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Full Length Research Paper

Trace metals bioaccumulation index and risk assessment in African giant land snail (*Archachatina margenata*) around University of Nigeria Nsukka sewage dumpsite, Enugu State Nigeria

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The study investigates some selected trace metals bioaccumulation index and risk assessment in African giant land snail (*Archachatina margenata*) around University of Nigeria sewage dumpsite. Soil samples were collected at 30 cm depth with the aid of soil auger and snail samples were collected from the University of Nigeria sewage dumpsite and Etana snail farms served as control. The soil samples were collected at random and their physicochemical properties such as pH, total nitrogen, total phosphorus, organic matter, total carbon and exchangeable cations (i.e., K^+, Mg^{2+} and Na^+) were analyzed using a standard method. The heavy metals in soils and vegetables, Al, As, Cd, Cr, Cu, Hg, and Pb were analyzed using Flame Atomic Absorption Spectrometer (FAAS). The risk assessment from the consumption of snails in the study area was calculated using standard methods and formulae. The result showed a significantly different ($p<0.05$) in soil physicochemical properties between soil from the sewage dumpsite and Etana farm soil, except K^+ which is not significant. The soil trace metal revealed that the mean concentrations of most of the trace metals were significantly ($p < 0.05$) higher at the sewage site compared to the control soil samples from the Etana snail farm. The concentration of trace metals (Al, As, Cd, Cr, Cu, Hg, and Pb) in snails from sewage dumpsite soil were 5.22, 3.85, 5.25, 3.42, 3.40, 3.60 and 2.88 mg/kg, while snails from Etana snail were 0.26, 0.33, 0.91, 0.28, 1.87, 0.06 and 0.11 mg/kg respectively. The concentration of trace metal in snail from sewage site were all greater than the maximum permissible limit of 0.5 (As), 2.0 (Cd), 0.3 (Cr), 0.04 (Cu), 3.60 (Hg) and 2.88 mg/kg (Pb) for snail given by FAO/WHO limits. The BAF of Cr, Cu, Hg, and Pb were greater than one (>1) in the test samples which indicates that snails are enriched with the trace metal from the soil (Bioaccumulators) while the control was all less than one. The result of DIM and HQ shows that sewage site snails have higher values of trace metals compared to the control farm. The Hazard Index (HI) shows that there is no harmful effect since the values obtained were below >1. But continuous consumption can accumulate in the food chain. This study showed that snails around the vicinity of sewage dumpsites were highly polluted with trace metals which can pose health risks.

Keywords: *Archachatina margenata*, bioaccumulation, sewage site, trace metals, risk assessment

INTRODUCTION

All human activities generate wastes and the way in which the wastes are handled, stored, collected and disposed, can pose risks to the environment and to public health (Zhu et al., 2008). Generally, most pollutants are introduced into the environment as solid waste, sewage and as compounds used to protect plants and animals.

For decades solid waste has been introduced into University of Nigeria dumpsites and it is suspected that this must have elicit pollution of the environment. Dumpsite consists of garbage, faeces, refuse, rubbish, dead animals, broken glasses, plastics, metals, food remnants, paper, wood, cloth etc., may either be biodegradable or non-biodegradable. Due to the multidimensional nature of waste and its negative effect on humans, animals, plants, microorganisms and environment, its management is difficult and required high effort. Snails being an important habitat in dumpsites due to the fact that decayed and composted wastes enhance soil fertility and increases nutrients to the snails, despites the important snails are readily exposed to heavy metals, which is bioaccumulated in human when consume through food chain (Nwachukwu et al., 2018, Oguh et al., 2019a). Snails thrive better in soils that are rich in organic matter. The decay of these solid wastes releases substances that can affect the soil, increase the concentration of heavy metals in the soil, altering the natural balance of nutrients available for snail's growth and development thereby affecting species diversity.

A. marginata (AM) belongs to the group Phylum *Mollusca* and Family *Achatinidae* belonging to the class Gastropoda (Nkop et al., 2016). Apart from insects, mollusca are the largest invertebrate group in the animal kingdom (Yoloye, 1994). *A. marginata* are bilaterally symmetrical invertebrates with soft segmented exoskeleton, inhabiting mostly marine environments, tolerating varied environmental conditions and thrive best in temperate and tropical areas, where soil pH ranges from 4.5-8.0 (Adediran et al., 2003). Organic manure and dead decay plant, and sewage soil ultimately maximize snail productivity and economic returns (Oguh et al., 2019a), but the side effects on snails are often neglected. *A. margenata* also known as Dodon kodi in Hausa, Igbin in Yoruba and Ejule in Igbo. Nutritionally, snails are of paramount important as source of high profile protein, low in fat and rich in iron food ideal for human nutrition especially for diabetic patients (Cobbinah, 1993, Awah, 2000). Snails serve as valuable sources of nutrition to human and animals with high levels of protein, iron, calcium, phosphorus and amino acid such as lysine, leucine, and arginine, relatively low amount of sodium, fat and cholesterol compared to poultry and other livestock (Wosu, 2003). Snail meat compares favourably with whole egg in all essential amino acids especially with regard to lysine, leucine, isoleucine and phenylalanine (Imebvore, 1990).

The term heavy metal refers to any metallic chemical element that has a relatively high density greater than 5 g/cm3 and is toxic or poisonous at low concentrations. Recent researchers have found that even low levels of mercury, cadmium, lead, aluminum and arsenic can cause a wide variety of health problems (Hassaan et al., 2016). Heavy metals toxicity can result in damaged or reduced mental and central nervous function, lower energy levels and damage to blood composition, lungs, kidneys, liver and other vital organs. Long-term exposure may result in slowly progressing physical, muscular and neurological degenerative processes that mimic Alzheimer's disease, Parkinson's disease, muscular dystrophy and multiple sclerosis. Allergies are not common and repeated long-term contact with some metals may cause cancer. Metals such as cadmium, mercury, arsenic and lead are non-essential and therefore have toxic effects on living organisms such as damage to the renal and nervous systems of fish as well as gill damage (severe destructive pathological changes, i.e. structural lesions) (Velcheva et al., 2010, Deore, and Wagh, 2012).

