Lead and Cadmium Accumulation in African Spinach (*Amaranthus cruentus* L.) Grown on Soil from a Dumpsite

Fawole, T. O.[1], Oyaniyi, T. O.[1], Omilabu, S. K.[2], Olla, N. O.[3], Bidmos, F. A.[4], Olakunle S. D.[5]

[1] Department of Crop Production Technology, Oyo State College of Agriculture and Technology P.M.B. 10, Igboora, Oyo State.

[2] Department of Agricultural Technology, Oyo State College of Agriculture and Technology P.M.B. 10, Igboora, Oyo State.

[3] Department of Soil Science Technology, Oyo State College of Agriculture and Technology P.M.B. 10, Igboora, Oyo State.

[4] Department of Horticultural Technology, Oyo State College of Agriculture and Technology P.M.B. 10, Igboora, Oyo State.

[5] Department of Forestry Technology, Oyo State College of Agriculture and Technology P.M.B. 10, Igboora, Oyo State.

Correspondence: olaoluwapeju@gmail.com, +2347039686610

Abstract

In this study, a nursery experiment was set up to assess the Lead (Pb) and Cadmium (Cd) accumulation in *Amaranthus cruentus* seedlings grown on contaminated soil. *Amaranthus* seeds were broadcast on the nursery bed and allowed to germinate for one week before sprouted seedlings were pricked and transplanted into polypots containing soil from a dumpsite, topsoil and sandy soil (5 kg each). The seedlings were arranged in a completely randomize design with 5 replicates per treatment. The physico-chemical properties, Lead and Cadmium concentrations in all soil types were determined before planting, while data were collected for eight weeks after planting. Weekly measurements of plant height, collar diameter, and number of leaves were done, while Lead and Cadmium concentrations in the plants were determined after harvest. Lead and Cadmium were detected in the dumpsite soil and *Amaranthus cruentus* grown on the dumpsite soil. Highest height, collar diameter and number of leaves occurred in the contaminated seedlings. *Amaranthus cruentus* absorbed Lead (7.66 mg/kg) and Cadmium (0.54 mg/kg), which were higher than the recommended safe limits for vegetables (0.3 mg/kg and 0.2 mg/kg, respectively). In addition, transfer factors for Lead (0.53 mg/kg) and Cadmium (0.51 mg/kg) indicated high bio-accumulation of the metals by the vegetables. Hence, vegetables produced from dumpsite soil may not be safe for consumption.

Keywords: Bioaccumulation; Lead; Cadmium; African Spinach; Transfer factor

Introduction

Amaranthus cruentus L. is a commonly cultivated vegetable in Africa, Asia and Southern Mexico with historical evidence that dates back to 6700 BC (1). In Nigeria, it is produced in home gardens, semi-urban gardens, inland valleys (Fadamas) and on farms using different cropping systems. It is a staple crop because about 40 % of the total production by farmers is consumed by family members, while the remaining is sold in markets (2).

Amaranthus (commonly called 'green' vegetable) is seen by many as a dicotyledonous pseudo-cereal crop of high nutritional value. The development of the plant as an alternative crop has attracted the attention of several researchers over the past decade (3, 4, 5). According to Aufhammer *et al.* (6), the leafy vegetable contains high crude protein, calcium, potassium, vitamins A and C. Thus, it contributes significantly to daily nutritional requirements of consumers (6, 7, 8). The seeds of the vegetable can be consumed raw or milled into flour and used as an ingredient when preparing porridge, pancakes, bread muffins, crackers and paste (9). Apart from their dietary importance, amaranth plants have a good history of medicinal uses. Fresh and dried leaf powder of the plant have been traditionally used to treat inflammation, gonorrhea and hemorrhoids, while pounded roots have been used to treat dysentery, and the leaf sap used for treating eye infections (10).

Amaranthus cruentus matures from 4 to 6 weeks and its cultivation is not labour intensive like some other vegetables such as Celosia, Pumpkin. The species grows optimally in high organic-matter and mineral reserve soils (11). These attributes make Amaranth one of the most important leafy vegetables, which provide income for semi-urban and urban dwellers.

