

Health Risk Assessment of Heavy Metals Via Consumption of Cassava, Cultivated on Reclaimed Mining Land Sites in Prestea-Huni Valley District, Ghana

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Abstract The soil determines the quality of food and hence the health of the individuals. However, most lands in the District are under mining or concessions to the detriment of livelihood. This situation resulted in the cultivation of food crops on reclaimed mining land sites contrary to the accepted norm. To assess the human health risk and the quality of cassava cultivated on reclaimed mining land sites in Prestea-Huni Valley District, soil and cassava samples were taken randomly in separate polyethylene bags in triplicates from four farms located on reclaimed mining land sites within the Prestea-Huni Valley District. Atomic Absorption Spectrophotometric (AAS) technique was employed in the determination of the metals after digestion. The precision and accuracy of the analytical methods were evaluated by analysing IAEA-soil-7 and 1547-Peach leaves, which are standard reference materials with recoveries ranging from 87% to 98%. The trend in metal tolerance for both the cassava tuber and its peel were similar and in the order Cd > Pb > Cr > Zn > As. Both soil Pollution Load Index (PLI) and Total Hazard Quotient (THQ) were below their respective values. Using the health index of $0.8017 < 1.0$ ($\sum THQ$) as a criterion for human health assessment, the cassava cultivated on reclaimed mining land sites in the Prestea-Huni Valley District, are safe and of high quality, since the health index was below the threshold level of 1.0, a level below which the cassava is deemed fit for human consumption.

Keywords Prestea-Huni Valley, Cassava, Peel, Heavy metals, Health index, Bioaccumulation

1. Introduction

The increasing demand for food and food safety is a major public concern and has drawn the attention of both governments and scientists to the risks associated with the consumption of contaminated food (i.e pesticides, heavy metals, and toxins in food) [1].

Cassava (*Manihot esculenta* Crantz) belongs to the family Euphorbiaceae. It is cultivated as a monocrop or intercropped with other food crops, either as a dominant or subsidiary crop. In terms of the quantity produced, cassava is the most important root crop followed by yam and cocoyam, but ranked second to maize in terms of area planted [2]. It is an important starchy staple crop in Ghana with per capita consumption of 154.0 Kg/ year [2,3]. Besides being a staple food crop, cassava can be processed into fermented and unfermented products. Fermented products include cassava bread, fermented cassava flour, fermented starch, *fufu*, *akyeke*, *agbelima*, whereas unfermented products include

tapioca, cassava chips, gari, and pellets. Fermentation is one of the oldest biotechnological approaches to solving food processing and preservation as well as beverages production [4]. Processing the root tuber into various products increases the shelf life and makes transportation to urban markets less expensive [5]. It is also a raw material for industrial starch and ethanol. The leaves of cassava are also edible, while the peel is either eaten by animals or used in the formulation of animal feed.

Cassava peel is the epidermal layer of cassava root tuber and as such after harvesting and processing, constitutes 25% of the whole plant. Cassava peels obtained from the products of cassava are usually either discarded as waste in the environment or used as animal feeds if properly processed. The large volume of processed cassava is proportional to the volume of cassava peels wasted without proper use [4].

The history of mining in Africa, in particular, and the third world in general, is a history of land appropriation, displacement of people from their lands, environmental devastation, and further marginalization and oppression of people belonging to the lower economic sectors of the society [6]. In Ghana, the mining sector, especially the mining of gold for export, has received renewed emphasis with new mining codes, clearly defining the rights of the

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foreign investor [6].

In the Western Region of Ghana alone, it is estimated that over 70% of the land previously used for farming activities is under mine concessions [2] to the detriment of livelihood. As a result, farmlands were lost to mining activities and mine concessions, forcing farmers to cultivate food crops on reclaimed mining land sites for their livelihood.

Reclamation is widely used to refer to the revegetation of degraded mining land sites. It aims to recover the productivity of a degraded site mostly using exotic tree species. The success criteria are the restoration of the disturbed site to acceptable physical appearance and acceptable after-use or the end-use objectives [7].

