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HEALTH RISK ASSESSMENT OF HEAVY METAL CONTAMINATION OF SOME INDIGENOUS VEGETABLES GROWN IN OIL SPILLED AGRICULTURAL AREA IN OGONILAND, RIVERS STATE, NIGERIA

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Abstract

This study seeks to evaluate the potential health risks associated with the consumption of selected heavy metals in soils and selected vegetables from polluted sites. The vegetable includes; *Gongrenema latifolium* (GL), *Ocimum gratissimum*(OG), *Vernonia amygdalina*(VA) and *Talinum triangulare* (TT), harvested from a crude oil polluted area in Bodocity, Ogoniland., Rivers State. The concentrations of Fe, Pb, Zn, Mg, Cd and Cr were determined using Atomic Absorption Spectroscopy (AAS). The mean concentrations of heavy metals (Fe, Pb, Zn, Mg, Cd and Cr) for the samples ranged from 0.36±0.01 in GL (Control) to 16.82±0.01 in the polluted soil (SS); <0.0001(BDL) to 0.48 in VA obtained from polluted site; $0.19±0.00^a$ in SS (control) to $2.67±0.01^a$ in OG (control); $3.51±0.01^b$ to $470.33±0.33^a$ in TT control; BDL to $0.04±0.00^b$ in VA polluted and BDL to $0.03±0.00^a$ in SS polluted (in mg/kg respectively). Pb concentrations in VA, TT and SS in polluted site exceeded the permissible limits of 0.2mg/kg, while other heavy metal levels were within safe limits. The metals were in the order of abundance Mg>Fe>Zn>Cr>Pb>Cd. Health Risk was assessed by computing the average Daily Intake of Heavy Metals, Target Hazard Quotient and Carcinogenic Risk for Heavy Metals. The daily intakes of Heavy metals from vegetable consumption were within the threshold values for Cr, Zn, Pb, and Cd except for contaminated soil, which exceeded the threshold value. The observed THQ of Cr, Zn, Pb and Cd were generally < 1. This observation is suggestive that consumers of vegetables from the study sites may not experience significant health risks from intake of individual metals through vegetable consumption except THQ value >1 for Fe which maybe a concern to consumers of crops planted in the Bodocity soil. The CR values of Pb, Cd and Cr in GL, VA, TT and SS were within 10^{-6} to10^{-4}, indicating that consumption of vegetables grown in the oil spilled area may likely result in cancer over a period of 70 years. The CR values indicated in this study were within the predicted permissible lifetime cancer risk (10^{-6}-10^{-4}) for all the samples assessed, constant consumption/exposure to these Heavy metals may result in their bioaccumulation overtime which is implicated in serious health risk.

Keywords: Daily Intake of Heavy Metals, Target Hazard Quotient, Carcinogenic Risk, Heavy metal, Health risk assessment, Bodocity, Ogoniland

Introduction

Heavy metal contamination is one of the major environmental hazards affecting people's health globally. This is mainly through the consumption of contaminated food, water and environmental exposure (Onyedikachi *et al.,* 2018; EFSA, 2021; Taghizadeh *et al.,* 2021). The essentiality of food for man's existence has attracted researchers to the risk associated with consumption of contaminated foodstuffs, i.e. pesticides, heavy metals and or toxins in vegetables (D'mello, 2016; Abdollatif *et al.,* 2019). The sources of potentially harmful heavy metals (HMs) in soils may not just be solely from the bedrock itself, but also from anthropogenic sources like solid or liquid waste deposits, agricultural inputs, and fallout of industrial and urban emissions (Kachenko and Singh, 2016). Excessive heavy metal accumulation in agricultural soils may result not only in soil contamination, but also in unfavorable consequences for food quality and safety. It is therefore essential to monitor food quality, given that plant uptake is one of the main pathways through which HMs enter the food chain (Antonious, 2020). Heavy metals are relatively non-biodegradable, hazardous in food and environment, and possess a long biological half-life. The agricultural sector suffers a great deal from the impacts associated with the level of contamination from these elements. As such, contamination and subsequent pollution of the environment by heavy metals have become of great concern due to their distribution and deleterious effects on the ecosystem (Hazrat and Ezzat, 2019).

Vegetable refers to the edible part of the plant that stores food in roots, stems, or leaves. Vegetables are green and leafy-like in appearance bearing edible stems or leaves and roots of plants (Javid *et al.,*2018).Vegetables take up HMs and accumulate them in their edible and non-edible parts to levels that could cause clinical problems to both animals and humans (Yeganeh *et al.,* 2017). As an example, the consumption of contaminated food can seriously deplete some essential nutrients in the body leading to weakened immune system, malnutrition and a high prevalence of upper gastrointestinal cancer (Guerra *et al.,* 2018). Zuo *et al.* (2019) reported that soil and vegetables contaminated with Pb and Cd in Copsa Mica and Baia Mare, Romania, significantly contributed to decrease in human life expectancy (9-10 years) within the affected areas. Heavy metals can be very harmful to the human body even at low concentration as there are no effective excretion mechanisms (Ghosh *et al.,* 2012; Onyedikachi *et al.,* 2019b). Lead (Pb) gives rise to adverse effects in several organs and systems in all known animal species, such as the blood, central nervous system, kidneys, reproductive and immune systems (NCM, 2016). Cadmium (Cd) is persistent and accumulates mainly in the kidney and liver of vertebrates, producing severe diseases in these organs (UNEP, 2018). Chromium (Cr) can cause skin ulcers and nasal septum perforations (USEPA, 2019). While both Cr (III) and Cr (VI) can be toxic to plants and animals, Cr (III) toxicity occurs in higher concentrations, and this form is actually an essential nutrient to human and other animals. Cr (VI), on the other hand, is toxic in much lower concentrations and tends to be more mobile and bioavailable than Cr (III) in surface and subsurface environments (Adriano, 2001). Compared with other pathways such as inhalation and dermal contact, dietary intake is the main route of exposure to heavy metals for most people (Yeganeh *et al.,* 2017).