African giant land snail (AGLS) are often found in many locations and have a very diverse type of habitat especially dump and dead decay sites and this may lead to the bioaccumulation of metals in AGLS, which is a major food chain route for the human body. Snails are not able to hear at all, but rely on their sense of touch to interact with each other and use their sense of smell to help them find food. Snails are more active at night than day time and may come out during the early morning hours as well. Land snails are particularly well adapted to changes in moisture and dry conditions and are able to remain sealed within their thick shells for two or more years. Most snails have thousands of microscopic tooth-like structures located on a ribbon-like tongue called a radula used to cut food into small pieces. Many snails are herbivorous, eating plants or rasping algae from surfaces with their radula. Snail farming is a lucrative business in most part of the world especially in Nigeria. However while some farmers in Asia and Europe have developed a technology for soilless snail farming, in Nigeria and most part of Africa snails are raised mainly in soil medium. Many snails ingest small amount of soil particles and rasp larger rocks or snail shells in order to obtain the Ca essential to reproduction, shell development (snail shells are composed mostly of calcium carbonate $CaCO_3$), and other physiological needs. In times of Ca demand, such as egg laying, snails mobilize Ca from their own internal organs and shells (Fournie and Chetail, 1984).

The sewage treatment plant receives sewage from all the students' hostels, staff quarters, laboratory, office wastewater, and wastewater from the university clinic. Wastewater contains a lot of nutrients, which increases snail yields around the area. However, it contains a variety of chemical substances and microbiological loads from domestic and industrial sources (Oguh et al., 2019b). A wide range of microbial pathogens have been found in sewage water and can be transferred to snail in such area. Survival of pathogens in the water and surrounding environment is mainly dependent on factors such as nutrient availability, temperature, organic matter content, competition with other microorganisms, pH and radiation. One major cause of snail's contamination could be the unavailability of hygienic environment. Pathogens can be transmitted to snail and cause outbreaks of illnesses when these are consumed. Risk assessment are series of calculation to estimate the nature and probability of adverse health effects in humans who may be exposed to chemicals in contaminated environmental media especially heavy metals. Human health risk assessment is the process to estimate the nature and probability of adverse health effects in humans who may be exposed to chemicals in contaminated environmental media, now or in the future (United States Environmental Protection Agency (USEPA), 2017).

Dumpsites contains trace toxic elements such as Aluminum, Arsenic, Cadmium, chromium, Copper, lead, mercury, and parasitic worms, which can induce severe risks to the human health and the environment (Mark et al., 2017, Shahid, 2017). Soil in and around dumpsite is usually nutrient rich, which improve soil properties such as organic matter, and nutrients, which increases snails yields and production. Thus despite this important and yield benefits, outbreaks of human infections and environmental hazard are associated with the consumption of snails around sewage dumpsite. Hence, this study was designed to evaluate trace element risk assessment in African giant land snail (*Archachatina margenata*) around University of Nigeria sewage dumpsite**.** The results are expected to create awareness among the public on the safety of consuming snails around in such areas.

MATERIALS AND METHODS

Study area

The study was carried out in farm around sewage dumpsite at University of Nigeria, Nsukka local government area in South-East, Enugu State of Nigeria. Nsukka is situated at 6.86^{o} North latitude, 7.39^{o} East longitude, 456 meters elevation above the sea level and has an area of 1810km^{2}. The University of Nigeria, Nsukka lies between longitudes 7°24′E and 7°26′E, and longitudes 6°51′N and 6°53′N (Figure 1). The sewage plant site is located at the northeast, of the university and covers an area approximately 700 m^{2}. Etana Snail farms, Nsukka Enugu State (A private snail farm was also taken into considerations) with coordinates (6E52'25.1 N, 7E22'10.0 E).

Sample collections

Soil and snail samples were major test materials used in this study. The snail samples used for the study were picked early hours of the day around university of Nigeria, Nsukka sewage dump site. Snail samples were also collected from Etana snail farms at Nsukka, Enugu state which serves as control. Snails were collected from the top soil or by burrowing between 0-30 cm from the topsoil. Soil samples were collected at same sites with the help of soil auger. Soils were the topmost soil ranging from 0-30 cm from the top soil on the three sites. Snails and soil samples were randomly sampled within the areas to get a representative sample. All samples were collected aseptically in a sterilized universal container and plastic bags. Analysis was conducted within 24 h arrival at the Laboratory.

Experimental design

The experiment was carried out under a Completely Randomized Design (CRD) with three replicate groups for each. The concentrations of the heavy metals both in dumpsite soils and snail sample, were done in two groups, from group 1 to 2, which are snail samples from dump site, and a control site (a snail private farm). Both soil and snail sample were randomly collected and analyzed for heavy metals (Al, As, Cd, Cr, Cu, Hg and Pb).

Preparation of Snail sample

The snail samples were sacrificed by striking with a wooden material on the shell carefully. The flesh/foot of the snail was carefully removed from the shell and washed with adequate distilled de-ionized water, then dried with a stainless plate in an oven at 115^{o}C to constant weight in three days. After drying, samples were crushed to fine powder using porcelain mortar and pestle, then sieved using a 0.4 mm mesh. The powdered samples were stored in 100 mL air tight bottles prior to digestion/analysis as describe by (Oguh et al., 2019a).

Determination of the physicochemical properties

The physicochemical properties measured were soil texture, pH, TN, TOC, OM, TP, and EI (Na^{+}, Mg^{2+} and K^{+}). The Physico-chemical properties of the soil were analysed in order to evaluate the biodegradable process. Physicochemical parameters of the contaminated soil and the control soil samples were determined according to (Nimyel et al., 2015).

