysis of the vegetable and soup samples
The samples were digested using the nitric-hydrochloric acid (1-3 ratio) digestion method of digestion (11). Buck Scientific Model 210 Atomic Absorption Spectrophotometer was used for the analysis of the heavy metals. Standards of arsenic, lead, cadmium, nickel, chromium and zinc were prepared. Values obtained from the analysis were converted to the wet weight using the formula of McClements (12).

Statistical analysis
Statistical Product and Service Solutions (SPSS) version 20.0 was used for data analysis. Descriptive statistics (mean and standard deviations) were used to summarize the data and an independent sample T-test analysis was carried out to determine differences in mean values. Significance was accepted at $p < 0.05$.

Results
The concentration of heavy metals in the selected raw vegetables is presented in Table 1. No lead and arsenic were detected in all the vegetables. In the organic samples, cadmium ranged from 0.00mg/100g - 0.09mg/100g, nickel ranged from 0.04mg/100g - 0.09mg/100g, zinc ranged from 0.24mg/100g - 0.47mg/100g. For the inorganic vegetables, Cd ranged from 0.00mg/100g-0.65mg/100g, Ni ranged from 0.06mg/100g - 0.6mg/100g, Cr ranged from 0.00mg/100g - 0.01mg/100g and Zn ranged from 0.34mg/100g - 0.44mg/100. There was no significant difference in the values observed in both vegetables.

Table 1: Heavy metal concentration (mg/100g) in the raw vegetable samples

	Corchorus olitorius			*Telfaria occidentalis*			*Celosia argentea*		
Heavy metal	Organic	Inorganic	p-value	Organic	Inorganic	p-value	Organic	Inorganic	p-value
As	0.00±0.00	0.00±0.00	-	0.00±0.00	0.00±0.00	-	0.00±0.00	0.00±0.00	-
Pb	0.00±0.00	0.00±0.00	-	0.00±0.00	0.00±0.00	-	0.00±0.00	0.00±0.00	-
Cd	0.00±0.00	0.00±0.00	-	0.00±0.00	0.00±0.00	-	0.09±1.33	0.65±0.09	0.82
Ni	0.09±0.02	0.60±0.01	0.21	0.04±0.05	0.06±0.02	0.69	0.07±0.03	0.06±0.00	0.42
Cr	0.00±0.00	0.01±0.02	0.54	0.00±0.00	0.01±0.01	0.33	0.00±0.00	0.00±0.00	-
Zn	0.42±0.00	0.44±0.02	0.76	0.47±0.37	0.34±0.11	0.68	0.24±0.17	0.41±0.18	0.18

The concentration of heavy metals in the cooked vegetables is shown in Table 2. No Pb and As were observed. Cadmium ranged from 0.00mg/100g to 0.37mg/100g in organic samples, Ni ranged from 0.05 to 0.14mg/100g, Cr ranged from 0.00mg/100g to 0.07mg/100g and Zn ranged from 0.22mg/100g to 0.51mg/100g. For the inorganic vegetables, no Cd, Pb, As was found. Chromium ranged from 0.00mg/100g to 0.01mg/100g and Zn ranged from 0.19 to 0.28mg/100g. No significant difference was found in the heavy metals content between the cooked organic and inorganic vegetables except for organic and inorganic *Corchorus olitorius* ($P = 0.03$) and Cr in raw and cooked organic *Telfairia occidentalis* ($P = 0.04$). Comparing the heavy metals in the organically and inorganically grown vegetables showed no significant difference ($p > 0.05$) for both the raw and cooked. However, the Cr content between raw and cooked organic *Telfairia occidentalis* was significantly different (P-value = 0.04). The raw vegetable had no Cr content while the cooked had 0.07mg/100g.

Table 2: Heavy metal concentration (mg/100g) in the cooked vegetable samples

	Corchorus olitorius			*Telfairia occidentalis*			*Celosia argentea*		
Heavy metal	**Organic**	**Inorganic**	**p-value**	**Organic**	**Inorganic**	**p-value**	**Organic**	**Inorganic**	**p-Value**
As	0.00±0.00	0.00±0.00	-	0.00±0.00	0.00±0.00	-	0.00±0.00	0.00±0.00	-
Pb	0.00±0.00	0.00±0.00	-	0.00±0.00	0.00±0.00	-	0.00±0.00	0.00±0.00	-
Cd	0.00±0.00	0.00±0.00	-	0.00±0.00	0.00±0.00	-	0.09±1.33	0.65±0.09	0.82
Ni	0.05±0.10	0.09±0.02	0.21	0.40±0.05	0.06±0.02	0.69	0.00±0.00	0.01±0.01	0.33
Cr	0.00±0.00	0.01±0.02	0.54	0.00±0.00	0.01±0.01	0.33	0.00±0.00	0.00±0.00	-
Zn	0.42±0.00	0.44±0.02	0.76	0.47±0.37	0.34±0.11	0.68	0.24±0.17	0.41±0.18	0.18