The issue of environmental contamination concerning heavy metals is the focus of research globally, especially in third world countries, as a result of the exploitation of mineral resources. Heavy metals are found in extremely small quantities in animals and plant tissues by consumption of food and water.

Naturally, heavy metals may accumulate in the environment through the processes of weathering and dissolution. However, heavy metals may also be introduced to the environment (soil and water) by humans during mining, agricultural, and industrial activities [8].

Heavy metals may enter the food chain as a result of their uptake by the edible parts of the plant, thus, determination of heavy metals in food crops especially those cultivated on reclaimed mining land sites is very important. Heavy metals from soil enter plants primarily through the root system. Plant roots are the most important sites for the uptake of toxic substances from the soil [8,9]. At varying levels, dependent on species, plants can extract or remediate toxins from the soil through their root system [9]. Once the heavy metal is present in the soil and water, these metals are likely to be passed on to humans through the food chain by the process of bioaccumulation.

Information on heavy metal uptake and concentration in edible tissues of food crops and their dietary intake are therefore very important for assessing human health risks [10]. Absorption of heavy metals in low doses by humans over a long period through food has been shown to have resulted in serious health consequences such as declining economic development in terms of low productivity as well as the direct cost of treating illnesses. Research has shown that high levels of Pb, Cd, Co, Cu, Ni, and Zn have cumulative effects since there is no homeostatic mechanism that regulates their toxicity. Some common health implications of heavy metals in humans are kidney diseases, damage to the nervous system, diminished intellectual capacity, heart diseases, bone fracture, and cancer [8]. Heavy metals in soil reduce the yield of vegetables by disrupting the metabolic processes of plants. Generally, the levels of heavy metals in soil are higher than the levels observed in food crops. This implies that only a small portion of soil heavy metals is transferred to plants (food crops) and the root acts as a barrier to the translocation of heavy metals within the plant [1]. It is also noticed that the application of phosphate

fertilizers to food crops is the main source of soil heavy metal pollution. This is because of the presence of Cd, As, Pb, Cr, Hg, and Ni as impurities in phosphate rocks [1]. The Soil may also become contaminated through mining and related anthropogenic activities [11].

The soil-to-plant transfer quotient which is termed bioaccumulation factor (BAF) is the main source of human exposure. The relative difference in bioavailability of metals to plants is the transfer quotient, which is a factor in determining metal tolerance by food crops. The transfer quotient or bioaccumulation factor threshold value of 1.0 is the benchmark above which the plant is said to be tolerant to a specified metal and indicates a high probability of food crop contamination by anthropogenic activities [1]. Information on heavy metal concentrations in edible tissues of food crops and their dietary intake is therefore very important for assessing human health risks [10]. Total Hazard Quotient (THQ) is the adverse health effect that a given metal is likely to produce if present in food and is the determining factor in Health Index (HI). A total hazard quotient value of 1.0 is the threshold value below which the food is safe for human consumption [5]. The sum of THQs is equal to HI.

This study was therefore set out to find out the quality of food crop (cassava), a staple food, cultivated on reclaimed mining lands sites, and its associated health risks to residents in the study area.