Ogoniland in Rivers State, Nigeria, is a part of the Niger delta region, which has a tragic history of pollution from oil spills and oil well fires because of pipeline failure, illegal bunkering, and artisanal refining (Whanda *et al.,* 2016; Njoku *et al.,* 2019). However, there is no systematic scientific information on the ensuing contamination and health risk implications of vegetables consumed in the area. Within Rivers State alone, Osuji and Adesiyan (2019) noted that oil spillage and gas flaring regularly occur, thus, destroying large part of the coastal vegetation, polluting water bodies, damaging fertile agricultural lands and this has led to ethnic and regional crisis (Wegwu *et al.,* 2017). Some studies have been done on health risks associated with consumption of root tubers, seafoods, drinking water, plantain and fish obtained from Ogoniland (Nwaichi *et al.,* 2014; Nkpaa *et al.,* 2016; Patrick-Iwuanyanwu and Udowelle, 2017; Nkpaa *et al.,* 2017; Dikioye *et al.,* 2018; Onyedikachi *et al.,* 2018). Due to paucity of data with regards to health risk associated with vegetable consumption in Bodocity, Gokana, Ogoniland, this study seeks to evaluate the potential health risks associated with heavy metals via consumption of selected vegetables; Bush buck (*Gongrenema latifolium)*, Clove basil (*Ocimum gratissimum)*, *bitter* leaf (*Vernonia amygdalina)*, and *water* leaf (*Talinium triangulare)* grown on oil contaminated sites in Bodocity, Ogoniland, and those grown in the reference location at Umuchichi village in Osisioma Ngwa LGA, Abia State. With the use of health risk assessment indices such as Daily Intake of Heavy Metals (DIM), the Target Hazard Quotients (THQs) and Cancer Slope Factors, we assessed the potential health risk exposure associated with consumption of foods grown in heavy metal polluted soils. (Onyedikachi *et al.,* 2018; Yabanli and Alparslan, 2018; Onyedikachi *et al.,* 2019a).

Material and Methods

Study area

Samples used in this study were obtained from Bodo city in Ogoniland Rivers State, Nigeria, located on Latitude 4°40′5″N and 4° 43′ 19.5″N and Longitude 7° 22′ 53.7″E and 7° 27′ 9.8″E. The rainfall distribution of the study area ranges from 2000 to 3000 mm (Oyegun and Ologunorisa, 2002) per annum in a bimodal with average temperature of about 28 to 35°C (Oyegun and Ologunorisa, 2002) depending on the season of the year. This vicinity is known for environmental pollution due to oil exploration activities. The reference (control) area is located at Umuchichi village in Osisioma Ngwa LGA, Abia State. Osisioma Ngwa lies within latitude 5^{0}19'32"N and longitude 07^{0}15′49" and 07° 25′23"E with an area of 198km^{2}. Umuchichi has marked lowlands and gentle slopes especially in areas around the Aba river, unlike the community close by (Ayaba Umueze) characterized with tablelands. There are two seasons: rainy and dry. The rainy season starts in early march and often terminates in November. Annual rainfall in Umuchichi usually ranges from 2000-2200mm, bi-modally distributed with peaks in July and September. The relative humidity is usually high, especially during the raining season soil (Nwankwo *et al.,* 2019). The selection of the reference area was based on availability of the samples and the absence of crude oil exploration activities.

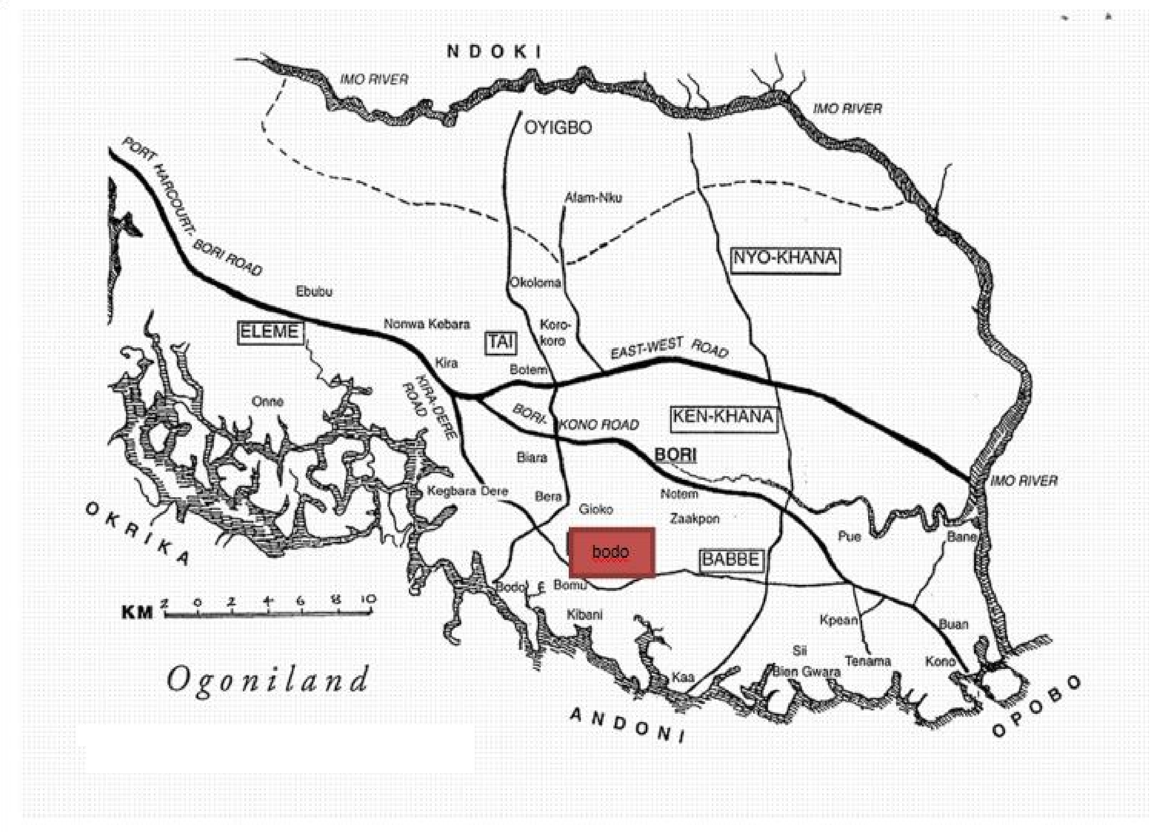

Fig 1: Map of Bodocity in Bodocity, Ogoniland (Ujoh and Ifatimehin, 2014)

Sample collection and preparation

A total of eight samples of four different vegetables namely; *Talinum triangulare*, *Gongronema latifolium*, *Ocimum gratissimum*, and *Vernonia amygdalina* were harvested from agricultural zones at Bodocity, Ogoni land(contaminated site) and Umuchichi(control site) each. Soil samples were also collected from the same farm zones.