Soil pH

Triplicate quantities of air-dried (20 g) of the soil samples were weighed into two separate groups of six 50 mL beaker and 20 mL of distilled water was added to one group and 30 ml of 1M KCl_2 was added to the other group. The mixtures were allowed to stand for 30 min

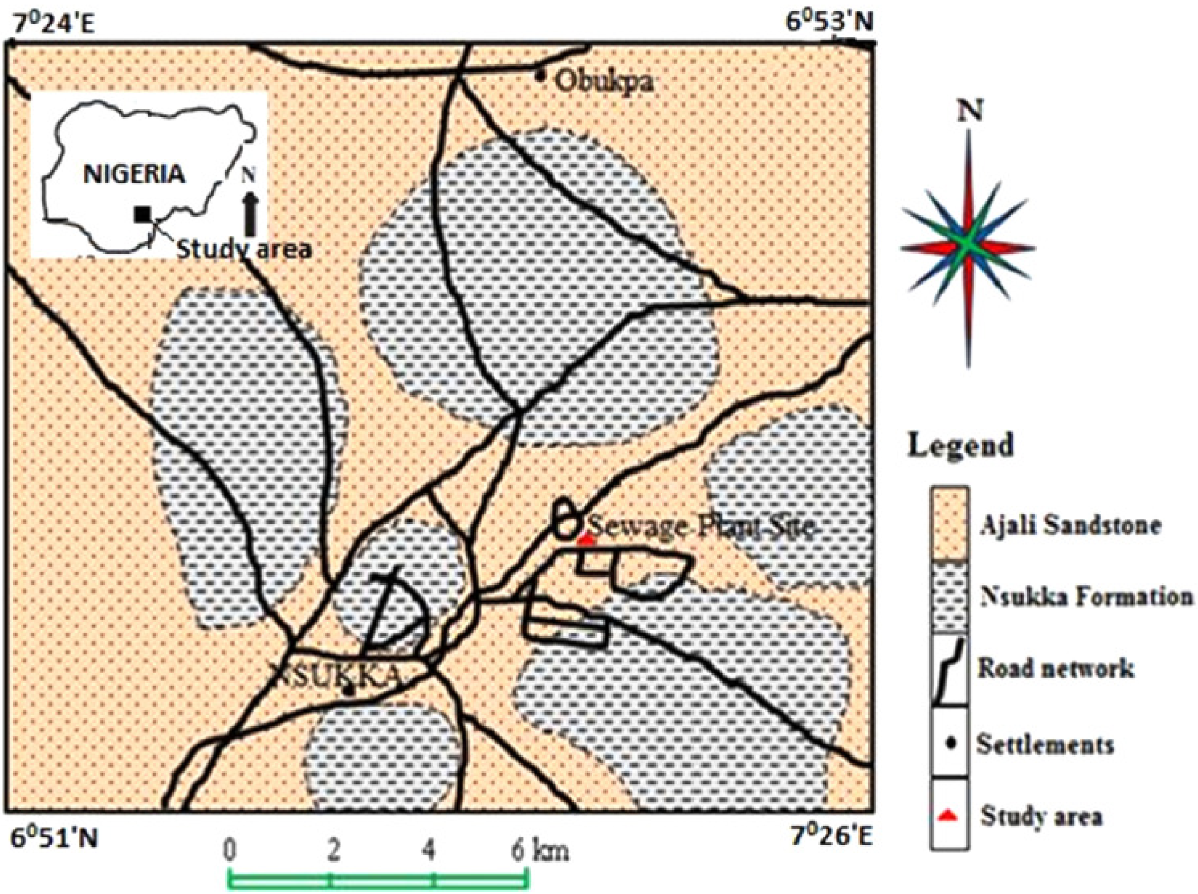

Figure 1. Map of the study area.

with occasional stirring using a glass rod. The electrode of the calibrated pH meter, MI 806 pH/EC/Temperature Portable Meter was inserted into mixtures and the pH value was read using pH meter. The results were reported as soil pH in 1M KCl_2 and soil pH in water (H_20). Three readings were recorded and then mean of it was calculated.

Organic matter

The soil samples were grounded to pass through 0.5mm sieve after which 1g was weighed in triplicate and transferred to 250 mL Erlenmeyer flasks. Exactly 10 mL of 1M potassium dichromate was pipetted into each flask and swirled gently to disperse the soil. Then 20 mL of concentrated, tetraoxosulphate (IV) acid was added. The flask was swirled gently until soil and reagents were thoroughly mixed. The mixture was then allowed to stand for 30 minutes on a glass plate to allow for the oxidation of potassium dichromate to chromic acid. Distilled water (100 mL) was added then 3- 4 drops of ferroin indicator or 1ml of diphenylamine indicator was added, after which the mixture was titrated with 0.5 M ferrous sulphate solution or ferrous ammonium sulphate till the colour flashes from blue-violet to green or bright green. A blank titration was similarly carried out. The percentage of organic matter is given by the following equation:

% organic matter = (M1V1$K_2Cr_2O_7$ – M2V2$FeSO_4$) × 0.0031 × 100 × F/Mass(g) of air dried soil
F = correction factor (1.33), M1 = mole of $K_2Cr_2O_7$, V1 = volume of $K_2Cr_2O_7$, M2 = mole of $FeSO_4$, V2 = volume of $FeSO_4$.

Total organic carbon

Exactly 1g soil sample was accurately weighed into a 500 mL conical flask. Then 10 mL of 1M potassium dichromate ($K_2Cr_2O_7$) solution was added to the sample using a bulb pipette, followed by 20 mL of concentrated sulphuric acid (H_2SO_4) while gently swirling the flask in a fume cupboard. It was allowed to stand and cool slowly on insulated pad like sheet of asbestos for about 25 min after which 200 mL of distilled water was added using a measuring cylinder. After this, 1 g of crystal sodium fluoride (NaF) was added to avoid interference by complexing $Fe^{3+,}$ obtaining a black colour mixture which was shaken vigorously. Finally, 1 mL of 1 % diphenylamine was added as an indicator and the mixtures were titrated immediately with 1 M ferrous sulphate ($FeSO_4$) solution in the burette. A blank without soil was prepared alongside the sample and titrated same way. End point was indicated a colour change from deep purple to green.