2. Materials and Methods

2.1. Study Area

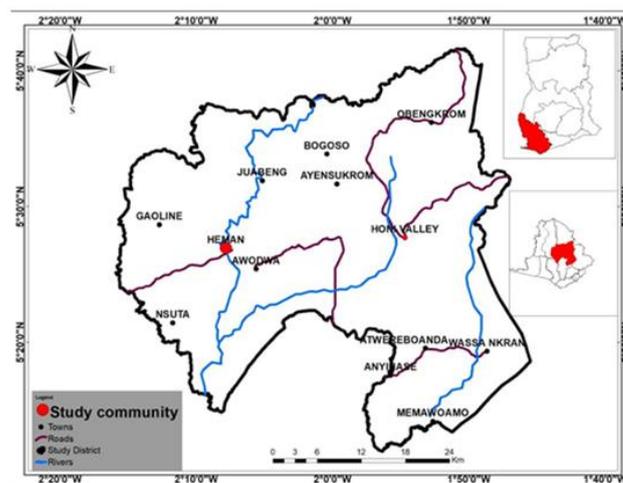


Figure 1. Map of Prestea-Huni Valley District Showing the sampling points

Prestea-Huni Valley District is located between latitude $2^{\circ}06'$ and longitude $5^{\circ}47'$ (Figure 1). The District has a land area of approximately $1,809 \text{ km}^2$. The land rises from about 240 m to about 300 m above sea level with the topography generally undulating with few scarps ranging between 150 m and 300 m above sea level. The district falls within the forest dissected plateau physiographic region. Precambrian rocks of

Birimian underlie the forest dissected plateau. The Birimian rocks are regarded as the most important formations due to their mineral potentials and thus the existence of minerals deposits in the District [12].

Some several rivers and streams drain the District, notable among them are Ankobra, Huni, Oppon, Bogo, Peme, Subri, Bona, and Mansi. The District is located in the rainforest zone of Ghana and enjoys a wet equatorial climate. The District experiences high rainfall with a mean annual rainfall of 187.83mm. Temperatures are high all year round with significant daily and seasonal variations. The annual average temperatures range between 26°C and 30°C. Humidity varies from 75-80% in the wet season and 70-80 percent in the dry season [12].

The District's major forest reserve is the Bona reserve. The other two reserves are Ben West and Nkontoben reserves. Activities of illegal mining and other illegal logging are posing a threat to the natural vegetation. Cocoa, oil palm, coffee, rubber, coconut, and citrus are some of the major cash crops grown in the area [12].

2.2. Sampling

The cassava samples cultivated on reclaimed mining land sites in the study area were harvested in triplicates during three sampling campaign periods from four farms randomly, at each farm, the cassava was harvested at four locations and put together from which a representative sample was obtained and labelled. The corresponding soil samples were taken from the respective farms using the auger, to a depth of 30.0 cm to cover the plough zone. At each farm, the soil samples were taken from three different places, bulked together, from which a representative sample was taken from the bulk and labelled. Both samples were packed into separate polyethylene bags and transported to the laboratory.

2.3. Sample Preparation and Analysis

The cassava tubers were washed with water to remove any surface deposit of soil particles that might act as contaminants. The washed cassava was then peeled using a stainless knife into an already washed plastic container. Both the cassava tuber and the peel (waste) were collected separately and labelled, put into an oven to dry at 105°C to a constant weight. The dried tuber and the peel were ground separately using cleaned ceramic mortar and pestle into powder and sieved using <40 mesh, homogenized, and packed in polyethylene bags. The samples were stored in the fridge for further analysis. Similarly, the soil samples were disaggregated, dried in an oven at 105°C to a constant weight, and taken through the same treatment as the cassava tuber and the peels.

Triplicate 2.0 g of each powdered cassava tuber and the peel were weighed and digested separately using aqua regia (a mixture of HCl and HNO₃ in the ratio 1:3) on a water bath at 80°C until a transparent solution appeared depicting the end of the digestion [13].

Similarly, triplicate 2.0 g of the powdered soil samples were also taken and digested in a 100.0 mL polytetrafluoroethylene teflon bombs. About 10.0 mL of concentrated HNO₃ was added to each soil sample and allowed to stand for 10 minutes. About 30% H₂O₂ was also added drop by drop to the mixture until the mixture no longer effervesced on the addition of H₂O₂. To each mixture in the teflon bomb, 2.0 mL of concentrated H₂SO₄ were added, followed by the addition of 5.0 mL of concentrated HClO₄ successively to each mixture. The resulting mixtures were digested for 25 minutes in a Milestone microwave oven (Ethos 900) using the following operating parameters: 250W for 2.0 min, 0W for 2.0 min, 250W for 6min, 400W for 5.0 min, 650W for 5.0 min and 5.0 min for venting [14]. The rotor was put into a bowl of water to cool the content of the tubes and also to reduce the associated pressure. The digested soil samples were then filtered using whatman (No 1) filter paper into 100.0 mL volumetric flask and made up to the mark using de-ionized distilled water. Calibration standards were prepared for the various analytes together with their respective blanks were used for the determinations, using Varian Fast Sequential Atomic Absorption Spectrophotometer at the respective wavelengths. Acetylene gas was used as the carrier gas in the determination, while argon was passed through the system to remove interfering gases between each reaction time [13,15].

