

# Transference Capacity of Some Heavy Metals in Cultivation-Consumption Chain of Plantain Harvested from Awka Etitu Anambra State

ENI-YIMINI SOLOMON AGORO<sup>\*1</sup>, OMOTAYO BABATUNDE ILESANMI<sup>2</sup>

<sup>1</sup>Directorate of Research and Quality Assurance, Federal University Otuoke, Bayelsa State Nigeria

<sup>2</sup>Department of Biochemistry, Faculty of Science, Federal University Otuoke, Bayelsa State Nigeria

## Abstract

**Background:** This research was carried out to investigate the potential bioaccumulation and translocation of heavy metals in plantain cultivation-consumption chain using the serum and vitreous humour as the final reservoirs. Soil, plantain, serum and vitreous humour were analyzed for heavy metals (HM) (Cr, Pb, Cd, As, Hg).

**Methods:** Twelve male albino rabbits were divided into two groups to constitute the sample size as validated by Mead's formula. The control group was fed with normal rabbit meal void of detectable HM sourced from the market, whereas the treatment group was fed with same normal meal along with plantain sourced from Awka Etitu Anambra State for ninety days. The heavy metal concentrations were analyzed using AAS. Data analysis was carried out using *t*-test through SPSS (version 22).

**Results:** The findings showed that soil and plantain cadmium concentrations in Awka Etitu were higher than World Health Organization (WHO) permissible limits. Similarly, heavy metals in the soil were higher than those found in plantain. Vitreous cadmium, chromium, and lead were higher in concentrations in the treatment group compared with the control, whereas arsenic concentration was the opposite. Similarly, serum cadmium and lead were lower in the treatment group, whereas chromium and arsenic were higher when compared with the control group.

**Conclusions:** Heavy metals bioaccumulation and translocation along cultivation-consumption chain is a reality and could be potentially disruptive to physiological processes in the body and the ecology in general.

**Keywords:** Metals, Awka Etitu, Bioaccumulation, Plantain, Serum

**How to cite this article:** Agoro ES, Ilesanmi OB. Transference Capacity of Some Heavy Metals in Cultivation-Consumption Chain of Plantain Harvested from Awka Etitu Anambra State. *Asia Pac J Med Toxicol* 2022; 11(1):19-24.

## INTRODUCTION

According to Kibria *et al.* [1], heavy metals are elements with an atomic weight greater than 20 and are characterized by similar atomic electronic configurations in the outer orbitals. They are harmful and usually persist and bioaccumulate in the environment and host organisms [2,3]. Arsenic (As), lead (Pb), cadmium (Cd), and Chromium (Cr) are the heavy metals of interest in this study. These metals are deleterious and affect several important organs of the body [4,2-3]. Based on this premise, the United States Environmental Protection Agency and the Agency for Toxic Substances and Disease Registry included these heavy metals amongst the 20 list of dangerous substances (ATSDR) [5-10]. Heavy metals are natural components of the environment, but human activities, notably industrial and mining processes, have been responsible for the wider diffusion of these elements. Plants are good soil quality indicators as they directly reflect soil quality [11].

Plantains (*Musa spp*) are plants producing fruits that remain starchy at maturity with more than 50% of worldwide

production [[12-14]. Nigeria is one of the largest plantains producing countries in the world [15]. Plantain thrives well in soils with high nutritional and waste polluted contents. This makes plantain a good indicator for heavy metal bioaccumulation and translocation. The issue of heavy metals and the contamination of soil and subsequent uptake and accumulation in food crops is rapidly becoming a major health concern as their presence pose serious health hazards to plants, animals, and humans [16].

The bioaccumulation and translocation of heavy metals from the environment to animals and then humans are pulsatile and installmental in nature. The portals of entry of these metals are quite different depending on the exposure, but the transportation in the body is through the traditional intracellular and extracellular fluids. Heavy metals picked up from food are transported to various parts of the body via the circulatory system. The blood is the major transporter of xenobiotics and nutrients in the body. Similarly, vitreous humour play a critical role in the transportation of ions and nutrients within the confines of the eyes [18-19]. These fluids are the choice for the assessment of heavy metal translocation and buildup in the body.