Sample preparation

At each study site, the diagonal length of each sampling site was marked into five equal points. Soil samples were taken using a soil auger at each marked point including the soil adhering to the roots of the vegetables. A composite soil composed of all the soil samples collected at each point was prepared for further analysis after the manual removal of non-soil particles like stone and wooden particles and then transported to the laboratory for further analysis (Onyedikachi *et al.,* 2019b). Soil and vegetable samples were stored (at ambient temperature) in paper bags in the field and transferred to the laboratory as soon as possible for pre-treatment and analysis (Zeng *et al.,* 2009; Rehman *et al.,* 2017). At the laboratory, the soil samples were air dried for three days i.e., after a steady weight was achieved, it was ground and sieved using a 2mm stainless steel mesh and stored at 4°C in the refrigerator prior to chemical analysis. Furthermore, the fresh vegetables were washed with distilled water to remove dirt particles. Afterwards, the vegetables leaves were separately plucked, selected and spread out on a flat foiled surface to air dry. After the water had evaporated, the vegetable samples were oven dried at 65°C for 48hours before crushing it into powder.

a. Soil sample digestion for heavy metal analysis

A measured weight (0.5g) each of the processed soil samples was added into different test-tubes, 10mls of aqua-regia HNO_3/HCL (1:3) at 110°C was added to each test-tube and heated for 2hours. After the time had elapsed, complete digestion did not take place, so additional 5mls of aqua-regia HNO_3/HCL (1:3) at 110°C was added and heated for extra one (1) hour to achieve complete digestion until the brown fumes disappeared. Then, 20ml of distilled water was added and heated until a colorless solution was obtained. The solution was allowed to cool and filtered into a standard volumetric flask (100ml) through Whatman No. 42 filter paper and the volume was made to the mark with distilled water (Iyaka, 2007).

b. *Vegetable Sample digestion for Heavy metal analysis*

A measured weight (0.5g) of each of the leaf sample was weighed into different test-tube and 10ml nitric acid was added to each sample in the beaker. It was heated in a water bath for 1-2hour(s) at 100°C for complete digestion to take place. Afterwards, 20ml of distilled water was added to the sample solution and the setup was heated until a colorless solution was obtained. The solution was then allowed to cool and filtered into a standard volumetric flask (100ml) through Whatman

No. 42 filter paper and the volume was filled to the mark with distilled water (Iyaka, 2007). The digested sample was analyzed for heavy metals (Fe, Pb, Zn, Mg, Cd, Cr) using Atomic Absorption Spectrophotometer (AAS) according to the methods of AOAC (2008).

Quality assurance and quality control

Distilled water was used throughout the study (Prajapati and Patel, 2010). Glassware and plastic ware (Merck, Germany) used were thoroughly rinsed with 10% HNO_3 followed by washing with distilled water. For quality control data assurance, each sample was analyzed in triplicate. Determination of metals was performed with Atomic Absorption Spectrophotometer (AAS) VGP 210 model. Analytical blanks were run in the same way as the samples, and concentrations determined using standard solutions prepared in the same acid matrix as reported by Yesudhason *et al.* (2013). Standards for the instrument calibration were prepared based on mono element certified reference solution Inductively Coupled Plasma Standard (Merck, Germany). The health risks associated with the consumption of heavy-metal-contaminated vegetables and soil sample were assessed based on the Daily Intake of Heavy Metals (DIM), Target Hazard Quotient(THQ) and Cancer Risk(CR).

Daily Intake of Heavy Metals (DIM)

According to Khan *et al.,* (2008); Mahmood and Malik (2014) and Onyedikachi *et al.* (2018), the daily intake of heavy metals is determined using the following equation:

$$\text{DIM} = \frac{\text{Concentration of heavy metal x Daily food intake} \ldots\ldots 1}{\text{Average weight}}$$

Calculations were made based on the standard assumption for an integrate USEPA risk analysis, considering an adult average body weight of 60kg and the average daily vegetable intake for adults is considered to be 0.345kg person^{-1} day^{-1} (Avila *et al.* ,2016; Wang *et al.,* 2018; Onyedikachi *et al.,* 2019b).

Target Hazard Quotient (THQ) & Hazard Index (HI)

The human health risks from consumption of vegetables by local populace were assessed based on the THQ. It is defined as the ratio between exposure and reference oral dose (RfD). This is used to express the risk of non-carcinogenic effects (Yu-Jun *et al.,* 2011). If the ratio is equal to or greater than 1, an exposed population experiences health risks. HI is the summation of THQs used to evaluate the potential risk to human health when more than one heavy metal is involved. The calculated sum is called the Hazard quotient. The methods used for the estimation of THQ and CR have been provided in USEPA Region III Risk-Based Concentration Table, January–June 1996 (Han *et al.,* 1998; USEPA 1996, 2011a; Chien *et al.,* 2015; USDOE, 2011) based on the equation thus:

$$\text{THQ} = \frac{\text{Concentration of heavy mental * Daily food intake} \ldots\ldots\ldots..(3)}{\text{RfD * Average weight}}$$

$$\text{THQ1} + \text{THQ2} + \text{THQ3} + \text{THQn} \ldots\ldots\ldots\ldots\ldots(4)$$

Where THQ is the target hazard quotient, DIM is the daily intake of heavy metals (mg/kg/day), heavy metal concentration in vegetables is expressed in mg kg^{-1}, average body weight is 60kg, and R_fD is the oral reference dose (mg/kg/day). The average daily vegetable intake for adults is considered to be 0.345kg person^{-1} day^{-1}(Avila *et al.,* 2016; Wang *et al.,* 2018; Onyedikachi *et al.,* 2019b). RfD is an estimate of a daily oral exposure for human population, which does not cause harmful effect during a lifetime; it is usually used in EPA's non-cancer health assessment (Mahmoud and Abdel-Mohsein, 2015; Guerra *et al.,* 2018).

