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Levels of toxic metal in *Achatina achatina* from parts of Akwa Ibom State, Nigeria

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Abstract: Tropical land snail (*Achatina achatina*) were collected from three areas viz. Nsit Ibom Local Government Area (NTB), Nsit Ubium Local Government Area (NTU) and Uyo Municipality (UYM), all in Akwa Ibom State to determine the levels of Ni, Pb, Zn and Cr in their shells and muscles. Generally, the levels of all the metals in the muscles were comparatively higher than that in the shells. Correspondingly, the metals appear to have been more stable in the muscles with comparatively lower coefficients of variation than in the shells. Moreover while there was no significant correlation between the levels of all the metals in shells and muscles of NTU samples, Pb and Zn in NTB correlated very significantly. The correlation between levels of Cr in NTB samples and of Zn in UYM samples in shells and muscles were equally significant. On the whole, the levels of these metals were found to be much lower in both shells and muscles of NTU samples obtained from the "enclosed and restricted" environment than in NTB and UYM samples collected randomly from "open and unrestricted" environments.

Key words: levels of toxic; *Achatina achatina*; Nigeria

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Introduction

Snails are typical univalves that belong to the phylum mollusca. They are either aquatic or terrestrial and the common terrestrial species known include *Helix aspersa* and *Helix pomatia* (Hickman, 1970; Laskowski, 1996). In Akwa Ibom State of Nigeria, however, the common edible species is known as *Achatina achatina*. This species is generally creeping and sluggish and in addition to browsing on vegetation often burrows into sand and mud. It is usually found in large numbers at night at/or near waste dump sites, plantations, covered farmlands and woody debris. Hence it often gets exposed to contamination by trace metals and other toxic substances.

The toxic metals Ni, Pb, Zn and Cr can enter the terrestrial environment via. natural processes such as rock and soil weathering as well as geothermal reactions. Therefore these metals are often found to be natural components in tissues of organisms. They could also be introduced into the environment anthropogenically through such industrial processes and other activities. All the above activities contribute directly or indirectly to levels of toxic metals in the environment and this in turn influence the levels of these metals in tissues of living things.

Studies with metals indicated that food and other particles are significant sources of toxic metals (Penthreath, 1973). Thus, *Achatina achatina*, a common edible species of snail in Southeastern zone of Nigeria just like many other univalves are capable of accumulating toxic metals from terrestrial environments. According to Jorhem and Sundstrom (Jorhem, 1993), diet is the main source of human exposure to toxic elements, and so the levels of these elements in basic foodstuffs are of great interest from toxicological and nutritional points of view. Although some studies have been conducted on metal levels in some univalves, there appears to be no information on levels of Ni, Pb, Zn and Cr in the tissues of *Achatina achatina* (a major protein supplement) prevalent in this part of Nigeria. Considering the present tight economic position of Nigeria and the determination by many citizens to survive, a good percentage of the populace will continue to resort to the cheap means of balancing their diets until the situation is reversed. In Akwa Ibom State, for instance, one of such cheap means is the practice of hunting for snails by grown up children at night during which time the animal is always available in large numbers. This investigation therefore became necessary to find out how safe for consumption (with reference to toxic metal content) are snails often collected randomly from "open and unrestricted" environments.

It is hoped that the findings from this study would be of benefit to snail and other animal farmers, consumers, environmental protection officials and the general public.

1 Materials and method

The samples were collected during the early part of the rainy season in 1997 from three areas: Nsit Ibom Local Government Area (NTB), Nsit Ubium Local Government Area (NTU) and Uyo Municipality (UYM), all in Akwa Ibom State, Nigeria (Fig. 1). Samples from NTB and UYM sites were collected randomly from different location while all sample from NTU site were obtained from a snail farm in the area. Samples collected were usually put into clean, labelled plastic containers and taken to the University's Central Research Laboratory for preservation. Altogether ten samples per site were produced for treatment and analysis.

