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Health Risk Assessment of Heavy Metal Toxicity Utilizing Eatable Vegetables from Poultry Farm Region of Osun State

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Abstract Heavy metals are present everywhere in the environment attributable to both geogenic and anthropogenic inputs and humans are vulnerable to them via various exposure pathways. Heavy metal accumulation in soils and plants is of grave concern because of the possible human health risks. This research article examined the seasonal concentrations of arsenic, cadmium, copper, iron, lead and zinc in leafy vegetables of *Talinum triangulare* and *Vernonia amygdalina* samples planted in some poultry farms and determine the risks involved in consumption of that vegetables. Flame Atomic Absorption Spectrophotometer was applied to evaluate the contents of the metals in the vegetables. Quality monitoring procedures comprised blank analysis, recovery test and calibration of standards. Descriptive and inferential statistics were adopted for data analyses. Hazard quotient, daily intake of metals, health risk index and transfer factor were also determined. The pattern of metals mean value in the soil was As > Fe > Zn > Pb > Cu > Cd while for vegetables was Fe > Zn > As > Cu > Pb > Cd. The TFs pattern for metals from soil to plant was Zn > Fe > Cd > As > Cu > Pb for *Talinum triangulare* and Fe > Zn > Cd > As > Cu > Pb for *Vernonia amygdalina* in both seasons. The sequence of the DIM value was Fe > Zn > As > Cu > Pb > Cd for both plants. The results indicated very high HRI and THQ values for As (22.60 and 61.00). This asserted that the intake of vegetables planted on the soil of the farms studied present a health risk for humans with regard to the evaluated metals.

Keywords: *metals, hazard quotient, health risk index, vegetable*

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1. Introduction

Heavy metals are present everywhere in the environment owing to both geogenic and anthropogenic inputs and humans are vulnerable to them via various pathways [1]. Poultry manure has substantial levels of nutrients like nitrogen, phosphorus, potassium, calcium, magnesium, organic matter and other excreted materials like vitamins, hormones, antibiotics, pathogens and heavy metals which are added by the use of feeds [1]. Organic manure comprises considerable amounts of potentially toxic metals, like As, Cd, Cu and Zn [2]. In very large amount, these elements can become toxic to plants, adversely affect organisms that feed on these plants and enter water ecosystems via surface run-off and seepage [3]. Heavy metals are introduced into poultry diets either inadvertently via contaminated feedstuffs or deliberately, as feed preservers applied to supply animals' requirements or – in much larger quantities – as veterinary medicines or growth enhancers [3]. Metals like Pb, Hg, Cd, and As are accumulated poisons, which cause environmental threats and are described to be extremely toxic [4].

Metals like Fe, Cu, Zn and Mn are necessary metals for humans, since they perform an essential role in organism systems, nevertheless the indispensable heavy metals can present harmful effects when their intake is extremely high [5]. Extreme buildup of heavy metals in agricultural soil by dint of poultry manure may not only give rise to soil contamination, but also bring about elevated heavy metal absorption by crops, and thus affect food quality and protection [6].

Heavy metal cumulation in soils and plants is of deeply concerned because of the possible human health risks. Food chain contamination is one of the crucial pathways for the entrance of these toxic pollutants into the human body. Heavy metal amassing in vegetables largely counts not merely on the texture of soil or media on which they grow, but also upon plant species and nature of plant, and the proficiency of varied plants in taking up metals is estimated by either plant uptake or soil-to-plant translocation factors of the metals [7].

Vegetables grew in poultry farm soils take up heavy metals in large enough extents to cause potential health hazards to the consumers. In a sequence to estimate the health risks, it is required to point out the potential of a causer to present risk agents into the ecosystem, estimate the amount of risk agents that come into contact with the human-ecosystem limits and quantify the health consequence of the exposure [7]. In line with National Research Council [8,9], this process encompasses four steps which are hazard identification, dose/response analysis, exposure quantification and hazard characterization. Chronic level intake of toxic metals has negative effects on humans and the associated harmful effects become apparent merely after many years of contact with [8]. However, the intake of heavy metal-contaminated food can seriously deplete some vital nutrients in the body that are further accountable for diminishing immunological defenses, intrauterine growth delay, impaired psycho-social abilities, disabilities inherent in malnutrition and high incidence of upper gastrointestinal cancer rates [7].

Poultry farming is a widespread practice in the world and recently a number of reports have been published on poultry farm soils contaminated with heavy metals [3,10]. Nevertheless, an additional insight into metal uptake, cumulation and assessment of human health hazard inherent in poultry farm soils is still needed. This study was conducted to investigate the appropriateness of poultry farm soils for vegetable planted and to assess the metal absorption by commonly consumed vegetables within the vicinity of some poultry farms in Osun State and also estimate the potential health risks inherent in human intake of vegetables contaminated with heavy metals. In this research *Talinum triangulare and Vernonia amygdalina* are selected for their heavy metal accumulation and translocation as a result of their consumption widely admired among the people of the area.