2.4. Quality Control/ Assurance

The chemicals used were of analytical grades obtained from Sigma Aldrich. The reproducibility and precision of the analytical procedures were tested by carrying out triplicate analysis on standard reference materials IAEA-soil-7 and Peach leaves [16,17]. Triplicate results did not differ by more than 5% of the mean. Recoveries from the reference materials ranged between (87-98)%.

2.5. Pollution Load Index (PLI)

Pollution Load Index is the degree of soil pollution in mined soil compared to a reference value. It was estimated using the pollution load index technique based on soil metal levels as shown in equation 1.

$$PLI = \frac{[C_{soil}]}{[C_{reference}]} \quad (1)$$

Where [C_(soil)] is the heavy metal concentration in soil and [C_{reference}] is the guideline values of metal levels in soil [16].

2.6. Bioaccumulation Factor (BAF)

The heavy metal transfer factor which is termed the Bioaccumulation factor of heavy metals in food crops was estimated using the plant bioaccumulation factor (BAF), which is the ratio of heavy metal concentration in food crop [C_{plant}] to that of contaminated soil [C_{soil}] as shown in equation 2 [16].

$$BAF = \frac{[C_{plant}]}{[C_{soil}]} \quad (2)$$

2.7. Estimation of Dietary Exposure and Health Risk

Estimated Average Daily Intakes (EADIs mg/g/day) of metals of interest were determined based on their average concentrations in cassava samples and a consumption rate of 154 Kg/person/day of cassava was assumed for Ghana [3]. For each type of exposure, the EADI was obtained by the equation 3.

$$EADI = \frac{[C] \times C_{factor} \times F_{ir}}{W \times D} \times 1000 \quad (3)$$

Where [C] is the concentration of the metals (mg/Kg) in cassava tuber, C_{factor} is the conversion factor from fresh weight to dry weight, F_{ir} is the mean annual intake of cassava in g / person/day. The cassava supply of 154 Kg/ person /year was divided by 365 days and the result was multiplied by 1000 for gramme conversion. The result is an intake of 421.92 g/person/day of cassava which is F_{ir} . W is the mean body weight (60 Kg) of an average adult and D is the number of days in a year (365 days) and 1000 is a unit conversion factor from Kg to gramme.

2.8. Reference Oral Dose (R_f Dose)

Reference oral dose (R_f dose) is an estimated exposure of metals to the human body per day that has no negative effect during a lifetime. The values of the oral dose were obtained from the literature [18].

2.9. Target Hazard Quotient (THQ)

The THQ is defined as the ratio of exposure (EADI) to toxic metals and the reference dose which is the highest level at which no adverse health effects are expected. The ratio is normally divided by 1000 as a unit conversion factor in equation 4

$$THQ = \left[\frac{EADI}{R_{fDose} \times 1000} \right] \quad (4)$$

Where EADI is the estimated average daily intake of cassava, R_{fDose} is the oral reference dose of the trace metals in mg/Kg/day.

2.10. Health Index (HI)

The health or hazard index (HI) is the summation of the

individual target hazard quotients of the metals assessed in cassava. The health index (HI) assumed that the consumption of a particular food type would result in simultaneous exposure to several potentially toxic metals. Even if individual THQs for the metals in the food item is lower than unity, the cumulative effects of consumption may result in adverse health effects. If HI is > 1, there is a potential adverse health effect. However, if the index is below the threshold level of 1.0, the food is considered safe for the consumer [18]. The equation for HI is given by equation 5 [19].

$$HI = \sum_{n=1}^i THQ_n \quad (5)$$

3. Results and Discussion

The reproducibility of the analytical method was assessed by analysing standard reference materials IAEA soil-7 [16] and 1547-Peach leaves [17]. Table 1 shows the recommended values for As, Cr, Pb, Zn, and Cd in soil-7 and 1547-Peach leaves against the experimental values obtained. The percentage recoveries ranged from 87% to 98%.