Environmental pollution occur as a result of increased industrialization, environmental disasters, indiscriminate waste disposal and other negative anthropogenic activities. Unfortunately, some of the inorganic contaminants introduced to the environment during pollution contain heavy metals, which are not biodegradable to non-toxic compounds (12). The presence of these heavy metals in the soil has been linked to some health problems such as neurological disorders, central nervous system destruction, cancers of various body organs (4, 5) and severe mental retardation in children (32).

Vegetables such as *Amaranthus cuentus* have the potential to absorb heavy metals from soil, air and nutrient solutions through their roots or foliage (22), thereby exposing consumers to risk of adverse health effects after consumption, especially when heavy metal contaminants are present on farmlands or abandoned dumpsite where the vegetable was cultivated (16, 26). In some rural areas, people produce vegetables on abandoned dumpsites. However, there is limited information on the concentration of heavy metals in the soil on these dumpsites. These heavy metals could bio-accumulate in vegetable crops. Therefore, this study compared the growth and yield of *Amaranthus cruentus* in dumpsite soil, topsoil and sandy soil. It also investigated the bio-accumulation of Lead and Cadmium in the *Amaranthus cruentus* seedlings.

Materials and Methods

The experiment was conducted in the screen house of the Department of Agricultural Technology, Oyo State College of Agriculture and Technology, Igboora, Oyo State. Igboora is the headquarters of Ibarapa Central Local Government Area in Oyo State. It is in the derived savanna zone along Latitude 7^0 26'1"N and Longitude $3^0$17'16"E. The city shares its boundaries with Abeokuta, Ibadan and Oke-Ogun regions (35). There are two distinct seasons; the dry season (from

November to March) and wet season (April to October). The average high temperature of the region is 33 °C, while the average low temperature is 22 °C (35).

A soil auger (3 cm depth) was used to collect composite soil samples from the dumpsite, top soil and sandy soil. The samples were subjected to physico-chemical and heavy metal analyses. The soil samples were air dried for 21 days, crushed and sieved to remove debris. The hydrometer method was used to determine sample particle size (9). The total nitrogen of the substrates was assessed using the macro-Kjeldahl method (19). The pH was determined with pH meter, while available phosphorus was estimated using the Bray 1 method (10).

The Lead (Pb) and Cadmium (Cd) content of each substrate were also determined. One gram of each substrate was weighed into a 100 ml beaker and 20 ml of 1:1 HNO_3 (analar grade) was added. The mixture was boiled in a fume cupboard to reduce the volume of HNO_3 to 5 ml. Thereafter, 200 ml of de-ionised water was added and the mixture boiled gently until the volume was 10 ml. The suspension was cooled and filtered through a Whatman No. 540 filter paper. The beaker and filter paper were rinsed with de-ionised water to 25 ml mark. The filtrate was later transferred to a 100 ml graduated flask, which was filled to the mark with more de-ionised water. Lead and Cadmium contents of the samples were determined using Atomic Absorption Spectroscopy (AAS) following the method of AOAC (3).

Amaranthus cruentus seeds were germinated on nursery beds for 2 weeks before they were transplanted into polypots containing 5 kg of substrate from dumpsite, topsoil and sandy soil, respectively. The polypots were arranged in a completely randomized design experiment with five replicates for each treatment. Two weeks after transplanting, growth variables such as plant height, number of leaves, collar diameter and leaf area were monitored and measured weekly for 8 weeks. After 8 weeks, *A. cruentus* were harvested and analysed for Pb and Cd content following the procedure described above.

Data were analysed using Analysis of Variance (ANOVA), while Duncan's Multiple Range Test was used to separate the significantly different means at $p \leq 0.05$ level of significance.

Transfer Factor (TF) or Plant Concentration Factor (PCF)

The transfer factor or plant concentration factor is an important component of human exposure to heavy metals along the food chain. The transfer factor describes the movement of heavy metals from soil to plants, and it is the ratio of heavy metal concentration in vegetables and the total heavy metal concentration in the soil (20). The TF of Pb and Cd in *Amaranthus cruentus* grown in different substrates were determined using equation 1:

$$TF = \frac{C_{plant}}{C_{soil}} \quad \ldots\ldots\ldots\ldots\ldots\ldots\ldots\ldots\ldots\ldots (1)$$

where, TF= Transfer factor, C_{plant} = concentration of heavy metal in fresh weight of plant (mg/kg) and C_{soil} = concentration of heavy metal in dry weight of soil (mg/kg).