Comparison of the heavy metals (mg/100g) in the raw and cooked vegetables

The comparison between the raw and cooked organic and inorganic vegetables is shown in Tables 3 and 4. The comparison between raw and cooked organic *Celosia argentea,* showed no significant difference in the Cd, Ni and Zn content (p = 0.42, 0.470, 0.37, respectively). Arsenic, Pb and Cr were not detected in the samples and there was no significant difference between the Ni and Zn of the inorganic samples. In the raw and cooked organic *Telfairia occidentalis,* there was no significant difference between the Cd, Ni and Zn (p = 0.32, 0.79 and 0.84, respectively) but there was significant difference in the Cr content p = 0.04). There was no significant difference in the Cd, Ni and Zn content of the raw and cooked inorganic *Celosia argentea* (p = 0.42, 0.38 and 0.65). There was also no significant difference in the Ni, Cr and Zn content of raw and cooked inorganic *Corchorus olitorius* (p = 0.20, 0.49, 0.12 respectively) as well as between the Ni, Cr and Zn in raw and cooked inorganic *Telfairia occidentalis* (p = 0.55, 0.96 and 0.74, respectively).

Table 3: Comparison of heavy metals in the raw and cooked organic vegetables

	Corchorus Olitorius			*Telfairia Occidentalis*			*Celosia Argentea*		
Heavy metals	Raw	Cooked	P-value	Raw	Cooked	P-value	Raw	Cooked	P-value
As	0.00±0.00	0.00±0.00	-	0.00±0.00	0.00±0.00	-	0.00±0.00	0.00±0.00	-
Pb	0.00±0.00	0.00±0.00	-	0.00±0.00	0.00±0.00	-	0.00±0.00	0.00±0.00	-
Cd	0.00±0.00	0.00±0.00	-	0.00±0.00	0.00±0.00	0.32	0.09±1.33	0.00±0.00	0.42
Ni	0.09±0.02	0.05±0.01	0.66	0.40±0.05	0.40±0.05	0.78	0.07±0.03	0.00±0.00	0.47
Cr	0.00±0.00	0.00±0.00	0.03	0.00±0.00	0.00±0.00	0.04	0.00±0.00	0.00±0.00	-
Zn	0.42±0.00	0.42±0.00	0.90	0.47±0.37	0.47±0.37	0.84	0.24±0.17	0.24±0.17	0.37

Table 4: Comparison of heavy metals in the raw and cooked inorganic vegetables

	Corchorus Olitorius			*Telfairia Occidentalis*			*Celosia Argentea*		
Heavy metals	Raw	Cooked	p-value	Raw	Cooked	p-value	Raw	Cooked	p-value
As	0.00±0.00	0.00±0.00	-	0.00±0.00	0.00±0.00	-	0.00±0.00	0.00±0.00	-
Pb	0.00±0.00	0.00±0.00	-	0.00±0.00	0.00±0.00	-	0.00±0.00	0.00±0.00	-
Cd	0.00±0.00	0.00±0.00	-	0.00±0.00	0.00±0.00	-	0.65±0.09	0.00±0.00	0.42
Ni	0.60±0.01	0.09±0.02	0.20	0.06±0.02	0.06±0.02	0.54	0.06±0.00	0.01±0.01	0.38
Cr	0.01±0.02	0.01±0.02	0.49	0.01±0.01	0.01±0.01	0.96	0.00±0.00	0.00±0.00	-
Zn	0.44±0.02	0.44±0.02	0.12	0.34±0.11	0.34±0.11	0.74	0.41±0.18	0.41±0.18	0.65

Table 5 shows the comparison of the heavy metal concentrations of the vegetables to levels permitted by the WHO. Levels of the heavy metal concentrations of the organic and inorganic vegetables were not above the permissible limit recommended by the WHO (13).

Table 5: Comparison of level of heavy metals (mg/100g) in vegetables and FAO/WHO standard

Vegetable	Zn	Cd	Pb
	Raw Organic		
Celosia argentea	0.24	**0.09**	0.00
Corchorus olitorius	0.43	0.00	0.00
Telfairia occidentalis	0.47	0.00	0.00
	Cooked Organic		
Celosia argentea	0.45	0.00	0.00
Corchorusolitorius	0.22	0.00	0.00
Telfairia occidentalis	0.51	**0.37**	0.00
	Raw inorganic		
Celosia argentea	0.41	**0.65**	0.00
Corchorus olitorius	0.44	0.00	0.00
Telfairia occidentalis	0.34	0.00	0.00
	Cooked inorganic		
Celosia argentea	0.28	0.00	0.00
Corchorus olitorius	0.23	0.00	0.00
Telfairia occidentalis	0.40	0.00	0.00
WHO/FAO standard	**No Maximum limits**	**0.02**	**0.03**

Discussions

Generally, heavy metal contents vary among different vegetables because of different absorption capacity (14). It has been reported that, there is also a difference in the root uptake rate of heavy metals by different vegetables (15). This might have accounted for the variation in the level of the different heavy metals detected in the raw vegetable samples. Studies have however shown that the variation in the vegetable with the higher heavy metal content could be because heavy metals contamination arising from natural sources (seepage from rocks, volcanic activities, forest fires) and anthropogenic sources (manures, sewage sludge, fertilizers, pesticides, traffic emission, waste incineration, urban effluent) (16).