Ingestion cancer slope factors

The Ingestion Cancer Slope Factors evaluate the probability of an individual developing cancer from oral exposure to contaminant levels over a period of a lifetime as described by USEPA (1998a) and ATSDR (2010). Ingestion cancer slope factors are expressed in units of (mg/kg/day)-^{1}.Lifetime probability of contracting cancer due to exposure to site-related chemicals is calculated as follows:

$$\text{ILCR} = \text{DIM x CSF} \ldots\ldots\ldots..(5)$$

Where, ILCR is Lifetime probability of cancer; DIM is the daily intake of each heavy metal (mg/kg/day), and CSF is the ingestion cancer slope factor (mg/kg/day)$^{-1}$.

Statistical Analysis for Metal Analysis

The four selected vegetables and soil analyzed individually in triplicate. The Least Significant Difference (LSD) was used to compare differences in each sample within treatments. Data was reported as mean ± S.E. One-way analysis of variance (ANOVA) was used to determine significant differences between groups, considering a level of significance of less than or equal to 0.05 ($p \leq 0.05$) using SPSS version 20.0 software.

Results and Discussion

Mean concentration of heavy metals (mg/kg dry weight) in soil and selected vegetables (*Gongrenema latifolium*, *Ocimum gratissimum*, *Vernonia amygdalina*, *Talinium triangulare*, Soil sample in contaminated (Bodocity, Ogoniland.) and control area (Umuchichi) are presented on Table 1. The results are expressed as triplicate mean ± S.E. Heavy metals analysed in this study are as follows; Lead (Pb), Cadmium (Cd), Chromium (Cr), Magnesium (Mg), Iron (Fe) and Zinc (Zn). The total concentrations were in the following abundance; Fe, Pb, Zn, Mg, Cd and Cr as follows, 10.45±0.05, BDL, 4.52±0.02, 726.50±33.38, BDL, 0.04±0.00 (Umuchichi) and 20.47±0.33, 1.16±0.00, 4.30±0.03, 736.08±2.68, 0.09±0.00, 0.05±0.00 (Bodocity, Ogoniland) mg/kg dry weight.

The concentrations of Fe in GL and OG from polluted site were higher than those from the control site due to high Fe levels from polluted soil is attributed to oil spills and burning of fossil fuel (LeCoultre, 2001; Nwaichi *et al.,* 2014; Wegwu *et al.,*2017; Onyedikachi *et al.,* 2017; Onyedikachi *et al.,* 2018; Onyedikachi *et al.,* 2019).

Also ,the mean concentration of Fe in OG, VA and TT did not show marked significant difference ($P > 0.05$), while those of TT and VA showed concentrations higher in samples from control site than those collected from the polluted site. This discrepancies observed indicates that Fe accumulation may not be basically as a result of Fe loads in the polluted site but could also be due to its participation in chlorophyll synthesis and photosynthesis (Mahfuza *et al.,* 2017). Apart from this, vegetables can be imparted aerially due to athropogenic activities in the environment and industries (Mahfuza *et al.,* 2017; Onyedikachi *et al.,*2018; Onyedikachi *et al.,*2019). Fe concentrations were generally higher than in vegetables, this agrees with many reports indicating that natural soils contains significant amount of Fe (Abdu *et al.,* 2011; Onyedikachi *et al.,*2018; Yahaya *et al.,* 2021). In addition, higher concentrations were observed in soil samples from Bodocity, Ogoniland than those of the control (Umuchichi) due to the high heavy metal levels in the crude oil polluted area (Nwaichi *et al.,* 2014). However, a high concentration of Fe in the soil cannot be conclusively attributed to a point source, as there may be other sources of Fe in the area. Besides, Fe has been reported to be a very abundant heavy metal in Nigerian soils (Yahaya *et al.,* 2021). The Fe concentration in soils ranged from 0.36±0.01mg/kg to 7.19±0.01mg/kg dry weight (Umuchichi) and from 0.38±0.01 to 16.82±0.01mg/kg dry weight (Bodocity, Ogoniland). These values were far lower than WHO/FAO safe limit of 425.00mg/kg in vegetables and 50000mg/kg in soil. Although, overtime, Fe loads may bioaccumulate in foods and soils resulting to several health risks associated with Fe toxicity. The toxicity of iron on cells has led to iron mediated tissue damage involving cellular oxidizing, reducing mechanisms, and their toxicity towards intracellular organelles such as mitochondria and lysosomes. Wide ranges of free radicals that are believed to cause potential cellular damage ensues excess intake of iron. The iron produced hydrogen free radicals attack DNA, resulting in cellular damage, mutation and malignant transformations that in turn cause array of diseases (Grazuleviciene *et al.,* 2009). High Fe toxicity shocks, hypotension, lethargy, tachycardia, hepatic necrosis, metabolic acidosis and sometimes death (Jaishankar *et al.,* 2014).