\*Correspondence to: Eni-yimini Solomon Agoro, PhD, Assistant Professor, Directorate of Research and Quality Assurance, Federal University Otuoke, Bayelsa State Nigeria

Tel: 08037434995, E-mail: enisagoro@gmail.com

Prolonged intake of heavy metals contaminated food has been implicated as a causative agent of cancer, nervous system disorders, cardiovascular diseases, renal problems, destruction of liver, lungs, and kidneys [19]. This research was, therefore, carried out to scrutinize the transference capacity of heavy metals (Pb, Cr, Hg, Cd and As) concentrations in the plantain cultivation-consumption chain. The chain includes soil, plantain, serum, and vitreous humour. Findings of this study are expected to provide further insights into the bioaccumulation, translocation, and distribution of heavy metals in soil, plantain, serum, and vitreous and its attendant potential effects amongst residents of Awka Etit.

## METHODS

### Research Setting

The soil and plantain samples were harvested from Awka Etit. Awka Etit, which is a village in Idemili South Local Government Area of Anambra State (Fig 1). The animal breeding and sacrifice were carried out at the Department of Biochemistry of the Federal University Otuoke, Bayelsa State. In a similar vein, the heavy metals analysis was conducted at the Eni-yimini Laboratories (eL) Ltd, Igbogene Epie, Bayelsa State.

### Research Design and Sample Size

Experimental research design was employed in this study including experimental and control groups. The sample size determination was derived from Mead's resource equation as postulated by Kirkwood and Robert, [20]. A total of twelve rabbits divided equally into control and treatment groups were used for the study. The collection of data was quantitative. Control group rabbits were fed with feeds void of studied heavy metals, whereas the treatment group diet included powdered unripe plantain sourced from various sites in Awka Etit. The plantains used for the study were of same specie as specified by a botanist in the Department of Biological Sciences of the Federal University otuoke. The study duration was put at three months excluding the two weeks of acclimatization. At the end of the three months, the rabbits were sacrificed mechanically following acceptable animal care rules.

### Selection Criteria

The rabbits used were all confirmed healthy and active by the university veterinarian. Rabbits showing signs and symptoms of illness and disorders were excluded. The research used only male albino rabbits of age range between seven to eight months and weight brackets of 1.8-2.0 kg. Soil and plantain samples were harvested randomly from central locations in Awka Etit into appropriate collectables aseptically.

### Ethical Approval

The ethical clearance was obtained from the Directorate of Research and Quality Assurance of Federal University Otuoke. The Animal Welfare Act of 1985 of the United State of America for Research and Institutional Animal Care and Use Committee (IACUC) protocols were stringently followed.

### Collection of Samples

#### a. Vitreous Humour and Blood

Vitreous humor samples were collected by the method of Coe [21] and that of blood by Ness [22]. Both samples were put into plain containers and centrifuged at 2000 rpm for ten minutes at 25°C. The supernatants were subsequently separated and used for the heavy metal analysis.

#### b. Soil

Twenty grams (20g) of soil sample was placed in a plain clean plastic container with a tight lid from several locations within Awka Etit. The samples were consequently transported to the laboratory for heavy metal analysis. All the processes involved in the collection were strictly void of contamination.

#### d. Plantain

Plantains harvested from Awka Etit were peeled, sun-dried, and grinded, and then put into a clean plastic container for laboratory analysis. Aseptic methods were strictly followed to avoid contamination during the collection and processing stages.

### Laboratory Procedure

Vitreous humour and serum samples with a total volume of a 1.2L each were thawed at room temperature and each sample was treated separately. A volume of 300µL of samples was added to 300µL of HNO<sub>3</sub> and 100µL of H<sub>2</sub>O<sub>2</sub>. Sample decomposition was then carried out in a water bath at 80°C for 30 minutes. After digestion/decomposition, samples were diluted to 10µL with deionized water. Similarly, 2g of soil or plantain samples were weighed and digested with aqua-riga for 1hour. The samples were allowed to cool and filtered into a 50ml volumetric flask after digestion was completed. The digest from all the prepared matrices were then analyzed using Varian Spectra A100 Atomic Absorption Spectroscopy (AAS) for Pb, Cd, Cr, Hg and As.