2. Materials and Methods

2.1. Study Area Depiction and Aptness

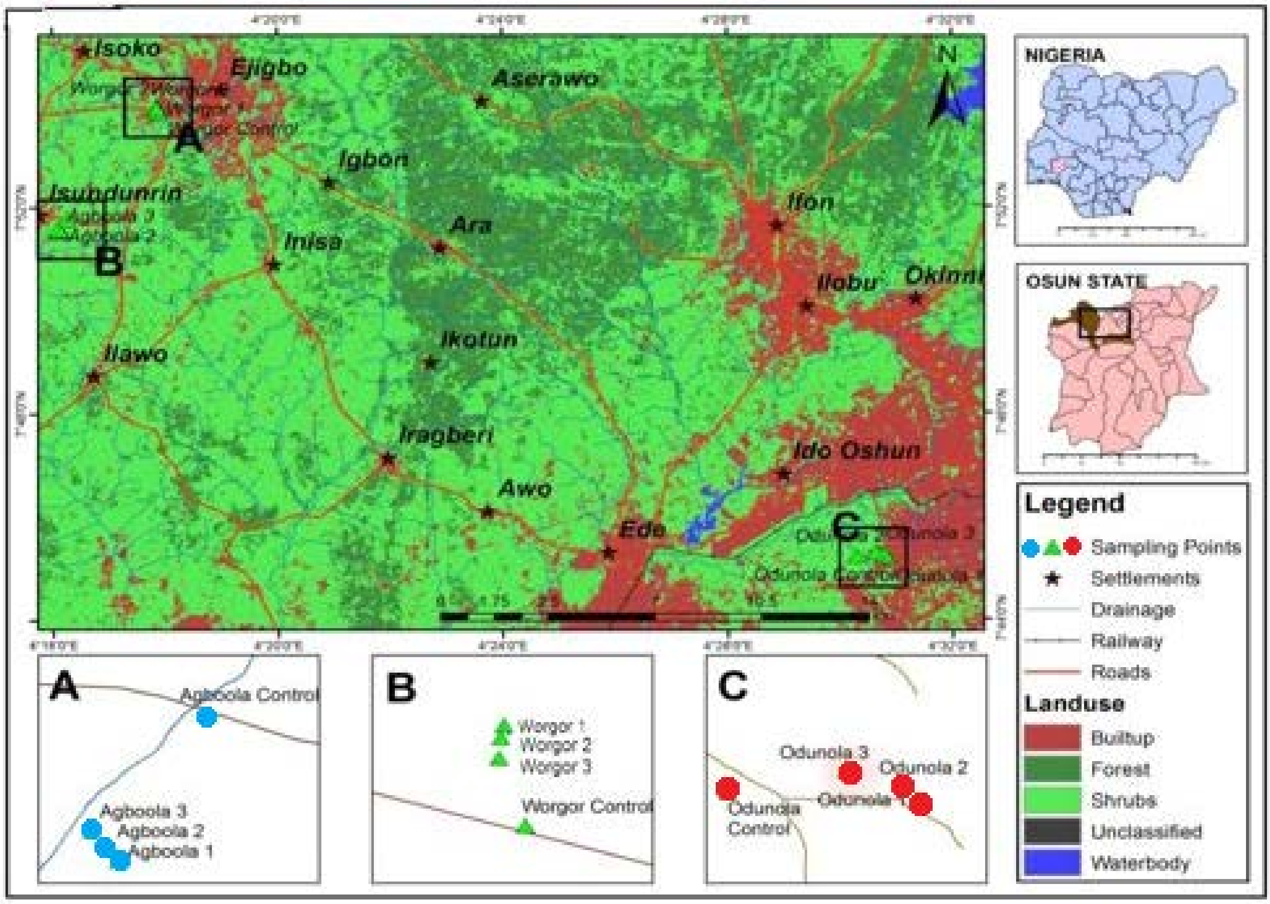

Figure 1. Map of the Study Area Revealing Sample Locations

The study area involved Ejigbo, Isundunrin and Osogbo poultry farms in Osun State, Southwestern Nigeria (Figure 1). Osun State covers an area (land mass) of about 14,875 square kilometers and positions between longitude 04°00'E and 05° S and latitude 05°558 'N and 08°07"W and has derived savanna vegetation, identified with Precambrian crystalline basement complex rocks. The prevailing rocks make up sets of gneisses and quartzite [11]. The estimated population for year 2009, obtained from the 2006 census is 3, 416,959 [12]. It is bordered by Ogun, Kwara, Oyo, Ondo and Ekiti States in the South, North, West and East, respectively. The tropical climate of the state has two largely characterized seasons: wet or rainy season that starts in March or April and ceases in October, and dry season that lasts between November and March. It has a temperature range of 21.80°C–31.40°C with a moderately high humidity. The annual rainfall varies from 1110 mm in the northern parts to 1277.70 mm in the southern parts [13]. The main livelihood of the people in the state is agriculture which offers 75% of employment to the people of the state [13]. The state is highly praised with ecological factors that foster people in the state to venture into animal rearing, like poultry; piggery; rabbitry; cane-rat; dairy; goat; sheep; bee keeping and honey production; snail domestication; fishing among others [13]. The sampling stations adopted for this study were considered appropriate because all of them have been in operation for more than thirty years and long-term land use history is familiar. Besides, seasonal study of heavy metal contamination of the soils and vegetables of the poultry farms selected was being investigated for the first time.

2.2. Soil Sampling and Pretreatment

Five sampling spots at a distance of 10 m from each other were figured out from which soil samples were taken within a given sampling site. Samples were taken during the dry season of 2014 and in the wet season of 2015 applying a clean stainless soil auger graduated from 0-15 cm depths. The sub-samples were taken along autonomous zig-zag paths to attain arbitrariness and lumped together to make a composite sample. The soil auger was carefully cleaned after each sampling exercise, to prevent contamination. The soil sampling sites were cleared of debris before sampling and labeled suitably. The soil samples were air-dried for seven days to stop microbial decompositions. Prior to analysis, the soil samples were crushed in a porcelain mortar and sieved by employing a 2 mm plastic sieve to get fine soil particles and then stored in labeled polyethylene bags until analysis [14].

2.3. Collection and Pretreatment of Vegetable Samples

Vegetable samples were picked up with gloved hands to avoid contamination. Samples of green vegetable bitter leaf (*Vernonia amygdalina*) and water leaf (*Talinum triangulare*) are taken by arbitrarily collecting some mature bottom leaves from the matured plants till a sizable bundle was assembled from each farm where soil samples were taken and one fallow plot where poultry manure was not dropping, respectively within the area under study. Samples from the non-poultry manure dropping farming plot were collected and assayed to provide reference data as an underlying condition of comparison to the farming areas. On getting to the research laboratory, per variety of vegetables, collections from each zone were washed with running tap water and distilled water to eliminate dirt and other particulate matters. Each collection or bundles were then sub-divided to provide triplicate samples weighing nearly 100 g fresh content and air-dried for 24 to 48 hours to prevent biochemical alterations. Previously to analysis, fresh vegetable samples were oven-dried to a constant scale at a temperature of 65^{o}C so as to prevent enzymatic action. Samples were then pulverized with a porcelain mortar, sieved utilizing 1 mm mesh sieve, and stored in polyethylene bags made ready for digestion and FAAS analysis.

2.4. Digestion of Samples

Carefully weighed 1 g of the air-dried (<2 mm) composite soil sample from each of the three areas (in triplicates) were first moistened with a few drops of water (to evade sputtering) next by the addition of 10 cm^3 concentrated nitric acid (HNO_3). The mixture was gradually heated over a period of 1 hour, on a hot plate. The solid residue got was digested with 20 cm^3 of a 3:1 mixture of concentrated HNO_3 and $HClO_4$ for ten minutes at ambient temperature before reheating was continued. The temperature of the hot plate was gradually heightened over a period of 1 hour till the fumes of $HClO_4$ begins to emit. Heating and restocking with acid mixture continued till a transparent solution was attained. The mixture was let to cool to ambient temperature. The cooled mixture was then quantifiably transferred into a 50 cm^3 volumetric flask and filled in to mark with distilled water. The digested samples were stored in polyethylene bottles in preparation for FAAS quantification.