% Total Carbon content = (B-T)×M×0.003×100×1.33/weight of soil sample taken.
Where; B = Blank titre, T = Test sample titre, M = Molarity of $FeSO_4$, 1.33 = Correction factor, 0.003 = mg equivalent of carbon.

Total phosphorus

Air-dried soil 2 g was weighed and dispensed in 20 ml of (0.025N HCl + 0.03N NH_4F) solution, shaken for 5 min and then filtered using whatman filter paper. After filtration, 3ml of the clear filtrate was put into a test tube, and 3 ml of (0.87N HCl, 0.38N ammonium molybdate, and 0.05% H_3BO_3) solution and 5 drops of (2.5g of 1–amino 2- tetraoxosulphsate (vi) acid, 5.0g Na_2SO_3, 146 g $Na_2S_2O_5$) solution were sequentially added to the prepared clear sample. A colorimeter (at wave length of 660 nm) was then used to take readings.

Total nitrogen

Total nitrogen was determined using the kjeldahl digestion method. Than 20 ml of concentrated tetraoxosulphate (VI) acid was added to a 1g measurement of air dried soil. A catalyst known as Kjeldahl TAB was also added and the solution was digested. After digestion, a clear solution was observed; this clear solution was distilled and subsequently titrated with 0.01M HCL.

Sodium, Magnesium and Potassium ion (Na^+ $Mg^{2+}$and K^+)

The exchange ions were determined calorimetrically using Flame photometer. Soil sample (5 g) was accurately weighed into No. 1 filter paper fitted into a funnel on a leaching rack with 100 mL volumetric flask for collecting the leachate. The soil sample was leached with 1 N NH_4OAC solution obtaining 100 mL volume of leachate. The Optical density readings for Na^+ Mg^{2+} and K^+ were obtained from the flame photometer.
$Na^+/mg^{2+}/K^+$ meq/100g = Optical density× correction factor×100/5

Determination of heavy metal

The soil samples was spread on stainless plates and then dried in an oven at 105°C for six hours. The dried soil was grounded and sieved through 0 – 5 cm mesh sieve. Exactly 1.0 g of the dried powders sample (soil and snail sample) were weighed accurately into a 50 ml beakers separately, to which 15 ml of tri-acid mixture (70% high purity HNO_3, 65%, $HClO_4$ and 70% H_2SO_4 in 5:1:1 ratio) were added. The mixture was digested at 80°C till the solution became transparent. The resulting solution were filtered and diluted to 50 ml using deionized water and analyzed for Al, As, Cd, Cr, Cu, Hg and Pb, by flame atomic absorption spectrophotometry describe by Oguh et al. (2019a).

Bioaccumulation factor (BAF)

The transfer factor was estimated by dividing the concentration of heavy metals in snail by the total heavy metals concentration in the soil. This index of soil to snail transfer or intake of elements from soil through snail was calculated using formula describe by (Adediran et al., 2003):
$BAF = C_{snail}/C_{soil}$

Where

BAF represent the transfer factor of snail or Bioaccumulation factor
C_{snail} = Concentration of metals in snail tissue, mg/kg fresh weight
C_{soil} = Concentration of metals in soil, mg/kg dry weight.
BAF > 1 indicates that the snails are much enriched with the elements from the soil
BAF < 1 shows that the snail are not enriched with the toxic elements from soil

Estimation of health risk assessment

Daily intake of metal (DIM)

The Daily intake of metals was calculated using the following formula describe by (USEPA, 2002).

$DIM = DI \times M_{snail}/WB$

Where

DIM = represents the average daily dose (mg,kg/d) of the metal
DI = represent the daily intake of snail for adult is 0.10274 kg/person/day
M_{snail} = Trace metals concentration in the snail tissues (mg/kg)
WB = Average body weight of adult (70kg for adults).

Hazard of metal to human (hazard quotient HQ)

The hazard quotient (HQ) was used to calculate the possible human health risks associated with the consumption of snail from contaminated soils. The following equation is used to calculate the Hazard Quotient of snail describe by (USEPA, 2017).
HQ is the ratio between exposure and the reference oral dose (RFD)
If the ratio is lower than one (1), there will be no obvious risk.
HQ = DIM/ RFDM

Where

DIM = represents the average daily dose (mg,kg/d) of the metal
RFDM = is the reference dose of the metal (mg,kg/d), which is define as the maximum tolerable daily intake of metal with no adverse effect

Hazard index (HI)

The hazard index (HI) was calculated to determine the overall health risk of exposure to all the heavy metals via the ingestion of a particular snail describe by (USEPA, 2002). The hazard index (HI) was calculated as the summation of the hazard quotient (HQ) arising from all the metals examined. The value of the hazard index is proportional to the magnitude of the toxicity of the snail consumed. HI > 1 indicates that the predicted exposure is likely to pose potential health risks. However, a hazard index <1 does not necessarily indicate that a potential adverse health effects will result, but only indicates a high probability of posing health risks.

$$HI = \sum HQ_{As} + HQ_{Cd} + HQ_{Cr} + HQ_{Cu} + HQ_{Hg} + HQ_{Pb}$$

Statistical analysis

The data obtained were analyzed using IBM Statistical Product and Service Solution (SPSS) version 20 and Microsoft excel 2013. The results were expressed as mean ± standard error (SE). One way analysis of variance (ANOVA) was carried out as $p<0.05$ considered statistically significant.

RESULTS

Physicochemical properties of sewage soil

The physicochemical properties of the soil were presented in (Table 1). The texture of the sewage site soil is loamy soil because of the decaying organic matter which contains more nutrients and humus and control soil is sandy loam. The pH of the soil in H_2O and KCl_2 were (5.64 and 7.78) and (6.04 and 8.01) in sewage site and control site respectively. The nitrogen, organic matter, organic carbon, total phosphorus, and exchangeable cation (K^+, Mg^{2+}, and Na^+) of sewage dump and control soil were 3.81, 34.18, 4.13, 10.51, (2.60, 4.24 and 6.73) and 2.06, 12.43, 1.82, 11.47 (2.54, 6.89 and 5.24) respectively.