### Statistics

Data were analyzed using statistical package for social (SPSS) program (SPSS inc., Chicago, IL, USA; Version 18-12) and Microsoft excel. *T-test* was used in comparing the means of the two groups. The level of significance was pegged at less than or equal to 0.05.

## RESULTS

Table 1 compared mean concentrations of some heavy metals in soil and plantain between World Health Organization (WHO) permissible to those obtained in Awka Etit in Anambra State. The comparison shows that cadmium obtained from the matrices in Awka Etit was higher than the permissive limit both in soil and plantain. Table 2 reveals significant higher concentrations of all the studied heavy metals in soil when compared with that of plantain. Vitreous cadmium, chromium, and lead was higher in concentration in the treatment group compared with the control, whereas arsenic concentration was on contrary (table 3). Serum cadmium and lead were higher in the treatment group, whereas chromium and arsenic were lower when compared with the control (table 4). Table 5 shows higher concentrations of all the studied heavy metals in serum control when compared to vitreous control. Cadmium and chromium were higher in the vitreous treatment when compared to serum treatment, whereas lead was on the contrary (table 6).

**Table 1. World Health Organization (WHO) permissible limits for heavy metals in plant and soil comparable to Awka Etti Anambra State mean concentrations**

Metals	WHO Acceptable Limits(Soil)	Imiringi Soil	Plantain WHO Acceptable Limits	Plantain (Imiringi)
Cadmium (mg/kg)	0.80	1.95	0.02	0.22
Chromium (mg/kg)	100.00	3.185	1.30	0.49
Arsenic (mg/kg)	5.00	0.98	1.46	0.65
Lead (mg/kg)	85.00	0.93	2.00	0.47
Mercury (mg/kg)	2.00	0.0075	0.02	>0.0001

Source: WHO (1996)

**Table 2. Comparison of some heavy metal concentrations between soil and plantain sourced from Awka Etti Anambra State**

Metals	Soil	Plantain	t-test	p-value
Cadmium (mg/L)	1.952±0.1470	0.2219±0.0715	21.180	0.000
Chromium (mg/L)	3.185±1.630	0.4856±0.1622	3.297	0.016
Arsenic (mg/L)	0.9809±0.2157	0.6457±0.1623	5.573	0.000
Lead (mg/L)	0.9283±0.2147	0.2147±0.1243	3.713	0.010
Mercury (mg/L)	0.0075±0.007	>0.0001	5.733	0.001

**Table 3. Comparison of some heavy metal concentrations between vitreous of control and treatment**

Metals	Vitreous Control	Vitreous Treatment	t-test	p-value
Cadmium (mg/L)	0.0002±0.0001	0.0219±0.0086	-5.033	0.002
Chromium (mg/L)	0.0020±0.0008	0.2820±0.0778	-7.196	0.000
Arsenic (mg/L)	0.1600±0.0163	>0.0001	5.573	0.000
Lead (mg/L)	0.0020±0.0008	0.0363±0.0085	-7.752	0.000
Mercury (mg/L)	>0.0001	>0.0001		

**Table 4. Comparison of some heavy metal concentrations between serum of control and treatment**

Metals	Serum Control	Serum Treatment	t-test	p-value
Cadmium (mg/L)	0.0023±0.0009	0.0052±0.0018	-2.880	0.039
Chromium (mg/L)	0.6030±0.1604	0.0288±0.0128	7.135	0.000
Arsenic (mg/L)	0.432±0.1714	>0.0001	8.515	0.000
Lead (mg/L)	0.0255±0.0082	0.0903±0.0161	-7.173	0.000
Mercury (mg/L)	>0.0001	>0.0001		