Likewise, 1 g of oven-dried and ground sample per variety of vegetable from each of the three regions were weighed (in triplicates) into a 100 cm^3 Teflon beaker. This was followed by the inclusion of 10 cm^3 mixtures of analytical grade acids, HNO_3 and $HClO_4$, in the ratio 3:1. The beakers were then protected with watch glasses and stayed overnight. Digestion was done on a hot plate at a temperature of about 90°C in a fume cupboard till about 4 cm^3 of the mixture was retrieved in the beaker. A 10 cm^3 of the acid mixture was then inputted and evaporated to a volume of about 4 cm^3 while still on hot plate, yielding a visible solution. The solution procured was cooled to ambient temperature and quantifiably transferred into a 50 cm^3 volumetric flask where it was filled in to the mark utilizing distilled water. These were stored in polyethylene bottles before analysis.

2.5. Quality Monitoring Measures Used

With the intention of determining the effectiveness of the HNO_3 – $HClO_4$ method of sample digestion, a recovery test was done by spiking 1 g of twenty-four (24) and ten (10) different soil and vegetables samples each with 1 cm^3 of standard solutions of the metals As, Cd, Cu, Fe, Pb and Zn. Recovery evaluation presented % recoveries > 85%. Metal contents in functioning standards and digested samples were performed with FAAS (Alpha Star Model 4, Chem Tech Analytical) at the Centre for Energy Research and Development (CERD) of the Obafemi Awolowo University, Ile-Ife, Nigeria. Instrumental conditions are as stated earlier [14]. Blanks were also prepared to determine the input of reagents to metal contents.

2.6. Reagents Made Use of and their Sources

All the reagents employed, namely nitric acid (HNO_3), (Riedel-deHaen, Germany), hydrochloric acid, (HCl), (Sigma-Aldrich, Germany), hydrofluoric acid (HF), (British Drug House, BDH, Chemical Ltd, Poole, England), sulphuric acid (H_2SO_4), (Sigma-Aldrich, Germany), perchloric acid ($HClO_4$), (Sigma-Aldrich, Germany), acetic acid (HOAc), (Sigma-Aldrich, Germany) and doubly distilled water, were of analytical grade. These were applied to formulate standard solutions.

2.7. Data Analysis

2.7.1. Health Risk Assessment

Risk to human health by the intake of metal contaminated vegetables was characterized utilizing [15] hazard quotient (HQ). This is a relation of determined dose to the reference dose (RfD). The consumers will be subjected to no risk if the proportion is below 1 and if the proportion is equivalent or above 1, so the indigenous inhabitants will undergo health risk. This risk assessment method takes on the subsequent equation:

$$HQ = \frac{[W_{plant}] \times [M_{plant}]}{RfD \times B_0} \quad (1)$$

where W_{plant} is the daily intake of vegetables (µg per day), M_{plant} is the content of metal in the vegetable (µg/g), RfD is the oral reference dose for the metal (µg/g of body weight per day), and Bo is the human body weight (kg).

The values of RfD for heavy metals were obtained from Department of Environment, Food and Rural Affairs [16] and Integrated Risk Information System [17].

2.7.2. Translocation Factor (TF)

The transfer ability of heavy metals from soil to the edible part of vegetables was largely defined utilizing the translocation factor [18]. Translocation factors (TF) of heavy metals were conducted in this manner:

$$TF = \frac{C_{plant}}{C_{soil}} \quad (2)$$

where C_{plant} and C_{soil} signify the heavy metal content in extract of vegetables and metal content in soil from the place where the vegetables were planted, respectively.

2.7.3. Daily Intake of Heavy Metal (DIM)

The daily intake of metals (DIM) was performed to generally estimate the daily metal loading into the body system of a specified body content of a consumer. This will make known the potential phyto-availability of metal. This does not take into consideration the likely metabolic ejection of the metals but can simply tell the likely ingestion rate of a precise metal. The daily intake of metal in this assessment was conducted obtained from the formula prescribed by Tsafe *et al*. [19]:

$$DIM = \frac{C_{metal} \times C_{rate} \times D_{food\ intake}}{BW(_{mean\ weight})} \quad (3)$$

where C_{metal} = heavy metals content in vegetables (µg/g), C_{rate} = conversion rate, $D_{food\ intake}$ = daily intake of vegetables.

The converting rate of 0.085 is to convert fresh vegetable weight to dry matter [19], whilst the average daily vegetable intake rate was determined by conducted an assessment where 100 people possessing average body mass of 60 kg were enquired for their daily intake of precise vegetable from the study region.

2.7.4. Health Risk Index (HRI)

By utilizing daily intake of metals (DIM) and reference oral dose (RfD), we obtained the health risk index. The subsequent formula was applied for the determination of HRI [15]:

$$HRI = \frac{DIM}{RfD} \quad (4)$$

where DIM is daily intake of metals and RfD is the oral reference doses. Oral reference doses were 3.00E-04, 1.00E-03, 4.00E-02, 3.00E-01, 4.00E-3 and 3.00E-01 µg/g/day for As, Cd, Cu, Fe, Pb and Zn, respectively [15,17]. If the concentration of HRI is not above 1 so the susceptible inhabitants is indicated to be free from risk [19].

3. Results and Discussion

Table 1 to Table 6 reveal the findings of the variables examined representing content of mean heavy metals in soil and vegetable samples, TF, HQ, DIM and HRI extracted from two distinct vegetables in dry and wet seasons. The values of the metals were generally higher in samples from the study site than the control site. There are also seasonal variations between dry and wet seasons samples. Heavy metal contents values were higher in soil samples in comparison to vegetables samples. Gupta *et al*. [35] recorded that the concentration of heavy metals in vegetables were usually lower than the soil samples. These results may be ascribed to root mechanism which implies to serve as a measure for bioavailability of metals (Table 2). It also signifies that other soil characteristics and plant physiologic processes perform a significant function in trace element translocation and absorption. In the same vein, from the Tables below vegetables harvested during dry seasons were heavily contaminated, while those harvested during wet seasons had lesser content of all the elements (Table 2). This could be ascribed to wet season rainfall which washed off contaminants from the vegetables.