Lead contents showed a higher value in *Vernonia amygdalina* leaf from Bodocity, Ogoniland compared to selected vegetables and soil sample from Umuchichi, which was below detection limit of 0.0001. Values ranged below detection limit of 0.0001 to 0.48±0.05mg/kg dry weight (Bodocity, Ogoniland), while samples from Umuchichi were below f 0.0001mg/kg. There was significant variation ($p < 0.05$) between lead content from Bodocity, Ogoniland and Umuchichi in *Gongrenema latifolium*(GL), *Ocimum gratissimum*(OG*)*, *Vernonia amygdalina*(VA) and *Talinum triangulare* (TT) and soil sample. Pb concentration in the soils did not exceed the permissible limit (100mg/kg) set by WHO. However, the Pb content in VA *Vernonia amygdalina* (0.48±0.05mg/kg dry weight) from Bodocity, Ogoniland had concentration above the WHO/FAO permissible limit of 0.3mg/kg. Conversely, Pb contents in selected vegetables and soil sample from Umuchichi were below detection limit of 0.0001mg/kg. This is attributable to the oil spills in Bodocity, Ogoniland, compared to Umuchichi, where there was a low level of heavy metals in vegetables due to low bioavailability of the heavy metals in its soil. In many plants, lead accumulation can exceed several hundred times the threshold of maximum level permissible for human consumption (Muhammad *et al.,* 2008); which is toxic to the body and is not required even in the smallest quantity (Onyedikachi *et al.,* 2018). It accumulates in the bones and teeth causing weakness in the wrist and joints leading to brittle bones. It affects the central nervous system, kidney and liver leading to anemia, brain and nervous system damage, which are the health consequences of most concern.

The mean concentration of Zinc showed values ranging from 0.4±0.01mg/kg to 2.49±0.01mg/kg dry weight from Bodocity, Ogoniland, while those from Umuchichi ranged from 0.50±0.01mg/kg to 2.67±0.01 mg/kg dry weight. This is lower than WHO/FAO safe limit of 100.00mg/kg in vegetables and 300mg/kg for the soil samples too. There was significant variation ($p < 0.05$) between zinc content from Bodocity, Ogoniland and Umuchichi in *Ocimum gratissimum*, *Gongrenema latifolium*, *Vernonia amygdalina*, and *Talinum triangulare* leaves and soil sample. Many heavy metals, such as zinc, copper, chromium, iron and manganese, are essential to the body. However, they are needed in very minute quantities. However, if they accumulate in quantities that ensues toxicity to the biological system, serious health issues may occur. For example, zinc is essential to all organisms and is important in metabolism, growth, development, and general wellbeing. It is an essential co-factor for a large number of enzymes in the body. Its deficiency leads to heart diseases and various metabolic disorders (Saraf and Samant, 2013). However, Zinc overexposure may cause the flu-like symptoms of metal fume fever; stomach and intestinal disturbances; and/or liver dysfunction. Besides, Zn can also interrupt the activity of microorganisms and earthworms thereby reducing the metabolism of organic matter (Zulfiqar *et al.,* 2020).

Mean concentration of magnesium showed values that ranged from 3.50±0.01 mg/kg to 470.33±0.33 mg/kg dry weight from Umuchichi when compared with values that ranged from Bodocity, Ogoniland (8.07±0.07 mg/kg to 415.67±0.67 mg/kg dry weight). One way ANOVA revealed there was significant variation ($p < 0.05$) between magnesium content from Bodocity, Ogoniland and Umuchichi in *Vernonia amygdalina* and *Talinum triangulare* leaves. On the other hand, one way ANOVA revealed that there was no significant variation ($p > 0.05$) in the Mg level in *Ocimum gratissimum*, and *Gongrenema latifolium* leaves and soil sample (Table 1). Mg is the eleventh most abundant element by mass in the human body and is essential to all cells and some 300 enzymes for proper functioning (Kontani *et al*., 2005; Ayuk and Gittoes, 2014; Megan, 2020). Magnesium ions interact with bio-molecules such as ATP, DNA and RNA. Magnesium compounds are used medically as antacids and laxatives. They are also known to stabilize blood pressure and as such used in the treatment of conditions like eclampsia. Although, overdose from dietary sources alone is unlikely because the kidneys promptly filter excess Mg in the blood except in condition of impaired renal function. In spite of this, mega dose therapy can lead to death.

Other common symptoms of overdose are nausea, vomiting, and diarrhea. Other symptoms include; hypotension, confusion, slowed heart and respiratory rates, deficiencies of other minerals, coma, cardiac arrhythmia, and death from cardiac arrest (Megan ,2020).

The results showed that cadmium ranged below detection limit of 0.0001 to 0.04mg/kg dry weight from Bodocity, Ogoniland, while those from Umuchichi were below 0.0001. There was significant variation ($p < 0.05$) between cadmium content from Bodocity, Ogoniland and Umuchichi in *Ocimum gratissimum*, *Gongrenema latifolium*, *Vernonia amygdalina*, and *Talinum triangulare* leaves and soil sample. When the present concentrations of heavy metals were compared with the permissible limits by FAO/WHO (2001), it was found that all levels were within the safe limit which is below 0.1mg/kg(vegetables) and3.00mg/kg in soil. The results showed that cadmium from Bodocity, Ogoniland has the highest mean concentration of 0.04mg/kg dry weight in VA impling that uptake of heavy metals is increased in plants that are grown in areas with increased soil concentration of heavy metals (LeCoultre, 2001; Onyedikachi *et al.,*2018). Cadmium poisoning may be caused by ingestion of food (e.g. grains, cereals, and leafy vegetables) and cigarette smoke. Occupational exposure to cadmium in metal plating, battery, and plastics industries may also occur. Overexposure may cause fatigue, headaches, nausea, vomiting, abdominal cramps, diarrhea, and fever. In addition, progressive loss of lung function (emphysema), abnormal buildup of fluid within the lungs (pulmonary edema), and breathlessness (dyspnea) may be present. In some cases, affected individuals may exhibit increased salivation; yellowing of the teeth, tachycardia; anemia, bluish discoloration (cyanosis) of the skin and mucous membranes due to insufficient oxygen supply to these tissues; and/or an impaired sense of smell (anosmia), osteomalacia, renal tubular dysfunction, minor changes in liver function(Nwaichi *et al.,* 2014; NORD, 2021).