**Table 5. Comparison of some heavy metal concentrations between serum of control and Vitreous of Control**

Metals	Serum Control	Vitreous Control	t-test	p-value
Cadmium (mg/L)	0.0023±0.0009	0.0002±0.001	-3.799	0.009
Chromium (mg/L)	0.6030±0.1604	0.0020±0.0008	-7.491	0.000
Arsenic (mg/L)	0.432±0.1714	0.1600±0.0163	-3.167	0.019
Lead (mg/L)	0.0255±0.0082	0.0020±0.0008	-5.686	0.001
Mercury (mg/L)	>0.0001	>0.0001		

**Table 6. Comparison of some heavy metal concentrations between serum of treatment and Vitreous of treatment**

Metals	Serum Treatment	Vitreous Treatment	t-test	p-value
Cadmium (mg/L)	0.0052±0.0018	0.0219±0.0086	4.346	0.005
Chromium (mg/L)	0.0288±0.0128	0.2820±0.0778	-6.423	0.001
Arsenic (mg/L)	>0.0001	>0.0001		
Lead (mg/L)	0.0903±0.0161	0.0363±0.0085	6.045	0.001
Mercury (mg/L)	>0.0001	>0.0001		

## DISCUSSION

The concentrations of heavy metals in an environment are measures of quality and safe inhabitation. An environment littered with above permissive limit of heavy metals is vulnerable to several diseases and ecological strains. The findings of this study point to vulnerability of inhabitants and ecological niche of Awka Etitu to potential long-term environmental disruptions associated with chronic heavy metals intoxication. The soil-to-plant transfer of heavy metals is a very important step in the trophic transfer in food chains. These metals are taken up by plants from polluted soil and subsequently transferred to herbivorous animals along the food chain [24]. Regarding the contamination of the human food chain, contamination of crops such as plantain is a very serious issue. Below is an empirical discourse on the transference capacity of some heavy metals within the plantain consumption chain;

The mean concentration of cadmium was the only heavy metals observed in soil and plantain that was above the permissive limit as established by World Health Organization (WHO) (table 1). Other parameters were detected but below the permissive limit, though with tendencies of long-term build up if the instigators resulting to the disposition are not halted. Heavy metal presence in an environment is a product of a lot of factors such as industrial encroachment, crude oil exploration, automobile repairs and many others [25-27]. The finding implies that the environment of Awka Etitu is predisposed to cadmium intoxication which is could be ecologically disruptive in the long-run.

Moreover, this study revealed higher concentrations of studied heavy metals in soil when compared to plantain (table 2). This is considerably not untrue due to the fact that plants derive its nutrients and other ancillary needs from the soil. The higher concentration of arsenic and other metals in soil and plantain harvested from Awka Etitu could be due to human activities such as pesticide use, mining, industrial activities, automobile emission, operating coal burning power plants, and waste disposal. Many heavy metals are environmentally stable and non-biodegradable, toxic to the living beings, plants, and animals [25]. Plants are known to bio-accumulate heavy metals from soil or atmosphere of which plantain is inclusive. A number of studies on metals accumulation in plants from cultivation sources have been previously reported [28-32]. This aspect of the study inferred that the translocation of heavy metals from soil to cultivated plants are a reality and could further pose risks to herbivores and other dependents.

Vitreous concentrations of cadmium, chromium, and lead were higher in the treatment group when compared with the control, whereas arsenic concentration was lower (table 3). The increase observed for vitreous chromium, cadmium, and lead in the treatment group could be as the result of bioaccumulation from the consumption of plantain grown in Awka Etitu. This has further consolidated the fact about the bioaccumulation and translocation of heavy metals within the food chain enclave. The detection of heavy metal in the vitreous is affirmations of possible heavy metal induce damage on the eye. A lot of eye diseases are without a known cause. The detection of heavy metals in the vitreous could be one of the reasons of the geometric increase in eye diseases and disorder. The presence of significant arsenic in the vitreous of the control rabbits call for a further probe, but a plausible reason could be as a result of environmental influence or other idiopathic reasons. This finding is in line with positions of Erie *et al.* [33], Erie, *et al.* [34] and Pamphlett *et al.* [35]. The stance of Pamphlett *et al.* [25] posited the hypothesis that accumulations of toxic metals in the retina could contribute to the pathogenesis of age-related macular degeneration.