Table 1. Mean Values (μgg-1) of Heavy Metals in Poultry Farm Soil of Area under Study (Dry and Wet Seasons)

Site	As	Cd	Cu	Fe	Pb	Zn
			Dry Season			
Agboola	855.89	1.84	92.64	793.34	76.09	243.12
Control	NA	NA	14.60	60.34	NA	14.40
Worgor	562.96	1.68	83.20	723.10	62.38	233.20
Control	NA	NA	10.70	48.82	NA	10.42
Odunola	516.98	1.40	70.40	687.24	88.40	265.00
Control	NA	NA	13.26	38.72	NA	18.58
Range	NA-855.89	NA-1.84	10.70-92.64	38.72-793.34	NA-88.40	10.42-265.00
Overall mean ± SD	643.94±101.25	1.64±0.08	47.47±6.25	391.96±96.09	75.62±10.63	130.79±16.72
			Wet Season			
Agboola	956.78	2.87	95.36	1146.92	102.17	312.50
Control	NA	NA	15.60	80.58	NA	20.50
Worgor	751.82	2.43	90.16	950.54	95.14	299.94
Control	NA	NA	11.80	63.00	NA	17.00
Odunola	762.86	2.22	119.80	1006.32	103.18	314.10
Control	NA	NA	17.00	45.20	NA	22.84
Range	NA-956.78	NA-2.87	11.80-119.80	63.00-1146.92	NA-103.18	17-314.10
Overall mean	823.82±106.12	2.51±0.13	58.29±8.63	548.76±100.41	100.16±10.75	164.48±18.45
***Elemental Conc. of a typical soil** *	7.20	0.35	25	26000	19	60
[21]	20-40	1-3	50-140	400-5000	50-300	150-300

Source: *Weil and Brady, [24]; NA= Not Available; [21].

Table 2. Mean of Heavy Metals Content (μgg-1) in Vegetables Grown in Poultry Farm Sites of Area under Study (Dry and Wet Seasons)

Site	Scientific Name	Common Name	As	Cd	Cu	Fe	Pb	Zn
				Dry Season				
Agboola	*Talinum triangulare*	Water leaf	25.50	0.09	0.56	165.60	0.13	68.54
	Vernonia amygdalina	Bitter leaf	9.96	0.04	0.60	235.15	0.10	18.55
Control			NA	NA	0.02	4.40	NA	8.20
Worgor	*Talinum triangulare*	Water leaf	33.00	0.10	0.62	193.40	0.28	94.94
	Vernonia amygdalina	Bitter leaf	10.98	0.06	0.64	240.05	0.08	20.50
Control			NA	NA	0.04	8.60	NA	11.60
Odunola	*Talinum triangulare*	Water leaf	38.50	0.13	0.76	203.20	0.16	83.14
	Vernonia amygdalina	Bitter leaf	14.96	0.05	0.82	250.05	0.15	30.54
Control			NA	NA	0.04	14.80	NA	9.40
				Wet Season				
Agboola	*Talinum triangulare*	Water leaf	23.10	0.06	0.54	139.34	0.08	58.20
	Vernonia amygdalina	Bitter leaf	8.90	0.05	0.34	152.22	0.03	18.57
Control			NA	NA	0.02	7.60	NA	8.80
Worgor	*Talinum triangulare*	Water leaf	30.15	0.11	0.58	140.32	0.07	64.40
	Vernonia amygdalina	Bitter leaf	7.78	0.04	0.40	163.13	0.06	24.53
Control			NA	NA	0.02	10.20	NA	10.60
Odunola	*Talinum triangulare*	Water leaf	28.11	0.10	0.44	144.96	0.09	73.20
	Vernonia amygdalina	Bitter leaf	12.77	0.06	0.52	204.30	0.08	26.51
Control			NA	NA	0.02	15.60	NA	9.80
Safe level [21]			5.00	1.50	30	48	2.50	60

The mean total level in poultry farm soil (μg/g) of As, Cd, Cu, Fe, Pb and Zn in both seasons were observed in the range of NA-956.78, NA-2.87, 10.70-119.80, 38.72-1146.92, NA-103.18 and 10.42-314.10, respectively (Table 1). The permissible level of As, Cd, Cu, Fe, Pb and Zn in the soil is between 20-40, 1-3, 50-140, 400-5000, 50-300 and 150-300, μg/g, respectively [21]. The values of all the metals determined apart from As were considerably below the tolerable limits. Prevailing man-made practices that have increased contents of As in the poultry farm soil are uses of As containing pesticides and poultry animal manure (for example Roxarsone (3-nitro-4-hydroxyphenylarsonic acid) in poultry drop and as the wood additive chromated copper arsenate (CCA). Arsenic was extensively used as a pesticide in the form of lead arsenate Ca_3AsO_4, Paris-Green (copper acetoarsenite), H_3AsO_4, MSMA (monosodium methanearsonate), DSMA (disodium methanearsonate), sodium arsenite, organic arsenical herbicides and cacodylic acid [25]. These commodities were employed to benothing insect and plant pests in the soil and its comprehensive application has resulted into localized soil As contents of 10 - 892 μg/g [36]. The utilization of As in the cause of Roxarsone in poultry feed to check parasites and provoke the efficiency of eggs in poultry has increased total As value by 14-76 µg/g in poultry drop [36]. The use of this waste onto soil can thereby elevate the As contents in the soil which is a routine practice in the area under studied. Most likely the most general but ignored origin of As in the soil is from the widespread utilization of CCA preserved lumber, which can largely increase As contents in soils next to where it is applied, where the treatment process occurred, or where inappropriately discarded. To meet dietary prerequisites, different micro-minerals are involved in feeds and preservatives for poultry beyond the bird's require. Use of these chemical in deliberately adds potentially toxic metals to the soil.

Heavy metals content revealed difference between two different vegetables harvested from Agboola, Worgor, Odunola and Control sites (Table 2). The differences in heavy metal contents in vegetables of the similar location may be attributed to the variations in their floristic and morphology for heavy metal absorption, complicity, cumulation and retaining [35]. Also content of all the elements evaluated differs from one site to the other. Vegetables varied in their capacity to amass and concentrate metals in their eatable portions, variations between them were significant which was well corroborated from the studies conducted by Anita *et al*. [22]. The differences in heavy metal contents in vegetables were as a result of differences in their uptake and cumulation trend. The absorption and bioavailability of heavy metals in vegetables is controlled by several factors like clime, atmospheric fallouts, the contents of heavy metals in soils, the attribute of soil and the maturity levels of the plants at the period of the collection [26]. The range and mean content of trace metals (µg/g) in leafy vegetables was presented in Table 2, respectively. In vegetables (*Talinum triangulare and Vernonia amygdalina*) the content of trace elements (µg/g) varied between NA-38.50 for As, NA-0.13 for Cd, 0.02-0.82 for Cu, 4.40-250.05 for Fe, NA-0.28 for Pb and 8.20-94.94 for Zn for the both seasons. The overall mean Fe concentration of *Talinum triangulare* was between (98.33 and 76.34 µg/g) while for *Vernonia amygdalina* was between (125.51 and 92.18 µg/g) for dry and wet seasons which were in good harmony with concentrations (111-378 µg/g) recorded in vegetables by Leblebici and Musa, [27]. The maximum uptake of Fe was in *Vernonia amygdalina* (250.05 µg/g) followed by *Talinum triangulare* (203.20 µg/g), whereas the values of Fe in all the vegetables were higher than the recommended safe level of FAO/WHO.