The mean concentration of chromium ranged from below detection limit of 0.0001 to 0.03±0.00 mg/kg dry weight from Bodocity, Ogoniland. While those from Umuchichi ranged from below detection limit of 0.0001 to 0.02±0.00 mg/kg dry weight. There was significant variation ($p < 0.05$) between chromium content from Bodocity, Ogoniland and Umuchichi in *Talinum triangulare*, and *Gongrenema latifolium* leaves and soil sample. Anova shows that there was no significant variation ($p > 0.05$) in the chromium level in the *Ocimum gratissimum* and *Vernonia amygdalina* leaves (Tables 1). When the present concentrations of heavy metals were compared with the permissible limits by FAO/WHO (2001), it was found that all levels were within the safe limit, which is 1.3mg/kg. The results showed that chromium from Bodocity, Ogoniland was of high mean concentration of 0.03mg/kg dry weight. This implies that uptake of heavy metals is increased in plants that are grown in areas with increased soil concentration of heavy metals (LeCoultre, 2001). Cr is used in the manufacture of cars, glass, pottery and linoleum. Over exposure to this metal may cause lung and respiratory tract cancer, kidney diseases, gastrointestinal disorders, heart diseases etc. Symptoms may lead to severe water-electrolyte disorders, increased mild acidity of blood and body tissues (acidosis), and/or inadequate blood flow to its tissues resulting in shock. Lesions on the kidneys, liver, and muscular layer of the heart (myocardium) may also develop (NORD, 2021).

The results showed that the total concentration was in the following abundance Fe, Pb, Zn, Mg, Cd and Cr as follows 10.45±0.05, BDL, 4.52±0.02, 726.50±33.38, BDL, 0.04±0.00 (Umuchichi) and 20.47±0.33, 1.16±0.00, 4.30±0.03, 736.08±2.68, 0.09±0.00, 0.05±0.00 (Bodocity, Ogoniland) mg/kg dry weight. The order of abundance is Mg>Fe>Zn>Cr>Pb>Cd (Umuchichi) and Mg>Fe>Zn>Cr>Pb>Cd (Bodocity, Ogoniland)

Health risk assessment of Heavy metal contamination of some indigenous vegetables grown in a crude oil contaminated site

Soil pollution with heavy metals due to the discharge of untreated crude oil spillage, gas flares is a major threat to ecological integrity and human well-being. In this study, our aim was to determine the human health risks associated, via food chain, with the contamination of some selected vegetables by heavy metals from crude oil spillage. Health risk assessment of heavy metals in contaminated vegetables is an important issue that is worth studying. The accumulation of these heavy metals in vegetables may represent a health risk, especially for populations with high consumption rates (Burger and Gochfeld, 2009; Diez *et al.,* 2009). Therefore, estimated daily intake or tolerable intake is widely used to define safe levels of intake of heavy metals (Kumar and Mukherjee, 2011). Hazard Quotient (HQ) characterized the health risk of intake of metal-contaminated vegetables to human health. The toxicological characteristic of the investigated metals presented on Table 3 shows the values for the reference dose and cancer slope factor used in the health risk calculation. This is a ratio of determined dose to the reference dose (R_fD). The population may not be at risk if value is less than one and if the value is equal or greater than one, then population is likely experience health risk. This risk assessment method has been used by researchers (Chien *et al.,* 2015; Onyedikachi *et al.,*2017; Onyedikachi *et al.,*2018; Wang *et al.,* 2018; Sridhara *et al.,* 2019) and proved to be valid and true. The values of R_fD for heavy metals were taken from Integrated Risk Information System (Abdollatif *et al.,* 2019) and Department of Environment, Food and Rural Affairs (DEFRA, 2020).

The Daily Intake of Heavy Metal (DIM) for individual vegetables is presented in Table 3. The DIM of iron from consumption of the selected vegetable and soil sample ranged from 0.002mg/kg for GL to 0.04 mg/kg body weight for soil in Umuchichi, while those from Bodocity, Ogoniland ranged from 0.002mg/kg body (VA) to 0.09mg/kg body weight for Soil sample in Bodocity, Ogoniland. The DIM for SS for both areas exceeded the established reference dose of 0.007mg/kg/day recommended by FAO/WHO (1993); USEPA (2011a) and USDOE (2011). Excess iron uptake is a serious problem and it increases the risk of cancer. The DIM of lead from consumption of the selected vegetable and soil sample from Umuchichi was below detection limit of 0.0001, while those from Bodocity, Ogoniland ranged below 0.0001mg/kg to 0.003mg/kg body weight. The highest DIM value of Pb was obtained from the consumption of *Vernonia amygdalina* leaf from Bodocity, Ogoniland. The DIM of lead was below established reference dose of 0.0035mg/kg/day recommended by FAO/WHO (1993), USEPA (2011a) and USDOE (2011).

The DIM of zinc from consumption of the selected vegetable and soil sample ranged from 0.001mg/kg to 0.015 mg/kg body weight for Umuchichi, while those from Bodocity, Ogoniland ranged from 0.002mg/kg to 0.014mg/kg. The highest DIM value of Zn was obtained from the consumption of *Vernonia amygdalina* leaf from Umuchichi. The DIM of zinc was below established reference dose of 0.3mg/kg body weight recommended by FAO/WHO (1993), USEPA (2011a) and USDOE (2011). The DIM of chromium from consumption of the selected vegetables and soil sample where below detection limit of 0.0001mg kg^{-1} body weight for both sites (Umuchichi and Bodocity, Ogoniland). The DIM for Chromium in this study was below the established tolerable daily intake of 0.03mgkg^{-1} body weight (FAO/WHO 1993; USEPA, 2011a; USDOE, 2011). The DIM of cadmium from consumption of the selected vegetable and soil sample from Umuchichi was below detection limit of 0.0001, while those from Bodocity, Ogoniland ranged from below detection limit of 0.0001 to 1.96E^{-05} mg/kg body weight. The highest DIM value of Cd was obtained from the consumption of *Vernonia amygdalina* leaf from Bodocity, Ogoniland (1.96E-05mgkg^{-1} body weight).The DIM recorded for Cd in vegetable and soil sample were also below the established tolerable daily intake of 0.001mgkg^{-1}.