When the transfer factor is greater than 0.5, it means that there is high mobility of heavy metals from the contaminated soil to the plant. This also indicates that the bio-accumulation of the heavy metal by the plant is high and could pose a health threat to consumers. However, a transfer factor below 0.5 indicates low bio-accumulation of heavy metals by the plant.

Results and Discussion

Physico-chemical Analysis of Substrates

The substrates obtained from the dumpsite, topsoil and sandy soil were slightly alkaline; with pH values of 7.67, 7.50 and 7.20, respectively. The essential nutrients were highest in the dumpsite e.g. nitrogen: 0.38% and potassium: 3.40 cmol/kg (Table 1). However, the essential nutrients were below the critical values of soils in the derived savanna ecological zones. This may be due to low levels of soil organic matter in the region (34). Lead (14.45 mg/kg) and Cadmium (0.15 mg/kg) were only detected in dumpsite soil samples, with no traces detected in substrates from topsoil and sandy soil (Table 1).

Table 1: Physical and chemical properties of soil from dumpsite, topsoil and sandy soil at Igboora, Oyo state, Nigeria

Elements	Dumpsite soil	Top soil	Sandy soil
pH (H_20)	7.67	7.50	7.20
Nitrogen (%)	0.38	0.27	0.20
Phosphorus (mg/kg)	30.25	29.20	15.50
Potassium (cmol/kg)	3.40	2.80	1.20
Lead (mg/kg)	14. 45	ND	ND
Cadmium (mg/kg)	1.06	ND	ND

Growth Variables of *Amaranthus cruentus* Seedlings in Three Substrates

There were significant differences in number of leaves of *A. cruentus* across treatments from the fourth to eighth week, after transplanting (Table 2). After 8 weeks, *A. cruentus* planted in dumpsite soil had the highest number of leaves (36.25) which was significantly higher than those planted in sandy soil (13.75) and top soil (12.50), respectively.

Table 2: Growth variables of *Amaranthus cruentus* planted in substrate from dumpsite, topsoil and sandy soil at Igboora, Oyo state, Nigeria

Substrate	2 weeks	3 weeks	4 weeks	5 weeks	6 weeks	7 weeks	8 weeks
		Number of leaves					
Dumpsite	9.00a	10.25a	13.00a	19.00a	24.75a	29.25a	36.25a
Sand soil	8.50a	9.25a	9.75b	10.75b	11.00b	12.00b	13.75b
Top soil	7.25a	7.50a	8.25b	9.75b	10.75b	11.50b	12.50b
			Collar diameter (mm)				
Dumpsite	1.97a	2.15a	2.90a	4.28a	5.30a	5.45a	6.05a
Sand soil	1.37b	1.53b	1.57b	2.00b	2.17b	2.33b	3.25b
Top soil	1.09b	1.25b	1.33b	1.66b	2.06b	2.19b	2.41b
			Plant height (cm)				
Dumpsite	5.60a	5.95a	9.47a	16.95a	21.42a	22.20a	28.82a
Sand soil	2.85b	3.55b	5.65b	8.03b	10.47b	11.15b	12.72b
Top soil	3.23b	3.65b	4.67b	6.45b	7.58b	8.22b	9.97b
			Leaf area (cm^2)				
Dumpsite	8.74a	11.45a	15.60a	24.78a	33.21a	42.49a	63.09a
Sand soil	2.62b	4.14b	6.05b	10.56b	13.72b	15.41b	16.38b
Top soil	1.86b	2.30b	3.28b	6.52b	7.66b	9.23b	13.73b

ND- Not Detected

Means with the same alphabet in rows are not significantly different at $p \leq 0.05$.

The collar diameter of *A. cruentus* planted in different substrates varied significantly during the study (Table 2). The stem collar diameter increased with time, with seedlings in the dumpsite soil having the highest collar diameter (6.05 mm), while topsoil was least (2.41mm), after 8 weeks.