Trace metal concentration in soils samples

The mean concentrations (mg kg-1) of trace metals were presented in (Table 2). The concentrations of Cd and Hg in sewage site soil were 5.29 and 3.46 mg/kg which were above the (WHO/FAO, 2001) limits of 3.0 mg/kg Cd and 2.0 mg/kg Hg of metal in soil. But the control soil recorded a value that was below the permissible limit. The concentrations of Al, As, Cr, Cu, and Pb in the sewage site were 5.24, 4.09, 2.88, 3.20 and 2.83 mg/kg while control site were 0.43, 1.11, 2.01, 2.55 and 0.27 mg/kg respectively which were all below the WHO/FAO, 2001 maximum limit of 20 (As), 100 (Cr), 100 (Cu) and 50 mg/kg (Pb) for soil. It revealed that the mean concentrations of most of the trace metals were significantly ($p < 0.05$) higher at the sewage site compared to the control soil samples from Etana snail farm (Table 2).

Trace metal concentration in snail samples

The mean concentration of trace metals in the snail indicates bioaccumulation from the soil. The concentration of trace metals (Al, As, Cd, Cr, Cu, Hg, and Pb) in snails from sewage dump site soil were 5.22, 3.85, 5.25, 3.42, 3.40, 3.60 and 2.88 mg/kg, while snails from Etana snail were 0.26, 0.33, 0.91, 0.28, 1.87, 0.06 and 0.11 mg/kg respectively. The concentration of trace metal in snail from sewage site were all greater than the maximum permissible limit of 0.5 (As), 2.0 (Cd), 0.3 (Cr), 0.04 (Cu), 3.60 (Hg) and 2.88 mg/kg (Pb) for snail given by (WHO/FAO, 2013; WHO/FAO, 2016). Etana snail farm snails were within the maximum permissible limit standard as shown on (Table 3). The result shows that snails from sewage dump site were contaminated with trace metals compared to the control.

Bioaccumulation factor (BAF) of trace metal to snail

The bioaccumulation factor (BAF) of trace metals gives the ratio of the concentration of metals in snail to the total concentration in the soil. The amount of elements Al, As, Cd, Cr, Cu, Hg, and Pb that is transferred from soil to snail in sewage site snails were 0.99, 0.94, 0.99, 1.18, 1.06, 1.04, and 1.01, snail from Etana farms were 0.60, 0.29, 0.45, 0.13, 0.73, 0.54 and 0.40 respectively.

Table 1. Physicochemical properties of soils samples.

Soil Properties	Physicochemical properties	
	Sewage site soil	Etana farms soil (Control)
pH H_2O	5.64 ± 0.10	7.78 ± 0.15
pH KCl	6.04 ± 0.16	8.01 ± 0.07
TN %	3.81 ± 0.10	2.06 ± 0.08
TP %	10.51 ± 0.09	11.47 ± 0.13
TOC %	4.13 ± 0.11	1.82 ± 0.07
OM %	34.18 ± 0.03	12.43 ± 0.10
K^+ meq/100g	2.60 ± 0.07	2.54 ± 0.05
Mg^{2+} meq/100g	4.24 ± 0.12	6.89 ± 0.07
Na^+ meq/100g	6.73 ± 0.08	5.24 ± 0.04
Texture	loamy	Sandy loam

Results expressed as Mean ± SD. n=3

Table 2. Concentration of trace metals in soil.

Elements (mg/kg)	Trace metals in soil		
	Sewage site soil	Etana farms soil (Control)	PL(mg/kg) in soil by (21)
Al	5.24 ± 0.07	0.43 ± 0.07	-
As	4.09 ± 0.03	1.11 ± 0.03	20
Cd	5.29 ± 0.06	1.98 ± 0.05	3.0
Cr	2.88 ± 0.08	2.01 ± 0.09	100
Cu	3.20 ± 0.04	2.55 ± 0.06	100
Hg	3.46 ± 0.10	0.11 ± 0.03	2.0
Pb	2.83 ± 0.10	0.27 ± 0.08	50

Results expressed as Mean ± SD. n=3

Table 3. Concentration of trace metals in snail.

Elements (mg/kg)	Trace metals in Snail		
	Sewage site snail	Etana farms snail (Control)	PL(mg/kg) snail FAO/ WHO, (22)**, (23)*
Al	5.22 ± 0.05	0.26 ± 0.04	-
As	3.85 ± 0.13	0.33 ± 0.09	0.5*
Cd	5.25 ± 0.07	0.91 ± 0.05	2.0*
Cr	3.42 ± 0.10	0.28 ± 0.01	0.3*
Cu	3.40 ± 0.07	1.87 ± 0.04	0.04**
Hg	3.60 ± 0.09	0.06 ± 0.01	0.1*
Pb	2.88 ± 0.08	0.11 ± 0.03	0.1*

Results expressed as Mean ± SD. n=3

The BAF of Cr, Cu, Hg, and Pb were greater than one in the test samples while the control were all less than one. Where the BAF is greater than one (>1) indicates that the snails are enriched with the elements from the soil (Bioaccumulators). Also where BAF is less than one (<1) means that the snails are not much enriched with the trace metals from the soil as shown in (Figure 2).

Estimation of health risk assessment

Daily intake of metal (DIM) for adult

The DIM for adult in snail was estimated according to the average snail consumption via food chain. The DIM values for trace metals (Al, As, Cd, Cr, Cu, Hg, and Pb) in sewage site (0.008, 0.006, 0.008, 0.005, 0.005, 0.006 and 0.004), Etana farm (0.0004, 0.0005, 0.0015, 0.0004, 0.0032, 0.0001 and 0.00001) respectively for trace metals. The result shows that sewage site snails have higher values of DIM (Figure 3).