Similarly, serum cadmium and lead was higher in the treatment group when compared with the controls, whereas chromium and arsenic were lower (table 4). The presence of cadmium and lead could be mostly due to the consumption of plantain diet, whereas that of the control could be environmentally induced or idiopathic. Though, both were exposed to similar environmental conditions, the presence of certain heavy metals in the controls and not in the treatments cannot be explained. However, the case of chromium could be different as it is important in a lot of metabolic processes. The fall in serum chromium in the treatment group could be due to displacement by other heavy metals. This is an affirmation of the displacement mechanisms known in the chemistry of heavy metals. The findings of Oves *et al.*, [36] and Hazrat *et al.* [37] concur with those obtained in this study.

A comparative analysis between the studied heavy metals in serum of the control and that of the vitreous revealed a significant increase in former (table 5). This stance was quite different in comparison between heavy metals in the treatment serum to that of vitreous treatment. The increase in serum compared with vitreous of the controls could be attributed to the restrictive attribute of the eye structure to molecular movements in a physiological state. This was on the contrary in the treatment group. The concentrations of cadmium and chromium were higher in the vitreous treatment

when compared to serum treatment, whereas lead was on the contrary (table 6). These alterations in concentrations between serum and vitreous in compositions of heavy metals of the treatment category could be as the result of the differences in the rate of ingestions, metabolism, storage, and interactions with biochemical transporters and transportation media. In a nutshell, the study has clearly shown the transmissibility of heavy metals between and within the biota milieu and ecosystems.

The limitations of the study were basically the paucity of funds and accessibility to reagents and more advanced instrumentation.

## CONCLUSION

Heavy metals are ubiquitous in terrestrial ecosystems. The ferocious effect of heavy metals is dependent on the concentration and its bioaccumulation index in the body. This study has shown that heavy metals are truly transmissible from soil to plant and then to the serum and vitreous of animals and humans. The presence of heavy metals in the body irrespective of the concentration is a potential pointer to bioaccumulation and potential systemic disruption. Metal such as cadmium was found to be higher in soil and plantain of the study location when compared with the permissive acceptable limit by the World Health Organization. Similarly, heavy metals such as cadmium, chromium, lead, and arsenic were detected in the various matrices of the study samples. The detection of these heavy metals in the serum and vitreous humour of rabbits upon consumption of plantain sourced from the study location is a further validation of the ease of translocation of heavy across matrices, membranes, and other restrictive barriers. This makes heavy metals a potential disruptor of the ecological and systemic status quo.

## Recommendations

A comprehensive study of the interactions and the transference of heavy metals in soil, plants, and animals show that serious steps should be taken to minimize the impact of these elements on human health and the environment. The following recommendations are therefore made :(i) Background concentrations of heavy metals should be analyzed and documented in the different environmental media around Nigeria to create a cut-off point regarding the permissive acceptable limits.(ii)The levels of potentially toxic heavy metals in crops, foods, water bodies, sediments, soils, and the resident biota should be assessed and monitored regularly.(iii) The public should be educated about the harmful effects of toxic heavy metals on human health and the environment.(vi) Scientific research on environmental assessment on toxic heavy metals should be encouraged and promoted by the allocation of appropriate funds for protecting human health and the environment.(vii) Further studies can be done in the future using this research design to replicate the current research and provide and incorporate a human model to validate this line of thought.

**Conflict of interest:** None to be declared.

## Author's Contribution

All authors contributed to the conception and design of the work. Conception and design, Interpretation of the results and revisions performed by E.S. Agoro. OB Ilesanmi performed data acquisition and analysis , and literature search and gathering. All authors read and gave final approval of the manuscript and are accountable for the originality of the work.

**Funding and support:** None

**Ethical approval:** The study protocol was approved by the Directorate of Research and Quality Assurance of the Federal University Otuoke, Bayelsa State, Nigeria. The ethical principles for medical research involving animal subjects as outlined in the Helsinki declaration in 1975 and subsequent revisions were strictly adhered to in the course of this study.