Similarly, seedling height significantly differed among the treatments. After 8 weeks, *A. cruentus* planted in dumpsite soil had the highest height (28.82cm), while top soil had the least (9.97cm).The leaf area significantly increased from the second to the eighth week (Table 2). After 8 weeks, *A. cruentus* planted in dumpsite soil had the highest leaf area (63.0 cm^2), while topsoil was least (13.73 cm^2). The higher growth performance observed for *Amaranthus cruentus* planted in dumpsite soil could be due to higher levels of nitrogen, phosphorus and potassium in the polluted soil (25).

Lead and Cadmium Concentrations in the Substrates and Plants

There was a transfer of heavy metals from the dumpsite soil into the plant biomass after 8 weeks (Table 3). The concentration of Pb (14.45 mg/kg) and Cd (1.06 mg/kg) in the dumpsite soil before planting, were higher than the recommended limits for Pb (1.6 mg/kg) and Cd (0.2 mg/kg) in soils (33). There was a translocation of heavy metals from the dumpsite soil to the *A. cruentus* tissues (Pb = 7.66 mg/kg and Cd = 0.54 mg/kg). These heavy metal concentration levels in the plants were higher than the safe limits recommended for Pb (0.3 mg/kg) and Cd (0.2 mg/kg) in vegetables (36).The transfer factors obtained for *Amaranthus cruentus* planted on dumpsite for Pb and Cd were 0.53 mg/kg and 0.51 mg/kg, respectively.

Accumulation of heavy metals in soils and plants have been a major concern because of their potential risks to human health. This study showed that *Amaranthus cruentus* grown on the dumpsite soil accumulated Pb and Cd, which corroborated the findings of Aljassir and Khalia (2). Leafy vegetables accumulate heavy metals at high rate and Ogunyemi *et al.* (27) had previously reported that vegetables grown in dumpsites of urban and peri-urban areas in Ibadan, contained high levels of heavy metals. These environmental contaminants are widely distributed in the air, water bodies and soils, and bio-accumulate in ecosystems, thus constituting a potential threat to human health (10). The vegetables absorb the heavy metals and store them in their leaves and other edible parts. These metals are subsequently transferred through vegetable consumption to humans and animals. This public health risk could cause damage to organs and increase vulnerability to cancers (27).

The efficiency of plant absorption of metals is evaluated by either plant uptake from soil or plant transfer factors of the metals (20, 29). In this study, the transfer factors were high for Pb (0.53 mg/kg) and Cd (0.51 mg/kg) implying high phytoextraction of the metals by amaranths. Hence, the selective permeability of *A. cruentus* roots is not a barrier to the translocation of the heavy metals and leafy vegetables have a high capacity to phytoextract heavy metals (13, 15, 18).

The uptake of Pb and Cd by the vegetables beyond safe limits recommended by WHO/FAO (36) suggests that the accumulation of these heavy metals in human body through the food chain poses serious health challenge to the consumers. This is corroborated by Fayinminnu and Adekunle – Jimoh (16) who reported that shoot (edible part) and root of *Amaranthus hybridus* L. from selected locations in Ibadan were contaminated with heavy metals (Pb and Cd) at levels exceeding permissible limits. Therefore, there is a likelihood that consumers may experience long term effects associated with consumption of *A. cruentus* grown in dumpsite soil.

Table 3: Lead and Cadmium concentrations in *Amaranthus cruentus* seedlings grown in three substrates for 8 weeks

Heavy metal composition	Lead mg/kg	Cadmium mg/kg
Dumpsite soil before planting	14.45	1.06
Sandy soil before planting	ND	ND
Top soil before planting	ND	ND
Dumpsite soil after planting	8.10	0.32
Sandy soil after planting	ND	ND
Top soil after planting	ND	ND
Heavy metals absorbed by *Amaranthus cruentus* planted on dumpsite soil	7.66	0.54
Heavy metals absorbed by *Amaranthus cruentus* planted on sandy soil	ND	ND
Heavy metals absorbed by *Amaranthus cruentus* planted on top soil	ND	ND
Transfer Factor (TF)	0.53	0.51
US-EPA maximum limits for heavy metals in soil (mg/kg)	1.6	0.2
WHO/FAO safe limits for heavy metals in vegetables (mg/kg)	0.3	0.2

ND: Not Detected

Conclusion

The concentration of Pb and Cd in the dumpsite soil exceeded the maximum limits recommended by regulatory bodies indicating that vegetables produced from this medium may be hazardous. Although, vegetable produced in the dumpsite had higher growth performance, than those in topsoil, they were unsafe. Hence, the use of such sites for cultivation of food must be discouraged, in order to avoid the bioaccumulation of heavy metals among local residents and other consumers.