The results for Target Hazard Quotient which is used to express the risk of an individual showing non-carcinogenic effects (Yu-Jun *et al.,* 2011) in this study is presented on Table 4. If the ratio is equal to or greater than 1, an exposed population experiences health risks. The interpretation of the THQ value is binary; THQ is either >1 or <1, where THQ >1 indicates a reason for non cancer health risk concern (Bassey *et al.,* 2014). The observed THQ of Cr, Zn, Pb and Cd were generally less than one. This observation is suggestive that consumers of vegetables from the study sites would not experience significant health risks from intake of individual metals through vegetable consumption. On the other hand, the observed THQ value >1 calculated for Fe may be of concern to consumers of crops planted in the soil sample from the contaminated sites. Li *et al.* (2014) and Krishna *et al.* (2014) reported that highest THQ value poses relatively higher potential health risk to human beings especially for the people residing in the area with serious metal pollution. The hazard index is a mathematical approach used to ascertain the risk ensued from an exposure to various chemicals (in this case heavy metals) through consumption of food. When the values are less than and equal to 1 it indicates that adverse effects are not likely, thus can be considered to be negligibly hazardous, however, the hazard index greater than 1, the exposure to these heavy metals is considered to be lethal. The result for Hazard Index showed also on Table 4 indicates that the exposure of the population to all samples from bodocity except for GL may cause adverse effect on their health. In addition, the hazard index was greater than 1 in VA grown in the control site (umuchichi), while other samples had values below 1, indicating that its heavy metal loads may not be a concern for the consumers in that region. However, it is pertinent to note that constant exposure to heavy metals overtime may bioaccumulate, resulting in various illnesses (Onyedikachi *et al.,* 2018).

The Ingestion Cancer Slope factor is estimated and expressed as a probability of contracting cancer over a lifetime of 70 years. In this present study, the average value of Ingestion Cancer slope factor for cancer risk for the Fe, Zn, Cd, Cr, Pb and Mg in this study is presented on Table 5. Fe, Zn and Mg did not show carcinogenicity as the Cancer Slope Factor. According to USEPA, CR between 10^{-6} (1 in 1,000,000) and 10^{-4} (1 in 10,000) represent a range of permissible predicted lifetime risks for carcinogens (Onyedikachi *et al.,* 2018). Contaminants, for which the risk factor is below 10^{-6} are eliminated from further consideration as a chemical of concern (Onyedikachi *et al.,* 2019). The ingestion cancer slope factors are not given for Fe, Zn and Mg because of its unique characteristics, therefore, the carcinogenic risk were not assessed. The carcinogenic health risk of a Pb in the polluted site for this study gave a range of 1.42E^{-05} in TT to 2.35E^{-05} in VA, indicating that consumption of vegetables planted in the oil spilled area may likely result in cancer over a period of 70 years. CR values for Cd and Cr had its highest value in VA 8.74E^{-05} and 8.63E^{-05} in soil respectively indicating that consumption of vegetables and exposure to soil in this area for Cd and Cr shows the probability of contracting cancer over a lifetime of 70 years. Generally, CR values indicated in this study were within the range of permissible predicted lifetime risks for carcinogens for all the samples assessed, constant consumption/exposure to these Heavy metals may result in bioaccumulation, which may result in serious health risk.

Conclusion

The toxicity level of Heavy metals [Lead (Pb), Cadmium (Cd), Chromium(Cr), Magnesium (Mg), Iron (Fe) and Zinc (Zn)] in selected Vegetables from Bodocity, Ogoniland, were analyzed and their potential health risks estimated. The result obtained from this study shows that the levels of Pb in VA, TT and SS exceeded the WHO/FAO permissible limit of 0.2mg/kg, which indicates a level of concern for the populace. They other metals analyzed were within permissible range. The discrepancies observed, as some metal concentrations were higher in control samples like in the case of Mg, Zn and Fe in GL and SS samples. However, prolonged consumption can pose great danger as these negligible quantities build up over time to become significant, thus endangering lives. The observed THQ of Cr, Zn, Pb and Cd were generally < 1, suggesting that consumers of vegetables from the study sites might not experience significant cancer health risks. The THQ for Fe because of abundance of Fe in soils may be important for further geochemical studies in Bodocity area. The CR values of Pb, Cd and Cr in GL, VA, TT and SS were within 10^{-6}to10^{-4} (range of permissible predicted lifetime risks for carcinogens) indicating that consumption of vegetables grown in the oil spilled area may likely result in cancer over a period of 70 years as a result of bioaccumulation overtime. Biological indicators have been widely applied for the monitoring of environmental pollutant in health exposure risk assessments. The relationship however, between external and internal levels of pollutants have not been adequately established, therefore it is difficult to accurately assess the potential health risk associated toxic metals. There is therefore need for the monitoring of these substances in the human diet as increased dietary intake from various foods may contribute in the development of various health disorders associated with heavy metal toxicity.

Table 1: Mean concentration of heavy metals (mg/kg dry weight) in soil and selected vegetables in contaminated (Bodocity, Ogoniland) and control area (Umuchichi). The results are expressed as triplicate mean ± S.E