The highest mean content of Zn was found in *Talinum triangulare* (94.94 µg/g) followed by *Vernonia amygdalina* (30.54 µg/g). The overall mean concentration of Zn in *Talinum triangulare* and *Vernonia amygdalina* (45.97 and 37.49; 16.47 and 12.65 µg/g) in vegetables of some poultry farms in Osun State was very similar to the vegetables from Beijing, China (32.01-69.26 µg/g) [28], as also from Rajasthan, India (21.1-46.4 µg/g) [27], but significantly below the Zn contents (3.00-171.03 µg/g) in vegetables from Titagarh Waste Bengal, India [35], Harare, Zimbabwe (1,038-1,872 µg/g) [34] and also the vegetables of Varanasi (59.61-79.46 µg/g) [22] and Delhi, India (46.7-91.9 µg/g) [28]. The maximum accumulation of Cd was found in *Talinum triangulare* (0.13 µg/g) while in *Vernonia amygdalina* (0.06 µg/g) which was lower than the FAO/WHO limit. The present work showed that the overall mean Cd level (0.11 and 0.09; 0.05 and 0.05 µg/g) evaluated in vegetables from some poultry farms of Osun metropolis was below the vegetables from Titagarh West Bengal, India (10.37-17.79 µg/g) [35] and vegetables from Turkey (25 µg/g) [27]. More so, our data was very far from the results of Anita *et al*. [22] (0.5-4.36 µg/g) in vegetables from Varanasi, India [27].

The overall mean Cu content in *Talinum triangulare* and *Vernonia amygdalina* (0.34 and 0.27; 0.36 and 0.22 µg/g) was below to the data recorded in Titagarh west Bengal, India (15.66-34.49 µg/g) [3] and also below the Cu concentration in vegetables (61.20 µg/g) from Zhengzhou city, China [28]. Notwithstanding, the difference of Cu content in vegetable in this study was strongly substantiated by the result (5.21-18.2 µg/g) of Leblebici and Musa, [27] and was also in good harmony with the concentrations recorded in Varanasi, India (10.95-28.58 µg/g) by Anita *et al.* [22]. Higher Cu content (0.82 µg/g) was detected in *Vernonia amygdalina* whilst the overall mean value was (0.34 and 0.27; 0.36 and 0.22 µg/g) for dry and wet seasons which were below the mean content (32.74 µg/g and 36.41 µg/g) respectively, described by Anita *et al*. [22] in Varanasi India of the same vegetables. Additionally, Cu contents in vegetables showed good harmony with the main contents in leafy vegetables (15.5-8.51 µg/g) from Samata Village, Jessor, Bangladesh obtained by Javid *et al*. [29]. The content of Cu value obtained in vegetables from this work shown the low absorption of the heavy metals in plants cultivated in poultry farm areas of Osun which was below those of above authors.

The maximum content of Pb was demonstrated by *Talinum triangulare* (0.28 µg/g) while in *Vernonia amygdalina* (0.15 µg/g) which was lower than the acceptable tolerance limit of WHO for Pb by more than seven times, respectively. Pb contents in eatable parts of all the vegetables studied in this research were lower than the permissible limits prescribed by WHO/FAO, India [29]. The overall mean Pb content in *Talinum triangulare* and *Vernonia amygdalina* (0.19 and 0.10; 0.11 and 0.06 µg/g) was below the concentrations recorded in Titagarh, West Bengal, (21.59-57.63 µg/g) [3] and also relatively below the Pb value recorded in China (0.18-7.75 µg/g) [28], (1.97-3.81 µg/g) [28] and in Varansi, India (3.09-15.74 µg/g) [22]. Nevertheless, it was also substantially below the mean content of Pb (409 µg/g) described in vegetables from Turkey by Turkdogan *et al*. [30]. Higher content of As demonstrated by *Talinum triangulare* (38.50 µg/g) was this seven-fold above the prescribed safe level of PFA (Prevention of food adulteration) [29]. The overall mean As contents in *Talinum triangulare* and *Vernonia amygdalina* was between (32.33 and 27.12; 11.97 and 9.88 µg/g) which was below the findings recorded by vegetables in Titagarh, West Bengal, India by Gupta *et al*. [35]. Notwithstanding, it was higher to the results of Anita *et al*. [22] (1.81-7.57 µg/g) in Varanasi, India; Leblebici and Musa, [27] (8.78-21.5 µg/g) in Delhi, India.

Mild fold higher contents of some of the heavy metals were revealed in all the vegetables. The dumping of poultry wastes on agricultural soil often throughout the year and not having a proper management blueprint may intensify the absorption and buildup of the heavy metals in the plants. This is in harmony with reports of mild contents of heavy metals in vegetables from poultry farm areas as in comparison to the cultivated plot of poultry area treated with poultry manure litters.

The data in Table 2 revealed that the increasing order of content of heavy metals in the vegetables was of the order Fe > Zn > As > Cu > Pb > Cd. Related findings were recorded by Abou Audu *et al.* [31] who reviewed the accumulation of heavy metals (Fe, Zn, Pb and Cd) on crops in Gaza Strip. The similar findings were also recognized by Do'a [37] who expressed that the maximum content was Zn, followed by Cu, Cr, Ni, Pb and Cd for two crops (*Cyperusmalaccensis* and *Scrpustripueter*). The findings signified that the metal content in the vegetables followed the trend *Talinum triangulare*>*Vernonia amygdalina*. Therefore, *Talinum triangulare* showed the strongest affinity to build up these metals from soils.

The overall pattern was that the metal contents were high in the soil but proportionally low in the plants. In the soil, the overall likelihood was As > Fe > Zn > Pb > Cu > Cd and in the plant, the pattern was Fe > Zn > As > Cu > Pb > Cd. The variation in the heavy metal contents in plants and soil was most likely as a result of differences in the sources of the metal content, accretion and retaining potential of soil and plant species. The total metal content of plants in this estimation was an input of the entire plants portion. Among all the monitored metal, the content of As and Fe in plant (9.88-32.33 µg/g and 4.40-250.05 µg/g) are above prescribed value [3].