References

1. Akanbi, W.B. and Togun, A.O. 2002. The influence of maize-stover compost and nitrogen fertilizer on growth, yield and nutrient uptake of amaranth. *Scientia Horticulturae*, 93: 1–8.

2. Aljassir, M. S. A and Khalia, M. A. 2005. Deposition of heavy metals on green leafy vegetables sold on roadsides of Riyadh city Saudi – Arabia. *Bulletin of Environmental Contamination and Toxicol*ogy, 75: 1020 - 1027.

3. AOAC, 2005. Official method of analysis. Association of Analytical Chemists. Washington DC. 15th ed.11-14.

4. ATSDR, 1999.Toxicological Profile for Cadmium and Nickel. Agency for Toxic Substances and Disease Registry, US

Department of Health and Human Services, Public Health Service, 205-93-0606.

5. ATSDR, 2000.Toxicological Profile for Arsenic. Agency for Toxic Substances and Disease Registry, US Department of Health and Human Services, Public Health Service, 205-1999-00024.

6. Aufhammer, W., Chuczorova, H.P.K. and Kruse, M. 1998. Germination of grain amaranth (*A. hypochondriacus* x *A. hybridus*): Effects of seed quality, temperature, light, and pesticides. *European Journal of the Agronomic Society* 8,127-135.

7. Axtell, N. R, Sternberg, S. P. K and Claussen, K. 2003, 'Lead and nickel removal using Microspora and Lemna minor', *Bioresource Technology* 89: 41-48.

8. Basta, N. and Gradwohl, R. 2000. 'Estimation of Cd, Pb and Zn bioavailability in smelter-contaminated soils by a sequential extraction procedure. *Journal of Soil Contamination* 9(2): 149-164.

9. Bouyoucos, G.J. 1962. Hydrometer method improved for making particle size analysis of soils. *Soil Science Society of America Proceedings* 26: 464-465.

10. Bray, R.H. and Kurtz, L.T. 1945. Determination of total organic and available forms of phosphorus in soils. *Soil Science*, 39:39-45.

11. Censis, P., Spoto, S.E.,Saiano, F. and Sprovieri, M. 2006. Heavy metal in coastal water system: A case of study from the northwestern Gulf of Thailand in chemosphere 64 (7):1167 – 117 doi:10.10161.chemosphere 2005.11.008

12. Costea, M. and DeMason, D. 2001. Stem morphology and anatomy in Amaranthus L. (Amaranthaceae)- Taxonomic significance". *Journal of the Torrey Botanical Society*, 128(3):254–281. doi:10.2307/3088717.JSTOR 3088717

13. Davies, B.E. and White, H.M. 1981.Trace element in vegetables grown on soils contaminated by base metal mining. *Journal of Plant Nutrition* 3: 387–396.

14. Denton, L. 1996. Expansion of Amaranth production and utilization in Nigeria *In: National Training Workshop on grain Amaranths production and utilization organized by* NlHORT between 7 and 8 August 1996. pp. 1-8.

15. Dosumu, O.O., Salami, N. and Adekola, F.A. 2003. Comparative study of trace element levels in some local vegetable varieties and irrigation waters from different locations in Ilorin, Nigeria. *Bulletin of Chemical Society of Ethiopia*, 17(1): 107-112.

16. Fayinminnu, O.O. and Adekunle – Jimoh, J.M. 2015. Evaluation of Heavy Metals and Health Risk Index in *Amaranthus hybridus* L. Vegetable Grown in Selected Farms in Ibadan, Nigeria. *Nigerian Journal of Science*, 49: 11-21.

17. Food and Agricultural Organization, FAO 1992. Conversion and rehabilitation of African Land. FAO.

18. Ghosh, A.K., Bhatt, M.A. and Agrawal, H.P. 2012. Effect of long-term application of treated sewage water on heavy metal accumulation in vegetables grown in Northern India. *Environmental Monitoring* and *Assessment*, 184: 1025-1036.