## REFERENCES

1. Kibria G, Haroon AKY, Nugegoda D. Climate Change and Chemicals-Environmental Biological aspects. 2010.
2. Kibria G, Hossain MM, Mallick D, Lau TC, Wu R. (2016a). Monitoring of metal pollution in waterways across Bangladesh and ecological and public health implications of pollution. *Chemosphere*. 2016; 165:1-9.
3. Kibria G, Hossain MM, Mallick D, Lau TC, Wu R. (2016b). Trace/heavy metal pollution monitoring in estuary and coastal area of the Bay of Bengal, Bangladesh and implicated impacts. *Marine Pollution Bulletin*. 2016b; 105:93-402.
4. Gall JE, Boyd RS, Rajakaruna N. Transfer of heavy metals through terrestrial food webs: a review. *Environ. Monit. Assess*. 2015;187:201.
5. ATSDR. Toxicological Profile for Barium U.S. Department of Health and Human Services, Public Health Service, Atlanta, GA (2007).
6. Xiong T, Austruy A, Pierart A, Shahid, M. Kinetic study of phytotoxicity induced by foliar lead uptake for vegetables exposed to fine particles and implications for sustainable urban agriculture *J. Environ. Sci*. 2016a; 1-12.
7. Xiong T, Dumat C, Pierart A, Shahid M, Kang Y, Li N, et al. Measurement of metal bioaccessibility in vegetables to improve human exposure assessments: field study of soil-plant-atmosphere transfers in urban areas, South China. *Environ. Environmental Geochemistry and Health*. 2016b; 38 (6):1283-1301
8. Khaled A. Trace metals in fish of economic interest from the west of Alexandria, Egypt. *Chemistry and Ecology*. 2009; 25(4): 229-246, 2009.
9. Rai PK (2018). Heavy metal phyto-technologies from Ramsar wetland plants: green approach. *Journal of Chemical Ecology*. 2018; 34 (8): 786-796.
10. Marschner P. (2012). Marschner's Mineral Nutrition of Higher Plants. (3rd ed.), Academic, London (2012)
11. Kabata PA, Mukherjee A.Trace Elements from Soil to Human. Springer-Verlag, Berlin. 2007.
12. Marrio J, Lancaster PA. Bananas and plantains. In: H.T. Chan (ed.), *Handbook of Tropical Food*. Dekker, New York. 1983; pp 85-143.
13. Robinson JC. Bananas and Plantains. CAB International, Wallington. 1996.
14. Food and Agriculture Organization. Production Yearbook 1990. FAO, Rome.1990