Table 3 shows the translocation factor (TF) of different heavy metals from soil to vegetables estimated as the ratio between the contents of heavy metals in vegetables and their mean content in soil. Translocation factor is the proportion of the content of heavy metal in a plant to the content of heavy metal in soil. It implies the proportion of heavy metals in the soil that finished up in the vegetable crop [4]. Translocation factor was determined to obtain a thorough knowledge of the extent of risk and related hazard as a result of ingestion emanating from heavy metal accretion in eatable part of vegetables [18]. Translocation factor of heavy metals controlled by bioavailability of metals, which in proper sequence regulated by its content in the soil, their speciations, variation in uptake potential and rate of growth of varied plant species [21]. Greater contents of TF imply poor retention of metals in soil and/or more translocation into plants. The higher uptake of heavy metals in leafy vegetables might be because of higher transpiration rate to support the growth and moisture content of these plants [20]. From Table 3 it can be observed that Zn contained the highest TF value (1.1132) and the patterns of heavy metals TF followed the sequence Zn > Fe >Cd > As > Cu > Pb and Zn > Fe > Cd > As > Cu > Pb in dry and wet seasons, respectively. In *Talinum triangulare*, the TF was Fe > Zn > Cd > As > Cu > Pb, and while *Vernonia amygdalina* was Fe > Zn > As > Cu > Cd > Pb. The results showed that Zn had the highest TF in all the vegetables in both seasons. Related results were recorded by Naser *et al.* [33] where they detected that Zn had the highest TF among other metals and the order was Zn, Fe, Cd, Ni, Co and Pb, they also expressed that the high mobility of Zn with a natural presence in the soil and the low retaining of Zn in the soil than other toxic cations may increase the TF of Zn.

In a study carried out by Opaluwa *et al.* [32], the highest TF of metals was for Cu and the occurrence was Cu, Co, Fe, As, Zn, Ni and Pb. The food-chain crops might take up sufficient quantities of heavy metals to become a potential health hazard to human [30], that implies that Zn, Fe, Cd, As, Cu and Pb present no serious risk of metal studied due to the low TF.

Table 3. Mean Translocation Factor Content in Vegetables Grown in Poultry Farm Sites of Area under Study (Dry and Wet Seasons)

Site	Scientific Name	Common Name	As	Cd	Cu	Fe	Pb	Zn
				Dry Season				
Agboola	*Talinum triangulare*	Water leaf	2.98×10^{-2}	4.89×10^{-2}	6.00×10^{-3}	2.09×10^{-1}	1.70×10^{-3}	2.82×10^{-1}
	Vernonia amygdalina	Bitter leaf	1.16×10^{-2}	2.17×10^{-2}	6.50×10^{-3}	2.96×10^{-1}	1.30×10^{-3}	1.30×10^{-1}
Control			NA	NA	1.40×10^{-3}	7.29×10^{-2}	NA	5.69×10^{-1}
Worgor	*Talinum triangulare*	Water leaf	5.86×10^{-2}	5.95×10^{-2}	7.50×10^{-3}	2.68×10^{-1}	4.50×10^{-3}	4.10×10^{-1}
	Vernonia amygdalina	Bitter leaf	1.95×10^{-2}	3.57×10^{-2}	7.70×10^{-3}	3.32×10^{-1}	1.30×10^{-3}	8.79×10^{-2}
Control			NA	NA	3.70×10^{-3}	1.76×10^{-1}	NA	1.1132
Odunola	*Talinum triangulare*	Water leaf	7.45×10^{-2}	9.29×10^{-2}	1.08×10^{-2}	2.96×10^{-1}	1.80×10^{-3}	3.14×10^{-1}
	Vernonia amygdalina	Bitter leaf	2.89×10^{-2}	3.57×10^{-2}	1.16×10^{-2}	3.64×10^{-1}	1.70×10^{-3}	1.15×10^{-1}
Control			NA	NA	3.00×10^{-3}	3.82×10^{-1}	NA	5.06×10^{-1}
				Wet Season				
Agboola	*Talinum triangulare*	Water leaf	2.41×10^{-2}	2.09×10^{-2}	5.70×10^{-3}	1.22×10^{-1}	8.00×10^{-4}	1.86×10^{-1}
	Vernonia amygdalina	Bitter leaf	9.30×10^{-3}	1.74×10^{-2}	3.60×10^{-3}	1.33×10^{-1}	3.00×10^{-4}	5.94×10^{-2}
Control			NA	NA	1.30×10^{-3}	9.43×10^{-2}	NA	4.29×10^{-1}
Worgor	*Talinum triangulare*	Water leaf	4.01×10^{-2}	4.53×10^{-2}	6.40×10^{-3}	1.48×10^{-1}	7.00×10^{-4}	2.15×10^{-1}
	Vernonia amygdalina	Bitter leaf	1.03×10^{-2}	1.65×10^{-2}	4.40×10^{-3}	1.72×10^{-1}	6.00×10^{-4}	8.18×10^{-2}
Control			NA	NA	1.70×10^{-3}	1.62×10^{-1}	NA	6.24×10^{-1}
Odunola	*Talinum triangulare*	Water leaf	3.68×10^{-2}	4.50×10^{-2}	3.70×10^{-3}	1.44×10^{-1}	7.00×10^{-4}	2.33×10^{-1}
	Vernonia amygdalina	Bitter leaf	1.67×10^{-2}	2.70×10^{-2}	4.30×10^{-3}	2.03×10^{-1}	8.00×10^{-4}	8.44×10^{-2}
Control			NA	NA	1.20×10^{-3}	3.45×10^{-1}	NA	4.29×10^{-1}

In overall mean for the both seasons, the trend in the TF for heavy metals in the study sites was in the ranking occurrence of Zn > Fe > Cd > As > Cu > Pb. Among the heavy metals, TF values were found to be mild for Zn, Fe, Cd and As whilst comparatively lower TF values were found in Cu and Pb. The food chain (soil-plant-human) is mostly detected as one of the main routes for exposure of human to soil contaminants. Soil-to-plant transfer is one of the key processes of human exposure to toxic heavy metals via the food chain [30]. When TF ≤ 1 or TF = 1, it signifies that the plant merely takes up the heavy metal but does not amass and when TF > 1, this signifies that plant amasses the heavy metals. Overall mean TF contents of As, Cd, Cu, Fe; Pb and Zn in dry and wet seasons were less than one (<1) in the vegetables which signify that plants merely take in the heavy metals. Different sorts of plants can take in and tolerate metals diversely. Generally, there is little evidence of vegetable contamination via poultry activity. The application of poultry manures may, nevertheless, elevate the metal concentration of unpolluted soils. This may present a threat to animals or children in the area who might ingest the poultry soil directly. The differences in heavy metal contents in vegetables were as a result of differences in their uptake and cumulation trend.