Hazard quotient (HQ) of trace metals

The HQ of trace metals through consumption of snails from sewage dump sites were given in (Figure 4). The HQ of trace metals Al, As, Cd, Cr, Cu, Hg, and Pb from snails in sewage dump site were 0.008, 0.0.012, 0.004, 0.016, 0.125, 0.06 and 0.04 and snails from Etana farms were 0.0004, 0.001, 0.0007, 0.0013, 0.08, 0.001 and 0.001

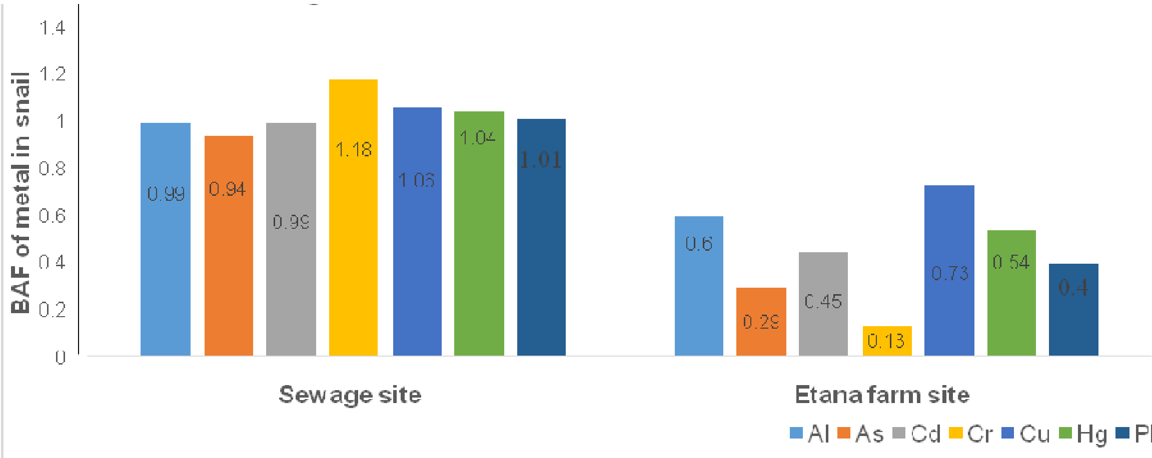

Figure 2. Bioaccumulation factor of metals in snail.

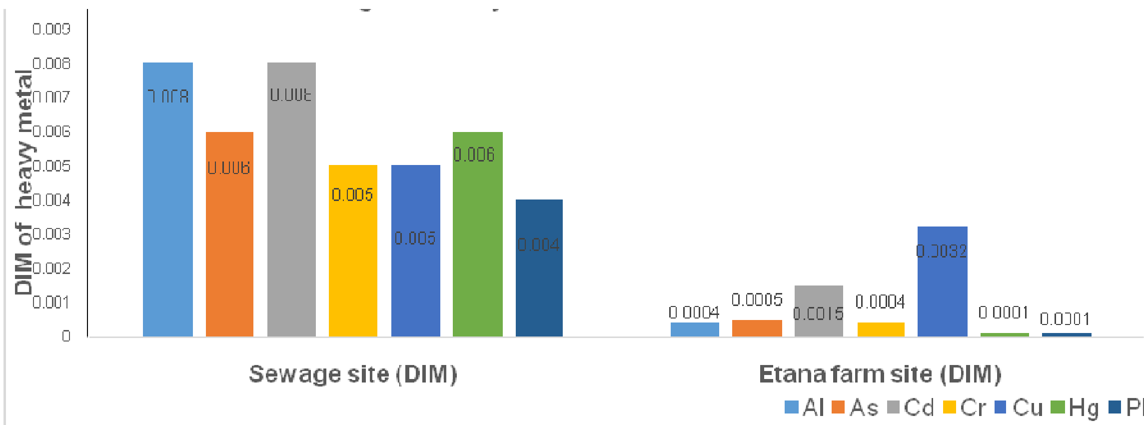

Figure 3. Daily intake of metals from snail.

respectively. The HQ for snails from sewage site were high than the control snail (Figure 4).

Hazard index (HI) of trace metals

The estimated HI in sewage site and control site were represented in (Figure 5). The HI shows the overall hazard risk of snail when consume. The total values of trace metals estimated in sewage dump site and Etana farm (control) were 0.265, and 0.085 respectively. However, the HI were all below one <1 which does not necessarily indicate that a potential adverse health effects will result, but only indicates a high probability of posing health risks.

DISCUSSION

The low pH value recorded in H_2O and KCl_2 (5.64, 6.04) at sewage site were as a result of the sewage waste materials, and high content of organic matter which moves toward acidity. The pH values in sewage site was below the recommended target limits (6.5 - 8.5) for agriculture (WHO-World Health Organization, 2006), while the control site is within the recommended limits. It has shown that heavy metals are more mobile at pH < 7 than at pH > 7 which has influence in heavy metal accumulation (Fonge et al., 2017, Oguh et al., 2019c). In connection with this findings, (Oguh et al., 2019c) also reported low pH in water (5.19) in sewage dump site. However, low pH can pose a significant health risk, leading to various chronic diseases, particularly in increased concentrations or in prolonged dietary intakes (Tirima et al., 2016). Solubility of trace metals tends to increase at lower pH value and decrease at higher pH values. The pH values obtained in this study are similar to that reported for dumpsites by other researchers (Elaigwu et al., 2007, Uba et al., 2008). The high total nitrogen in the sewage site soil is as a result of the waste nitrogenous substances such as decay plant and animals materials in the sewage. The results reported by (Osazee