19. Jackson, M.L. 1958. Soil Chemical Analysis. Prentice Hall of India Pvt. Ltd., New Delhi, 498p.

20. Kachenko, A. and Singh, B. 2006. Heavy Metals contamination in vegetables grown in Urban and metal Smelter contaminated sites

in Australia. *Water, Air and Soil Pollution* 169 (1):101 – 123.

21. Leon, R.G., Knapp, A.D. and Owen, M.D.K. 2004. Effect of temperature on the germination of common water hemp (*Amaranthus tuberculatus*), Giant Foxtail *(Setaria faberi*), and Velvetleaf (*Abutilon theophrasti*). *Weed Science*, 52, 67-73.

22. Lokeshwari, H. and Chandrappa, G.T. 2006. Impact of heavy metal contamination of Bellandur Lake on soil and cultivated vegetation. *Current Science,* 91(5): 622- 627.

23. Messiaen, C.M. 1989. Le potager tropical. 2nd Edition. Presses Universitaires de France, Paris, France. 580 pp.

24. Muyonga, J.H., Nabakabya, D., Nakimbugwe, D.N. and Masinde, D. 2008. Efforts to promote amaranth production and consumption in Uganda to fight malnutrition. In: Robertson G. L., Lupien J. R., editors. Using Food Science and Technology to Improve Nutrition and Promote National Development. Toronto, Canada: International Union of Food Science and Technology, pp. 1-10.

25. Nafiu, K.A., Togun, A.O., Abiodun, M. and Chude, V.O. 2005. Effect of NPK fertilizer on growth, dry matter production and yield of eggplant in southwestern Nigeria. *Agriculture and Biology Journal of North America*, 2(7): 1117-1125.

26. Nirmal, K.J.I., Hiren, S. and Rita, N.K. 2007. Characterization of Heavy Metals in Vegetables Using Inductive Coupled Plasma Analyzer (ICPA). *Journal of Applied Science* and *Environmental Management,* 11(3): 75-79.

27. Ogunyemi, S., Bamgbose, O.O. and Awodoyin, R.O. 2003. Heavy metal contamination of some leafy vegetables growing within Ibadan metropolis, South-western Nigeria. *Tropical Agricultural Research and Extension Journal,* 6(2):71-76.

28. Qadir, M. and Ghafoor, A. 1999. Irrigation with city effluent for growing vegetables – A silent epidemic of metal poisoning. *Pakistan Academy of Science,* 2: 219 -222.

29. Rattan, R.K ., Datta, S.,Chhonkar, P.K and Saribabu, K.2005.Long – term impact of irrigation with sewage effluent on heavy metal content in soils, crops and ground water- A case study in Agriculture. *Ecosystems and Environment,* doi:10. 01611

30. Rubatzky, V.E and Yamaguchi, M. 1997. World vegetables: Principles, production and nutritive values. 2nd ed. International Thomas Publishing, 1997. New York: Chapman and Hall.

31. Saunders, R. M. and Beckar, R. 1983. Amaranthus. In: Advances in Cereal Science and Technology Y. Pomeranz, (ed.) American Association of Cereal Chemistry, St., Paul, Minnesota, U.S.A Vol. 6 chapter 6.

32. Udedi, S.S. 2003. From Guinea Worm Scourge to Metal Toxicity in Ebonyi State, Chemistry in Nigeria as the New Millennium Unfolds, 2(2): 13–14.

33. US-EPA-IRIS, 2006. United State Environmental Protection Agency, Integrated Risk Information System. https//www.epa.gov/iris/subst.

34. Vanlaure, B., Diels, J., Deckers, J. and Lyasse, O. 2002. Fertility status of soils of the derived savanna and northern guinea savanna and response to major plant nutrients influenced by soil type and land use management. *Nutrient Cycling in Agroecosystems,* 62: 139-150

35. Weatherspark. Https.//weatherspark.com

36. WHO/FAO, 2001. Joint World Health Organisation/Food and Agricultural Organisation. Food standards programme codex committee on contaminant in foods, 5th session pp.64-89.