Chabukdhara *et al.* [4] also recorded greatest translocation factor for heavy metals of some leafy vegetables. In spite of the fact that TF does not precisely indicate the hazard aligned with the metal in any form, the use of poultry manures may, notwithstanding, elevate the metal concentration of uncontaminated soils and as a result, metal absorption by plants increasing on that soils. The degree of toxicity of heavy metals to human beings determined by their daily intake [20,22]. Data on DIM shown that Fe contained highest vegetable metal content (250.05 µg/g) and the highest DIM (1.42 × 10^{-1}) value, whilst its HRI was second to the highest. The patterns of DIM was found to be in the sequence Fe > Zn >As > Cu > Pb > Cd (Table 5). From another point of view, HRI results revealed that the sequence of HRI in *Talinum triangulare* was As > Fe > Zn >Cd > Pb > Cu while in *Vernonia amygdalina* it was As > Fe > Cd > Zn > Pb > Cu (Table 6). The continual intakes of *Vernonia amygdalina* and *Talinum triangulare* plants in this soil seemed to be at greater risk of As pollution as its values were somewhat high (NA-72.67 and NA-28.27 for *Talinum triangulare* and *Vernonia amygdalina*), respectively.

Hazard quotient results revealed the hazard inherent in subject to the metals for the period of lifespan regarded in this research. The HQ values revealed that the residents were vulnerable to health hazards related to these metals in the sequence As > Fe > Cd > Zn > Pb > Cu (*Vernonia amygdalina*) and As > Fe > Zn > Cd > Pb > Cu (*Talinum triangulare).*The THQ for Cd, Cu, Pb and Zn were far below 1 inferring that they did not present direct health effect. Nonetheless, the THQ values for As and Fe were so high that the occupants were dreaded to be vulnerable to health hazard in relation to As and Fe to a greater extents. The input for As apart from others was near 99% to the sum total HQ (Table 4). This revealed that in *Vernonia amygdalina and Talinum triangulare*, As content was above prescribed value, thereby these vegetables were not fit for intake. Arsenic is poisonous elements that can be toxic to plants, despite plants generally demonstrate potential to accumulate considerable quantities of As with no apparent modifications in their look or form. In countless plants, As accretion can transcend many hundred times the threshold of highest value tolerable for human [23]. The entry of As into the food chain may alter human health and as such, assessments regarding As cumulation in vegetables present matter of serious concern [4]. By and large, *Vernonia amygdalina and Talinum triangulare* that were considered in this assessment were contaminated by As and they were therefore harmful to the eaters. *Talinum triangulare* demonstrated severely large accumulation trend in relations to As, Fe, Cu, Zn and Pb. In this way, it could be meant for the extraction of the heavy metals from the polluted soil, sediment, sewages and other solid waste. From another point of view, their massive intake could be detrimental because they contain high bioaccumulative content values for almost all of the heavy metals.

Table 4. Mean Hazard Quotient Content in Vegetables Grown in Poultry Farm Sites of Area under Study (Dry and Wet Seasons)

Site	Scientific Name	Common Name	As	Cd	Cu	Fe	Pb	Zn
				Dry Season				
Agboola	*Talinum triangulare*	Water leaf	566.67	6.00×10^{-1}	9.33×10^{-1}	3.68×10^{+1}	2.17×10^{-1}	1.52×10^{+1}
	Vernonia amygdalina	Bitter leaf	221.33	2.67×10^{-1}	1.00×10^{-1}	5.23×10^{+1}	1.67×10^{-1}	4.12×10^{-1}
Control			NA	NA	3.33×10^{-1}	9.78×10^{-1}	NA	1.82×10^{-1}
Worgor	*Talinum triangulare*	Water leaf	733.33	6.67×10^{-1}	1.03×10^{-1}	4.30×10^{+1}	4.67×10^{-1}	2.11×10^{+1}
	Vernonia amygdalina	Bitter leaf	244.00	4.00×10^{-1}	1.07×10^{-1}	5.33×10^{+1}	1.33×10^{-1}	4.56×10^{-1}
Control			NA	NA	6.67×10^{-2}	1.91×10^{-1}	NA	2.58×10^{-1}
Odunola	*Talinum triangulare*	Water leaf	855.56	8.67×10^{-1}	1.27×10^{-1}	4.52×10^{+1}	2.67×10^{-1}	1.85×10^{+1}
	Vernonia amygdalina	Bitter leaf	332.44	3.33×10^{-1}	1.37×10^{-1}	5.56×10^{+1}	2.50×10^{-1}	6.79×10^{-1}
Control			NA	NA	6.67×10^{-2}	3.29×10^{-1}	NA	2.09×10^{-1}
				Wet Season				
Agboola	*Talinum triangulare*	Water leaf	513.33	4.00×10^{-1}	9.00×10^{-2}	3.10×10^{+1}	1.33×10^{-1}	1.29×10^{+1}
	Vernonia amygdalina	Bitter leaf	197.78	3.33×10^{-1}	5.67×10^{-2}	3.38×10^{+1}	5.00×10^{-2}	4.13×10^{-1}
Control			NA	NA	3.33×10^{-3}	1.69×10^{-1}	NA	1.96×10^{-1}
Worgor	*Talinum triangulare*	Water leaf	670.00	7.33×10^{-1}	9.67×10^{-2}	3.12×10^{+1}	1.17×10^{-1}	1.43×10^{+1}
	Vernonia amygdalina	Bitter leaf	172.78	2.67×10^{-1}	6.67×10^{-2}	3.63×10^{+1}	1.00×10^{-1}	5.45×10^{-1}
Control			NA	NA	3.33×10^{-3}	2.27×10^{-1}	NA	2.36×10^{-1}
Odunola	*Talinum triangulare*	Water leaf	624.67	6.67×10^{-1}	7.33×10^{-2}	3.22×10^{+1}	1.50×10^{-1}	1.63×10^{+1}
	Vernonia amygdalina	Bitter leaf	283.79	4.00×10^{-1}	8.67×10^{-2}	4.54×10^{+1}	1.33×10^{-1}	5.89×10^{-1}
Control			NA	NA	3.33×10^{-3}	3.47×10^{-1}	NA	2.18×10^{-1}

Table 5. Mean Daily Intake of Heavy Metals Content in Vegetables Grown in Poultry Farm Sites of Area under Study (Dry and Wet Seasons)