Mean As level in soil samples from the farms in the study area was 0.04 mg/Kg (Table 2). This level of As in soil was lower than the WHO/FAO, (2017) [20] permissible level of 5.0 mg/Kg of As in soil. The PLI of As in the soil was 0.008, as shown in Table 2 which was less than the threshold value of 1.0 above which the soil is deemed to have been polluted.

The arsenic level (mean) in cassava tuber and its peel from all the farms was 0.02 mg/Kg (Table 2). The bioaccumulation factor of As in cassava tuber and its peel was 0.01 (Table 3) This factor is less than 1.0 [22] a threshold above which the cassava tuber and its peel are said to be a potential accumulator of As. The calculated estimated daily intake of As in cassava tuber was 0.0119 and that of the total hazard quotient was 0.039 (Table 3). All these values were below the benchmark of 1.0, an indication of safe levels of As in cassava. Data on animal food ingestion (F_{ir}) and reference dose of As in peels were not available, hence, EADI and THQ for peel values were not calculated.

Table 1. Recovery results from the standard reference materials on dry weight basis

Analyte	IAEA- Soil-7			1547- Peach Leaves		
	Recommended value, mg/Kg	Experimental value, mg/Kg	%Recovery	Recommended value, µg/g	Experimental value, µg/g	% Recovery
As	13.4	12.8	95	0.06	0.056	93
Cd	1.3	1.25	96	0.026	0.025	96
Cr	60.0	58.6	98	1.0	0.88	88
Pb	60.0	56.2	93	0.87	0.85	97
Zn	104.0	102.89	98	17.9	17.5	97
Cu	11.0	9.61	87	3.7	3.5	94
Mn	631	623	98	98	96.40	98

Table 2. Mean metal levels (mg/Kg dry weight) in Soil, Cassava Tuber, and Peel with corresponding soil Pollution Indices (PLI)

Sample	As	Cd	Cr	Pb	Zn
Soil	0.03-0.04	0.03-0.06	0.06-0.38	0.03-0.04	1.86-1.90
Mean	0.04	0.048	0.285	0.04	1.885
Soil Pollution index (PLI)	0.008	0.048	0.0029	0.007	0.009
WHO/FAO, 2007 value in soil	5.0	1.0	100.0	60.0	200.0
Cassava tuber	0.02-0.02	0.02-0.03	0.04-0.06	0.01-0.02	1.39-1.40
Mean	0.02	0.025	0.048	0.02	1.403
Cassava Peel	0.01-0.02	0.02-0.03	0.02-0.03	0.01-0.02	0.93-1.0
Mean	0.02	0.022	0.023	0.02	0.995
WHO/FAO, 2007, value in food	2.0	0.02	1.30	0.03	99.40

Table 3. Calculated values of Bioaccumulation Factor (BAF), Estimated Daily Average Intake (EADI), and Total Hazard Quotient (THQ) of metals

Metal	Bioaccumulation factor (BAF)		EADI, mg/day/kg /body wt.		Rf Dose, mgKg/day		Total hazard quotient (THQ)		$HI = \sum THQ$
	Cassava tuber	Peel	Cassava tuber	Peel	Cassava tuber	Peel	Cassava tuber	Peel	
As	0.01	0.01	0.0119	-	0.0003	-	0.039	--	
Cd	1.25	1.1	0.747	-	0.001	-	0.747	-	
Cr	0.037	0.017	0.0278	-	0.003	-	0.0095	-	
Pb	0.667	0.667	0.0119	-	0.0035	-	0.0034	-	0.8017
Zn	0.014	0.01	0.838	-	0.3	-	0.0028	-	
Reference							[18]		