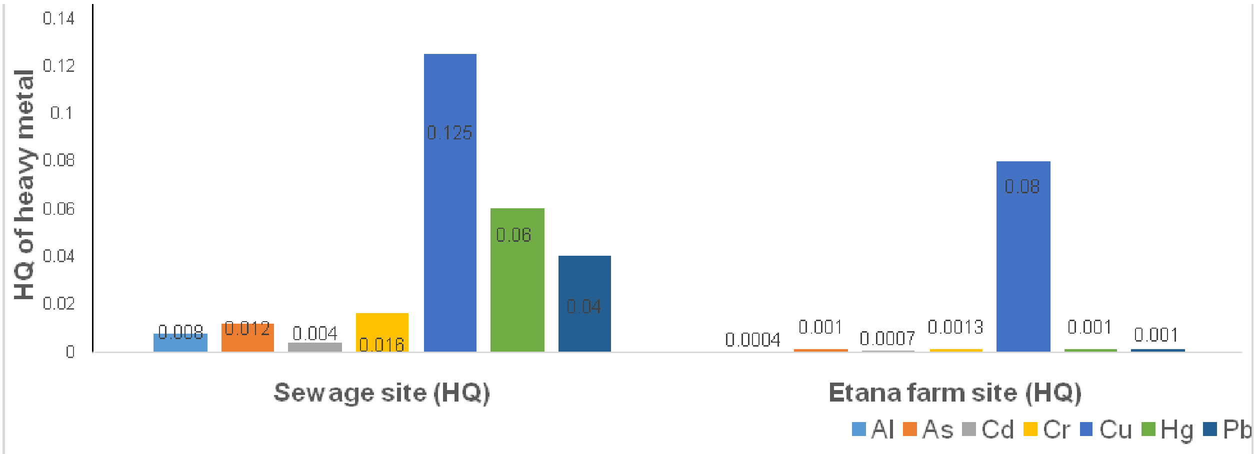

Figure 4. Hazard quotient of trace metals.

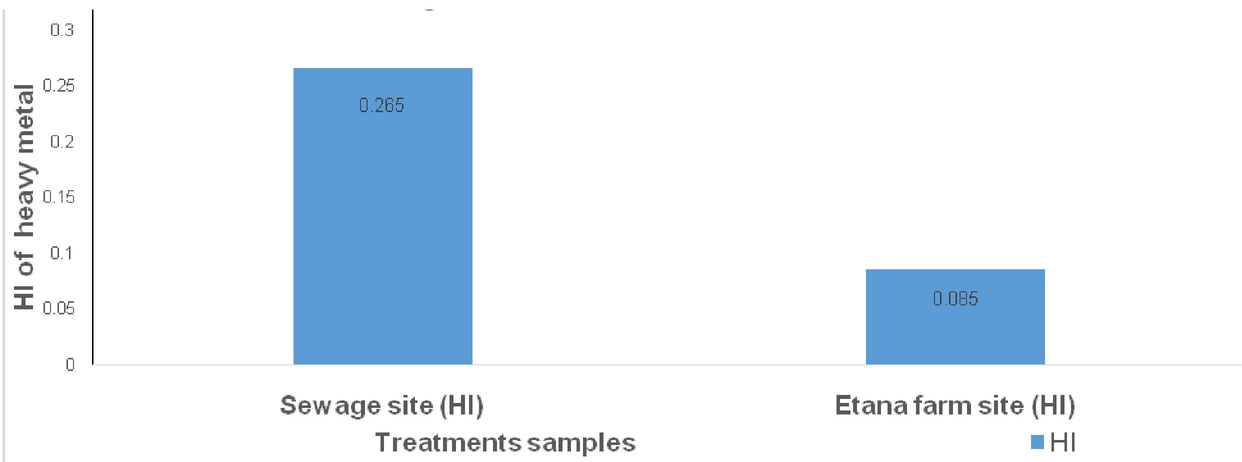

Figure 5. Estimation of hazard index.

et al., 2013) had the range 3.476 to 4.522 % which is within range of the present research with 3.81 in sewage site. Organic matter and total organic carbon in the sewage site soil were higher than that of the control sample. Decrease in total phosphorus content in the sewage site soil can be attributed to low phosphorus content in the sewage dump site areas. The increase of cations exchange capacity is due to the presence of clay content and increase in organic matter. The Analysis of variance (ANOVA) carried out shows significant differences ($p < 0.05$) in the sewage site soil and control soil (Table 1). The result of physicochemical carried out on the sewage soil changes the properties of soil, especially the sewage site soils. The alteration of the physicochemical properties of the soil is therefore expected to affect the survival of certain species of animal such as snails which depends on soil for survival and hence their diversity (Table 1). As a result of growing human activities, heavy metals in soils have become an alarming threat to ecosystem, animals and human health. The results of the soils showed that the concentration of trace metals in the soil increased at the sewage site than the control sites for all the trace metals (Al, As, Cd, Cr, Cu, Hg, and Pb). The concentration of Cd and Hg in sewage sites soil (5.29, and 3.46 mg/kg) respectively were above the WHO/FAO, (2001) permissible limits of 3.0 mg/kg (Cd) and 2.0 mg/kg (Hg) for soil except for control soil with mean value below the permissible limit. The variations increase could be attributed to the nature, composition and amount of Cd and Hg containing wastes disposed of in the sewage site. The mean concentrations of Al, As, Cr, Cu and Pb recorded were below the WHO/FAO, (2001) maximum permissible limit of 20 (As), 100 (Cr), 100 (Cu), and 50 (Pb) mg/kg respectively for soil. This result conforms to the findings of (Oguh et al., 2019c) who also reported an increase of heavy metals on sewage dumpsite. This was in agreement with the findings of (Tanee and Eshalomi-Mario, 2015) results which showed that the concentration of heavy metals in the soil increased at the dumpsites than the control sites for all the metals (Fe, Pb, Cd and Zn). The study revealed that the concentrations of all trace elements analyzed were significantly (P < 0.05) higher at the sewage sites compared to the control soil samples (Table 2). The apparent increase of trace metals concentration in sewage site compared to the control site certainly confirms the sewage dump site waste as the potential source of soil contamination and their accumulation in snail (Table 2). Trace metals are considered the most important constituents of environmental pollution from the terrestrial environment due to toxicity and accumulation by land organisms, such as snails. The entire snail samples from sewage site and control site contained detectable levels of the trace elements studied. The accumulation of these metals in snails may represent a health risk, especially for populations with high consumption rates of snails. Results One-way Analysis of variance (ANOVA) revealed a significant (P<0.05) variation in the concentrations of trace metals in sewage site and that of the controls site, which is an indication of the extent of metal pollution from the soils. Generally sewage site had higher heavy metals concentrations (Al, As, Cd, Cr, Cu, Hg, and Pb) than the controls, which were all above the FAO/WHO, (2013) and FAO/WHO, (2013) permissible limit of 0.5 (As), 2.0 (Cd), 0.3 (Cr), 0.04 (Cu), 0.1 (Hg) and 0.1 mg/kg (Pb) (Table 3). The mean values recorded at all control sites were below the FAO/WHO acceptable value except Cu which is above the WHO/FAO, (2013) permissible limit of 0.04 mg/kg for snails. Trace metals and nutrients absorbed by snails are usually translocated to different parts of the snail such as tissue which could limit the concentrations in the soil. However, availability of metals in the soil and continuous absorption by the snail could lead to higher concentration in the snail which is transfer to human when consume. The ability of snails to accumulate essential metals equally enables them to acquire other nonessential metals from the soil. High concentration of Al may be due to high content of Al in the sewage waste. Aluminium toxicity is one of the major factors that limit plant growth, effect animals and development in many acid soils. However, strong interaction of Al^{3+}, the main Al toxic form, with oxygen donor ligands (proteins, nucleic acids, polysaccharides) results in the inhibition of cell division, cell extension, and transport. Aluminium have some problems to plant and animals at levels greater than 0.5 ppm. High concentration of As and Cd at sewage site may be due atmospheric deposition of the metal from non-ferrous metal activities, combustion, etc. which is absorbed into foliage and translocated through the snail. The levels of As recorded in this study was however much higher than the values of 0.21 to 0.81 mg/kg reported for snail by (Eneji et al., 2016).