Heavy Metals	**GL**	**OG**	**VA**	**TT**	**SS**	**WHO/FAO FOOD**	**SOIL**
IRON(Fe) C.	0.36 ± 0.01^{a}	1.34 ± 0.01^{b}	0.62 ± 0.01^{b}	0.94 ± 0.01^{b}	7.19 ± 0.01^{a}	**425.00**	**50000**
P.	1.17 ± 0.01^{b}	1.42 ± 0.01^{b}	0.38 ± 0.01^{b}	0.68 ± 0.29^{b}	16.82 ± 0.01^{b}		
LEAD(Pb) C.	BDL ^{a}	BDL ^{a}	BDL ^{a}	BDL ^{a}	BDL ^{a}	**0.30**	**100**
P.	BDL	BDL	0.48 ± 0.05^{b}	0.29 ± 0.00^{b}	0.39 ± 0.00^{b}		
Zinc(Zn)	0.59 ± 0.00^{a}	0.50 ± 0.01^{a}	2.67 ± 0.01^{a}	0.57 ± 0.00^{a}	0.19 ± 0.00^{a}	**100**	**300**
C.	0.47 ± 0.00^{b}	0.41 ± 0.01^{b}	0.47 ± 0.00^{b}	2.49 ± 0.01^{b}	0.46 ± 0.01^{b}		
Magnesium(Mg) C.	103.00 ± 1.53^{b}	72.33 ± 31.18^{b}	77.33 ± 0.33^{a}	470.33 ± 0.33^{a}	3.51 ± 0.01^{b}	**--**	**--**
P.	113.00 ± 0.58^{b}	79.67 ± 0.67^{b}	119.67 ± 0.67^{b}	415.67 ± 0.67^{b}	8.07 ± 0.07^{b}		
Cadmium(Cd) C.	BDL^{a}	BDL^{a}	BDL^{a}	BDL^{a}	BDL^{a}	**0.10**	**3.00**
P	BDL^{a}	BDL^{a}	0.04 ± 0.00^{b}	0.03 ± 0.00^{b}	0.02 ± 0.00^{b}		
Chromium(Cr) C.	0.02 ± 0.00^{a}	BDL^{b}	BDL^{b}	0.02 ± 0.00^{a}	BDL^{b}	**0.20**	**100**
P.	0.01 ± 0.00^{a}	BDL ^{b}	BDL ^{b}	0.01 ± 0.00^{a}	0.03 ± 0.00^{a}		

Values in different superscript letters (a, b) in the same row are significantly different at $P \leq 0.05$while same superscript letters (b) in the same column are not significantly different at $P > 0.05$. BDL - Below detection limit of 0.0001 mg/kg. Gongrenema latifolium GL, Ocimum gratissimum OG, Vernonia amygdalina VA, Talinium triangulare (TT), Soil sample(SS)

Table 2: Toxicological characteristic of the investigated metals

Metals	**Ingestion reference dose RfD (mg kg^{-1} day^{-1})**	**Ingestion carcinogenic slope factor CSF(mg kg^{-1} day^{-1})$^{-1}$**
Chromium (Cr)	0.003	0.5
Lead (Pb)	0.001	0.38
Lead (Pb)	0.0035	0.0085
Zinc (Zn)	0.3	0
Iron (Fe)	0.007	0

Source: FAO/WHO (1993), USEPA (IRIS, 2010), USDOE (2011), USEPA (2011a)

Table 3: Daily Intake of Heavy Metals in selected vegetables (mg/kg/day)

Heavy metals	Sample location	GL	OG	VA	TT	Soil
Fe	Control	0.002	0.008	0.004	0.005	0.041
	Polluted	0.007	0.008	0.002	0.004	0.097
Pb	Control	0.000	0.000	0.000	0.000	0.000
	Polluted	0.000	0.000	0.003	0.002	0.002
Zn	Control	0.003	0.003	0.015	0.003	0.001
	Polluted	0.003	0.002	0.003	0.014	0.003
Mg	Control	0.592	0.416	0.445	2.704	0.020
	Polluted	0.650	0.458	0.688	2.390	0.046
Cd	Control	0.000	0.000	0.000	0.000	0.000
	Polluted	0.000	0.000	0.000	0.000	0.000
Cr	Control	0.000	0.000	0.000	0.000	0.000
	Polluted	0.000	0.000	0.000	0.000	0.000

BDL- Below detection limit of 0.0001 mg/kg. CSF- Cancer Slope Factor. Gongrenema latifolium.(GL), Ocimum gratissimum (OG), Vernonia amygdalina (VA), Talinium triangulare (TT), and soil sample(SS) in contaminated (Ogoniland) and control area (Umuchichi)

Table 4: Target Hazard Quotient and Hazard Quotient of selected vegetables (mg/kg)

Heavy metals		RFD	GL	OG	VA	TT	SS
Fe	control	0.007	0.296	1.101	0.509	0.772	5.906
	polluted	0.007	0.961	1.166	0.312	0.559	13.816
Pb	control	0.0035	0.000	0.000	0.000	0.000	0.000
	polluted	0.0035	0.000	0.000	0.789	0.476	0.641
Zn	control	0.3	0.011	0.010	0.051	0.011	0.004
	polluted	0.3	0.009	0.008	0.009	0.048	0.009
Mg	control	0	-	-	-	-	-
	polluted	0	-	-	-	-	-
Cd	control	0.001	0.000	0.000	0.000	0.000	0.000
	polluted	0.001	0.000	0.000	0.230	0.173	0.115
Cr	control	0.003	0.038	0.000	0.000	0.038	0.000
	polluted	0.003	0.019	0.000	0.000	0.019	0.058
Hazard index	control	0.35	1.11	0.56	0.82	5.91	
Hazard index	polluted	0.99	1.17	1.34	1. 27	14.64	

BDL- Below detection limit of 0.0001 mg/kg.. Gongrenema latifolium.(GL), Ocimum gratissimum (OG), Vernonia amygdalina (VA), Talinium triangulare (TT), and soil sample(SS) in contaminated (Ogoniland) and control area (Umuchichi)

Table 5: Cancer Risk of Heavy metal in selected vegetables (mg/kg/day)

		CSF	GL	OG	VA	TT	SOIL
Fe	control	0	_	_	_	_	_
	polluted	0	_	_	_	_	_
Pb	control	0.0085	_	_	_	_	_
	polluted	0.0085	_	_	2.35E-05	1.42E-05	1.91E-05
Zn	control	0	_	_	_	_	_
	polluted	0	_	_	_	_	_
Mg	control	0	_	_	_	_	_
	polluted	0	_	_	_	_	_
Cd	control	0.38	_	_	_	_	_
	polluted	0.38	_	_	8.74E-05	6.56E-05	4.37E-05
Cr	control	0.5	5.75E-05	_	_	5.75E-05	
	polluted	0.5	2.88E-05	_	_	2.88E-05	8.63E-05

BDL- Below detection limit of 0.0001 mg/kg. CSF- Cancer Slope Factor. Gongrenema latifolium. GL, Ocimum gratissimum OG, Vernonia amygdalina VA, Talinium triangulare TT, and soil sample in contaminated (Ogoniland) and control area (Umuchichi)

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