15. Food and Agriculture Organization. Production Yearbook 2004. FAO, Rome.2006
16. Khan S, Cao Q, Zheng YM, Huang YZ, Zhu YG. Health risks of heavy metals in contaminated soils and food crops irrigated with wastewater in Beijing, China. *Environmental Pollution*. 2008; 152(3):686–692.
17. Helmenstine AM. Heavy metals definition. About.com. 2014.
18. Agoro ES, Wankasi MM, Ombor OJ. Biochemical Patterns of Cardio-renal Biomarkers in Serum and Vitreous Humor of Rabbits after Chronic CO Exposure. *Ann Environ Sci Toxicol*. 2019; 3(1): 001-006.
19. Agoro ES, Ben-Wakama EN, Alabrah pW. Review on chronic impacts of carbon monoxide intoxication on some routine vitreous and blood investigations. *Toxicology and Forensic Medicine – Open Journal*. 2020; 5(1): 16-25.
20. Kirkwood J, Robert H. The UFAW Handbook on the Care and Management of Laboratory and Other Research Animals. Wiley- Blackwell. 2010; 29.
21. Coe JI. Vitreous potassium as a measure of the postmortem interval: an historical review and critical evaluation. *Forensic Science International*.1989; 42:201-13.
22. Ness RD. Clinical pathology and sample collection of exotic small animals. *The Veterinary Clinics of North America: Exotic Animal Practice*. 1999; 2(3), 591-620.
23. WHO Permissible limits of heavy metals in soil and plants (Geneva: World Health Organization), Switzerland. 1996.
24. Nica DV, Bura M, Gergen I, Harmanescu M, Bordean DM. Bioaccumulative and conchological assessment of heavy metal transfer in a soil-plant-snail food chain. *Chemistry Central Journal*. 2012; 6(1):55. 2012.
25. Vijaya CB, Kiran K, Nagendrappa G. Assessment of Heavy Metals in Water Samples of Certain Locations Situated Around Tumkur, Karnataka, India. CODEN ECJHAO E- Journal of Chemistry.2010; 7(2): 349-352.
26. Akhilesh J, Savita D, Suman M. Some Trace Elements Investigation in Ground Water of Bhopal and Sehore District in Madhya Pradesh: India. *Journal of Applied Sciences and Environmental Management*. 2009; 13(4): 47 – 50.
27. Xilong W, Sato T, Baoshan X, Tao S. Health risk of heavy metals to the general public in Tianjin, China via consumption of vegetables and fish. *Science of the total Environment*. 2005; 50: 28-37.
28. El Hamiani O, El Khalil H, Sirguy C, Ouhammou A, Bitton G, Schwartz C, et al. Metal concentrations in plants from mining areas in South Morocco: health risks assessment of consumption of edible and aromatic plants. *Clean: Soil, Air, Water*. 2015; 43 (3): 399-407
29. Bolan S, Kunhikrishnan A, Seshadri B, Choppala G, Naidu R, Bolan NS, et al. Sources, distribution, bioavailability, toxicity, and risk assessment of heavy metal(loid)s in complementary medicines. *Environ Int*. 2017;108:103-118.
30. Kim HS, Kim K-R, Kim W-I, Owens G, Kim K-H. 2017. Influence of Road Proximity on the Concentrations of Heavy Metals in Korean Urban Agricultural Soils and Crops. *Archives of Environmental Contamination and Toxicology*. 2017; 72(2): 260–268.
31. Kim SW, Chae Y, Moon J, Kim D, Cui R, An G, et al. In Situ Evaluation of Crop Productivity and Bioaccumulation of Heavy Metals in Paddy Soils after Remediation of Metal-Contaminated Soils. *J Agric Food Chem*. 2017;65(6):1239-1246. doi: 10.1021/acs.jafc.6b04339. Epub 2017 Feb 2. PMID: 28150951.
32. Kohzadi S, Shahmoradi B, Ghaderi E, Loqmani H, Maleki A. Concentration, Source, and Potential Human Health Risk of Heavy Metals in the Commonly Consumed Medicinal Plants. *Biol Trace Elem Res*. 2019; 187, 41–50
33. Erie JC, Good JA, Butz JA. Excess lead in the neural retina in age-related macular degeneration. *American Journal Ophthalmology*. 2009; 148(6):890-4.
34. Erie JC, Butz JA, Good JA, Erie EA, Burritt MF, Cameron JD. Heavy metal concentrations in human eyes. *American Journal Ophthalmology*. 2005; 139(5):888-93.
35. Pamphlett R, Cherepanoff S, Too LK, Kum Jew S, Doble PA, Bishop DP. The distribution of toxic metals in the human retina and optic nerve head: Implications for age-related macular degeneration. *PLoS One*. 2020; 29;15(10):0241054.
36. Oves M, Khan MS, Zaidi A, Ahmad E. (2012). Soil contamination, nutritive value, and human health risk assessment of heavymetals: an overview A. Zaidi, P.A. Wani, M.S. Khan (Eds.), *Toxicity of Heavy Metals to Legumes and Bioremediation*, Springer, New York. 2012; pp. 1-27
37. Hazrat A, Ezzat K, Ikram I. Environmental Chemistry and Ecotoxicology of Hazardous Heavy Metals: Environmental Persistence, Toxicity, and Bioaccumulation. *Journal of Chemistry*. 2019; <https://doi.org/10.1155/2019/6730305>