Mean Pb levels in soil from the farms were 0.04 mg/Kg (Table 2). This level was lower than the WHO/FAO, 2017 permissible level of 60.0 mg/Kg. The PLI of Pb in soil was 0.007 (Table 2), which was below the threshold level of 1.0 an indication that there was no likelihood of Pb loading in analysed soil. A similar study [21] conducted around small-scale gold-mining areas in Wassa-Amenfi-West District of Ghana reported Pb PLI of 54.18 in soil. An analogous study conducted in crude oil contaminated soil at Ikot Ada Udo in Nigeria reported 0.51 mg/Kg of Pb in cassava tuber (NR-8082 variety) [23]. This level was higher than the 0.04 mg/Kg reported in this study. Both the cassava tuber and its peel had Pb bioaccumulation factor of 0.667 (Table 3), a value which is indicative of the insignificant likelihood of adverse health effects of lead poisoning on the consumption of cassava tuber and its peel by humans and animals in the study area. The bioaccumulation factor for Pb from soil to cassava tuber cultivated on crude oil contaminated soil at Ikot Ada Udo in Nigeria was 0.37 in NR-8082 variety [23]. A related study reported a Pb level of 0.608 mg Kg/ of Pb in cassava leaf cultivated in the River State of Nigeria [24], with a bioaccumulation factor of 0.239 in cassava leaf [24]. Lead showed a strong positive correlation of 0.926 for Pb transfer from soil to cassava tuber, suggesting that, the cassava tuber is a good accumulator of Pb. The calculated EADI and THQ for Pb in cassava tuber were 0.0119 and 0.0034 respectively (Table 3).

Chromium levels in soil ranged from (0.06-0.38) mg/Kg

with a mean level of 0.285 mg/Kg (Table 2). This level was lower than the acceptable level of 100.0 mg/Kg in agricultural soils. A similar study reported Cr level of 0.02 mg/Kg in crude oil-contaminated soils in Nigeria [23], and a bioaccumulation factor of 0.02 in NR-8082 cassava tuber variety [23]. The soil PLI for Cr was 0.0029 (Table 2) which was far below the threshold of 1.0. However, the bioaccumulation factor of Cr in the cassava tuber was 0.037, while the peel had a bioaccumulation factor of Cr in the peel to be 0.017 (Table 3). The cassava tubers were more Cr tolerant compared to its peel. Nevertheless, the bioavailability of Cr in terms of bioaccumulation factors in the cassava tuber and its peel was less than the threshold value of 1.0, below which there is no obvious health risk to both humans and animals.

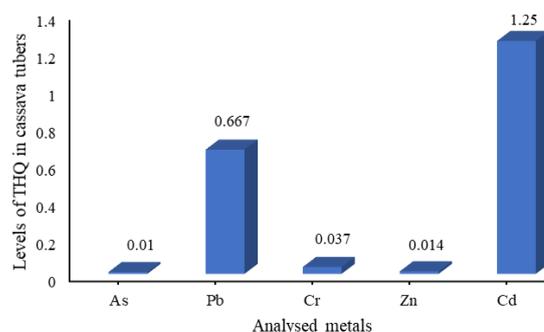


Figure 2. Relative contributions of analysed metals to health index in cassava tubers

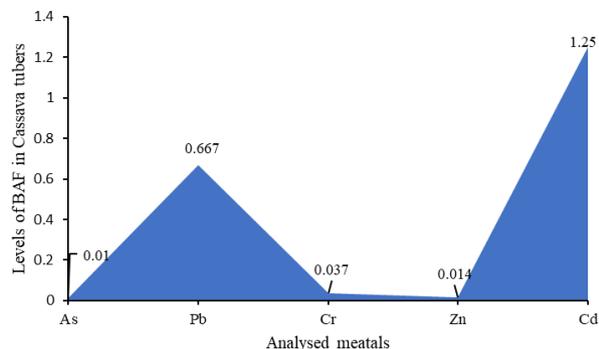


Figure 3. Relative (bioaccumulation factor) tolerance of analysed metals in cassava tubers