Generally, the heavy metals in the raw inorganic vegetable samples exceeded the heavy metals found in organic vegetables but were not significantly different. A study reported that conventional vegetables generally contained higher amounts of heavy metal studied compared to their organic counterpart but the author further stated that the observation did not suggest that conventional vegetables were less safe than their organic counterparts because the organic vegetables in some cases had higher amounts of heavy metals for some vegetables (17). Although, the popularity of consuming organic foods has grown, more recent studies revealed that organically grown foods have no significant overall differences in their nutritional quality when compared to inorganic foods (18). Also, it has been reported that consumption of organic foods may reduce exposure to pesticide residues and anti-biotic resistant bacteria but there is no strong evidence that shows organic foods are more nutritious than inorganic foods (19).

After cooking the vegetables, it was observed that the concentration of heavy metals in some samples increased while a decrease was observed in others. It has been reported in a study that minerals are not lost due to heat, but are usually leached if cooked in boiling water, this might account for the general reduction in the level of heavy metals in *Corchorus olitorius,* since it was cooked in boiling water unlike *celosia argentea* and *Telfairia occidentalis* that were steamed (20). It was also observed that there was a general increase in the concentration of heavy metals found in the cooked organic and the inorganic *Telfairia occidentalis.* Research has shown that *Telfairia occidentalis* has high water and oil absorption capacities (21). Cooking methods such as boiling and frying can alter the content of a toxic element through the loss of water and volatiles, the solubilization of the metal, and to some extent, metal binding to macronutrients such as carbohydrate but toxic elements are not evaporated or broken down to safer compounds. Hence, the elements which are removed from foodstuff during cooking migrate from food to the frying oil, boiling water or cooking stock (22) but in the case of *Telfairia occidentalis* the leaves absorb a high quantity of the heavy metals from the water and oil because of its high absorption capacity.

The concentration of Ni in both organic and inorganic *Celosia argentea* and *Telfairia occidentalis* in this study increased after cooking.

This could be because stainless steel cookware leaches Ni into foods during cooking (23). Studies have shown that stainless steel cookware leaches nickel into cooked foods (24, 25). The concentration of heavy metals in the organic and inorganic vegetables was observed to be lower than those found in previous studies. A study reported a range of 0.71-15.89mg/kg, 0.07-0.97mg/kg, 0.18-5.05mg/kg and 0.18-1.59mg/kg for Ni, Cd, Cr and Pb, respectively on dry matter basis (26). This could be because the values reported are on the dry matter while the values reported in this study were converted back to the wet weight. It might also be because the study was conducted on marketing sites as some studies have reported that transportation and marketing systems play a significant role in elevating contaminant levels of heavy metals of vegetables than production sites (27, 28).

No lead was detected in all the vegetables in this study. Generally, plants do not absorb or accumulate lead, possibility of absorption only occurs in soil high in lead, but the lead is highly immobilized in soils (3, 29). A study also reported that lead in plants is associated with proximity to automobile traffic (30). In another study carried out to check for heavy metal contamination of vegetables in urban city in Lagos, it was observed that the test vegetables contained high concentration of heavy metal especially Pb because of atmospheric depositions of heavy metals at the time of transportation and marketing (27). This might explain the reason why the vegetables in this study contained no Pb, they were freshly harvested from the farmlands without exposure to automobile traffic during transportation or marketing. Backyard gardening would be a good choice for household's vegetable source, since vegetables would be harvested freshly without exposure to automobile traffic. Lead poisoning causes inhibition of the synthesis of hemoglobin, dysfunctions in the kidneys, joints and reproductive systems, cardiovascular system and acute and chronic damage to the Central Nervous System (CNS) and peripheral nervous system (31).

There was little or no detection of the metal As in all the vegetable samples. Arsenic has been reported to be associated with hypertension and serious impacts on the cardiovascular system including hepatic damage at high doses (4). Arsenic can cause nausea and vomiting, decreased production of white and red blood cells, abnormal heart rhythm, damage to blood vessels and a sensation of pins and needles in hands and feet (9). With the level of arsenic detected in the vegetables, there is no threat of arsenic poisoning attached to the consumption of the vegetables studied.

The cadmium content observed ranged from 0.00mg/100g to 0.65mg/100g. No cadmium was detected *in Telfairia occidentalis* and *Corchorus olitorius* (both organic and inorganic samples). Only raw *Celosia argentea* had some amount of cadmium and it has been reported to be a traffic pollutant (32). This explains why the value was low compared to those obtained in literatures, since the vegetables were gotten directly from farmlands.

Zinc was the most prevalent heavy metal in the vegetable samples. It ranged from 0.24mg/100g to 0.47mg/100g. Although zinc is referred to as heavy metal, but it is needed by the body for various functions which include bone formation, taste acuity, immune function. However, its toxicity is associated with supplemental intakes at 5 or more times than the RDI (15).

Conclusions

There were different levels of arsenic, lead, chromium, cadmium, nickel and zinc in study samples. The heavy metal concentration of both organic and inorganic vegetables were not significantly different.

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