Arsenic affects almost all organs during its acute or chronic exposure. Liver has been reported as target organ of arsenic toxicity. Toxicity is due to arsenic's effect on many cell enzymes, which affect metabolism, DNA repair and brain problem. The most prominent chronic manifestations of As involve the skin, lungs, liver and blood systems. The result of this study were higher than that of (Adamou et al., 2018) who recorded the average cadmium concentrations ranged from 0 to 0.032 mg/kg while those from lead ranged from 0.047 to 0.342 mg/kg in ALGS. Adedeji et al. (2011) recorded a concentration of 0.01 mg/kg Cd in snail from Alaro River within Oluyole industrial area in Ibadan Nigeria. Cadmium is a dangerous element because it can be absorbed via the alimentary track; penetrate through placenta during pregnancy and damage membrane and DNA. Significant concentration of Cd may have gastrointestinal effect, reproductive effect on livestock and causes both acute and chronic poisoning, adverse effect on kidney, liver, vascular and the immune system (Maobe et al., 2012). The concentration of Cr in this study was lower than (Eneji et al., 2016) who recorded level of Cr in all the sampling areas ranged from 40.8 to 857 mg/kg in snail. High dose of chromium is observed to cause Bronchopneumonia, chronic bronchitis, diarrhea, emphysema, headache, irritation of the skin, itching of respiratory tract, liver diseases, lung cancer, nausea, renal failure, reproductive toxicity, and vomiting. Copper is indeed essential, but in high doses it can cause anaemia, diarrhea, headache, metabolic disorders, nausea, vomiting, liver and kidney damage, stomach and intestinal irritation on human health. According to (Maobe et al., 2012) high levels of copper can cause metal fumes fever with flu-like symptoms, hair and skin decolouration, dermatitis, irritation of the upper respiratory tract, metallic taste in the mouth and nose. High level of Pb symptoms include blindness, deafness, brain damage, digestive problems, kidney damage, lack of coordination and mental retardation. Trace metal Pb has no beneficial biological function and is known to accumulate in the body and causes both acute and chronic poisoning and thus, poses adverse effects on kidney, liver, vascular and immune system. An increase concentration of Pb can cause serious injury to the brain, nervous system, red blood cells, low IQ, impaired development, shortened attention span, hyperactivity, mental deterioration, decreased reaction time, loss of memory, reduced fertility, renal system damage, nausea, insomnia, anorexia, and weakness of the joints when exposed to high Pb. The level of occurrence of the trace metal in sewage site increased in the order Pb < Cu < Cr < Hg < As < Al < Cd in (Table 3). The bio-accumulated trace metals on the snail may interact directly with biomolecules such as nucleic acid, protein, carbohydrate, disrupting critical biological processes, resulting in toxicity and the concomitant transfer of these metals through the food chain could ultimately pose risk to human life.

The soil-snail BAF is one of the key components of human exposure to toxic trace metals through the food chain. The BAF values decreased with the increasing respective total metal concentrations in the soils, indicating an inverse relationship between BAF and total metal concentration. The BAF in sewage site were significantly different from the control which indicates the high presence of metals in the sewage site. The higher the metals in the soil the more the bioaccumulation on the snail. The soil-snail BAF in sewage site for Cr, Cu, Hg, and Pb were greater than one (>1) which may be due to high levels of trace metals in soil and indicate that snails take up more of this metals from the contaminated soil. The control samples were all below one (<1) which indicate that the snail do not take up toxic element from the soil shown in (Figure 1). The hazard quotient (HQ) values were significantly <1; indicating that consumers of snails from these sites are not exposed health risk of the analyzed trace metal. The Hazard index (HI) shows the overall risk of exposure to all the metal in sewage site and the control site were below <1 which indicate that the consumption of snail in such environment cannot pose health risk to consumer. This finding shows that DIM, HQ, HI on the consumption of snail from sewage areas is nearly free of risks, but continuous consumption can lead to bioaccumulation in the food chain.

Conclusion

The present study revealed that African giant land snail tissue bioaccumulate significantly high levels of trace metals from contaminated sewage dump area, which exceeds the maximum prescribed limits for elements in snails, which toxicity disrupts natural ecosystems and affects the food chain, leading to deleterious health problems in humans and animals.

Author's declaration

We declared that this study is an original research by our research team and we agree to publish it in the journal.

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