Zinc levels in the soil ranged from (1.86-1.90) mg/Kg with a mean of 1.885 mg/Kg (Table 2). The level of Zn was below the permissible WHO/FAO, 2017 guideline level of 200.0 mg/Kg. The PLI of Zn in soil was 0.009 (Table 2), which is insignificant considering a threshold of 1.0. The Zn levels in NR-8082 variety reported in crude oil-contaminated soils in Nigeria was 17.5 mg/Kg [23] and the corresponding bioaccumulation factor of Zn in the same cassava tuber variety was 0.26. Bioaccumulation factors of Zn in the cassava tuber and its peels were 0.014 and 0.01 respectively (Table 3). These values were lower than the threshold value of 1.0 suggesting an adverse effect of Zn on the consumption of cassava and its peel by humans and animals. A related study reported a soil to plant bioaccumulation factor of 0.894 in cassava leaf cultivated in the river State of Nigeria [24]. There was a positive correlation of 0.689 for Zn transfer from soil to cassava tuber, illustrating a high preference for Zn in the cassava tuber. The EADI and THQ of Zn in cassava tuber were 0.838 and 0.0028 respectively (Table 3).

Cadmium levels in soil ranged from 0.03 mg/Kg to 0.06 mg/Kg with a mean concentration of 0.048 mg/Kg (Table 2) compared to the WHO/FAO, 2017 standard value of 1.0 in agricultural soil. The PLI of Cd in soil was 0.048 (Table 2) which is lower than the threshold level of 1.0. Cadmium levels reported in a similar contaminated soil in Nigeria reported 0.13 mg/Kg of Cd in NR-8082 variety cassava tubers, a level higher than 0.048 mg/Kg in the present study [23].

Cadmium had a bioaccumulation factor of 1.25 in the cassava tuber and 1.1 in its peels (Table 3). This implied that cassava and its peel are natural accumulators of Cd since the bioaccumulation factors were all above a threshold value of 1.0. These factors of bioaccumulation in cassava and its peels are of much concern because, Cd is a hazardous metal that competes with Calcium in bone deposition and replaces calcium in bone, making the mammalian bone to be fragile. Cadmium poses a potential health hazard to both humans and animals via the consumption of cassava and its peel from the study farms. A similar study reported soil to cassava leaf bioaccumulation factor of 0.013 in the River State of

Nigeria [24]. The EADI and THQ for Cd in cassava tuber was 0.747.

The sum of the total health quotient for all the metals was 0.8017 (Table 3). This value is lower than a threshold of 1.0, below which health effects are not obvious. Although peels are not consumed by humans directly, animals that feed on cassava peels can pass it on to humans through the food chain.

Metal tolerance in terms of bioaccumulation was in the order Cd > Pb > Cr > Zn > As while that of the peel was in the order Cd > Pb > Cr > Zn > As. Both cassava tuber and the peel seemed to follow the same trend in metal tolerance. Cassava had a higher tolerance for Cd compared to the other metals. Cadmium also recorded the highest total hazard quotient of 0.747, a factor in determining the human health index, hence contributing positively to the health index.

4. Conclusions

Evaluation of the soil pollution load index (PLI), suggested that all the metals were below the threshold level of 1.0, an indication of good metal levels for agricultural soil. However, the soil-peel-cassava transfer which is the bioaccumulation factor for Cd in cassava tuber, and its peel were 1.25 and 1.1 respectively, and were all above the threshold of 1.0, indicating that cassava and its peel had a higher tolerance for Cd compared to the other metals. Cadmium also recorded the highest total hazard quotient of 0.747, a factor in determining the human health index, hence contributing positively to the health index.

In general, the cumulative Health Index (HI) of all the analysed metals of 0.8017 < 1.0 (a threshold level below which the food is deemed to be safe) which is the sum of all the total hazard quotients (exposure to heavy metals) indicated that there is no potential health effect associated with the consumption of cassava tubers cultivated on reclaimed mining land sites in the Prestea-Huni Valley District.

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Conflict of Interest

The authors have no conflict of interest to declare.

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