Site	Scientific Name	Common Name	As	Cd	Cu	Fe	Pb	Zn
				Dry Season				
Agboola	*Talinum triangulare*	Water leaf	1.45×10^{-2}	5.10×10^{-5}	3.17×10^{-4}	9.38×10^{-2}	7.37×10^{-5}	3.88×10^{-2}
	Vernonia amygdalina	Bitter leaf	5.64×10^{-3}	2.27×10^{-5}	3.40×10^{-4}	1.33×10^{-1}	5.67×10^{-5}	1.05×10^{-2}
Control			NA	NA	1.13×10^{-5}	2.49×10^{-3}	NA	4.65×10^{-3}
Worgor	*Talinum triangulare*	Water leaf	1.87×10^{-2}	5.67×10^{-5}	3.51×10^{-4}	1.10×10^{-1}	1.59×10^{-4}	5.38×10^{-2}
	Vernonia amygdalina	Bitter leaf	6.22×10^{-3}	3.40×10^{-5}	3.63×10^{-4}	1.36×10^{-1}	4.53×10^{-5}	1.16×10^{-2}
Control			NA	NA	2.27×10^{-5}	4.87×10^{-3}	NA	6.57×10^{-3}
Odunola	*Talinum triangulare*	Water leaf	2.18×10^{-2}	7.37×10^{-5}	4.31×10^{-4}	1.15×10^{-1}	9.07×10^{-5}	4.71×10^{-2}
	Vernonia amygdalina	Bitter leaf	8.48×10^{-3}	2.83×10^{-5}	4.65×10^{-4}	1.42×10^{-1}	8.50×10^{-5}	1.73×10^{-2}
Control			NA	NA	2.27×10^{-5}	8.39×10^{-3}	NA	5.33×10^{-3}
				Wet Season				
Agboola	*Talinum triangulare*	Water leaf	1.31×10^{-2}	3.40×10^{-5}	3.06×10^{-4}	7.90×10^{-2}	4.53×10^{-5}	3.30×10^{-2}
	Vernonia amygdalina	Bitter leaf	5.04×10^{-3}	2.83×10^{-5}	1.93×10^{-4}	8.63×10^{-2}	1.70×10^{-5}	1.05×10^{-2}
Control			NA	NA	1.13×10^{-5}	4.31×10^{-3}	NA	4.99×10^{-3}
Worgor	*Talinum triangulare*	Water leaf	1.71×10^{-2}	6.23×10^{-5}	3.29×10^{-4}	7.95×10^{-2}	3.97×10^{-5}	3.65×10^{-2}
	Vernonia amygdalina	Bitter leaf	4.41×10^{-3}	2.27×10^{-5}	2.27×10^{-4}	9.24×10^{-2}	3.40×10^{-5}	1.39×10^{-2}
Control			NA	NA	1.13×10^{-5}	5.78×10^{-3}	NA	6.01×10^{-3}
Odunola	*Talinum triangulare*	Water leaf	1.59×10^{-2}	5.67×10^{-5}	2.49×10^{-4}	8.21×10^{-2}	5.10×10^{-5}	4.15×10^{-2}
	Vernonia amygdalina	Bitter leaf	7.24×10^{-3}	3.40×10^{-5}	2.95×10^{-4}	1.16×10^{-1}	4.53×10^{-5}	1.50×10^{-2}
Control			NA	NA	1.13×10^{-5}	8.84×10^{-3}	NA	5.55×10^{-3}

Table 6. Mean Health Risk Index of Heavy Metals Content in Vegetables Grown in Poultry Farm Sites of Area under Study (Dry and Wet Seasons)

Site	Scientific Name	Common Name	As	Cd	Cu	Fe	Pb	Zn
				Dry Season				
Agboola	*Talinum triangulare*	Water leaf	48.17	5.10×10^{-2}	7.90×10^{-3}	3.10×10^{-1}	1.84×10^{-2}	1.29×10^{-1}
	Vernonia amygdalina	Bitter leaf	18.81	2.30×10^{-2}	8.50×10^{-3}	4.40×10^{-1}	1.42×10^{-2}	3.50×10^{-2}
Control			NA	NA	2.80×10^{-4}	8.30×10^{-3}	NA	1.55×10^{-2}
Worgor	*Talinum triangulare*	Water leaf	6.23	5.70×10^{-2}	8.80×10^{-3}	3.70×10^{-1}	3.98×10^{-2}	1.79×10^{-1}
	Vernonia amygdalina	Bitter leaf	20.73	3.40×10^{-2}	9.10×10^{-3}	4.50×10^{-1}	1.13×10^{-2}	3.87×10^{-2}
Control			NA	NA	5.70×10^{-4}	1.60×10^{-2}	NA	2.19×10^{-2}
Odunola	*Talinum triangulare*	Water leaf	72.67	7.40×10^{-2}	1.10×10^{-2}	3.80×10^{-1}	2.27×10^{-2}	1.57×10^{-1}
	Vernonia amygdalina	Bitter leaf	28.27	2.80×10^{-2}	1.20×10^{-2}	4.70×10^{-1}	2.13×10^{-2}	5.77×10^{-2}
Control			NA	NA	2.27×10^{-5}	8.39×10^{-3}	NA	1.78×10^{-2}
				Wet Season				
Agboola	*Talinum triangulare*	Water leaf	43.67	3.40×10^{-2}	7.70×10^{-3}	2.63×10^{-1}	1.13×10^{-2}	1.10×10^{-1}
	Vernonia amygdalina	Bitter leaf	16.80	2.80×10^{-2}	4.80×10^{-3}	2.88×10^{-1}	4.30×10^{-3}	3.50×10^{-2}
Control			NA	NA	2.80×10^{-4}	1.44×10^{-2}	NA	1.66×10^{-2}
Worgor	*Talinum triangulare*	Water leaf	57.00	6.20×10^{-2}	8.20×10^{-3}	2.65×10^{-1}	9.90×10^{-3}	1.22×10^{-1}
	Vernonia amygdalina	Bitter leaf	14.7	2.30×10^{-2}	5.70×10^{-3}	3.08×10^{-1}	8.50×10^{-3}	4.63×10^{-2}
Control			NA	NA	2.80×10^{-4}	1.93×10^{-2}	NA	2.00×10^{-2}
Odunola	*Talinum triangulare*	Water leaf	53.00	5.70×10^{-2}	6.20×10^{-3}	2.74×10^{-1}	1.28×10^{-2}	1.38×10^{-1}
	Vernonia amygdalina	Bitter leaf	24.13	3.40×10^{-2}	7.40×10^{-3}	3.87×10^{-1}	1.13×10^{-2}	5.00×10^{-2}
Control			NA	NA	2.80×10^{-4}	2.95×10^{-2}	NA	1.85×10^{-2}

4. Conclusion

The manuscript assessed the heavy metals composition of the vegetables planted in the poultry farm region of Osun State, South-Western Nigeria. The levels of metals discovered in both the soil and the vegetables indicated obvious contamination of the samples by heavy metals as shown by the various hazard assessments like DIM, THQ and HRI.

This assessment revealed that long-term and indiscriminate use of untreated poultry manure directly to agricultural field may result in accumulation of harmful metals in surface and sub-surface soils. The metals contents in the leafy vegetables were greater than the prescribed values in some instances. It was implied that additional monitoring and assessment of bioaccumulation of these metals should be carried out for other vegetables planted on the soils to determine their heavy metal bioaccumulation potential and safety for human consumption.

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Statement of Competing Interests

The authors declare that they have no competing interests.

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