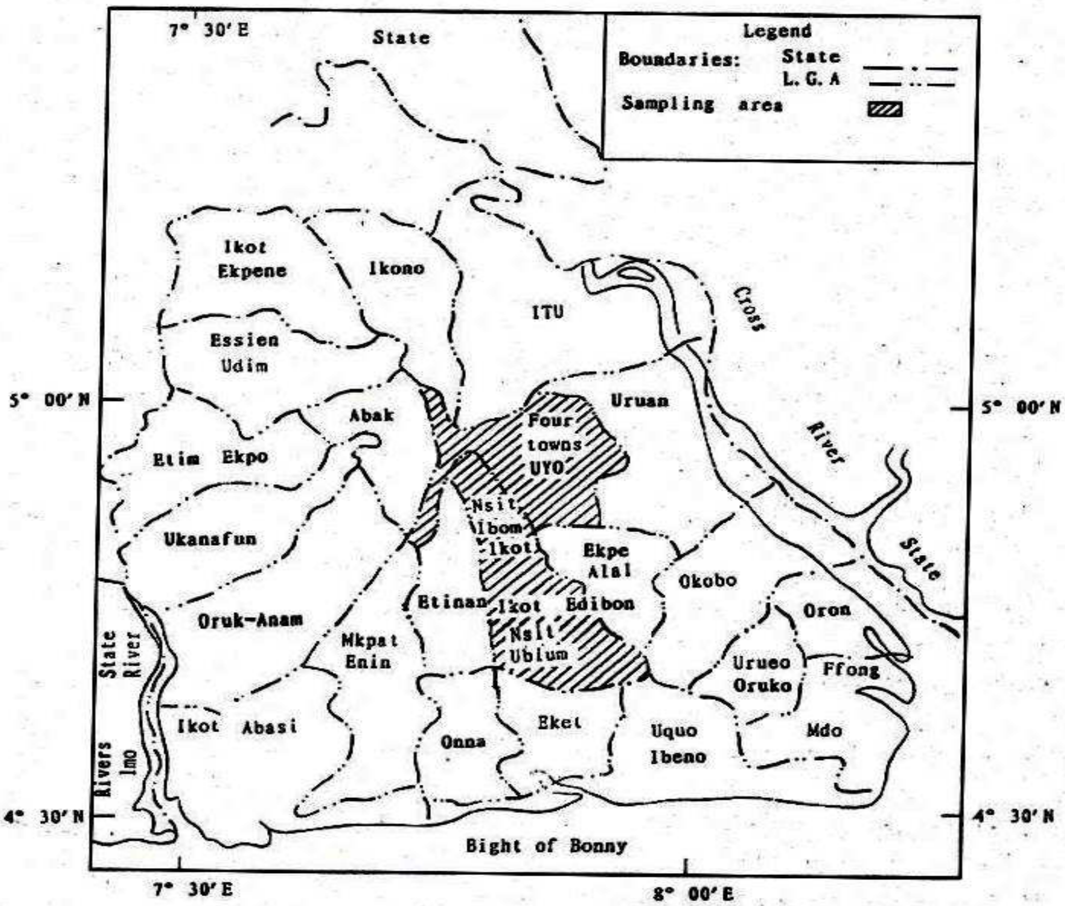

Fig. 1 Akwa Ibom State showing sampling area

After washing the samples thoroughly, the muscles were carefully removed from the shells. These were then separately dried to constant weights in an oven at a temperature of 50℃ for 48 hours. All dried samples were later milled into powder using an electric blender and stored in an appropriately labelled containers for further use.

Each sample (1.000 g) was weighed into a dry pyrex beaker and placed in a muffle furnace initially set at 200℃ to char; and then at 500℃ until the ash was devoid of traces of carbon (A.O. A.C., 1975). The ash was then dissolved in 6 mol/L HCl solution (20 cm^3) and heated on a steam bath until the volume was reduced to about one third. This was quantitatively transferred to a 25 cm^3 volumetric flask and the solution made up to the mark with ultrapure water before aspirating into a Unicam 919 Solar System Atomic Spectrophotometer equipped with an automatic readout device after appropriate dilutions. Replicate analyses of the samples and reagent blanks were also carried out. Recovery studies carried out on all the metals gave results ranging between 95% and 98%.

2 Results and discussion

The metal levels in shells and muscles of the samples were measured as μg/g on a dry weight basis. However, emphasis will be more on the muscles because of its dietary significance. Table 1 contain the mean levels, ranges, standard deviations and coefficients of variation of the metals Ni, Pb, Zn and Cr in shells and muscles of samples collected from NTB, NTU and UYM sites (Fig.1).

Table 1 Metal concentrations (μg/g) of Ni, Pb, Zn and Cr in samples

Metal	Sample	NTB		NTU		UYM	
		Mean ± *S.D.*	*r*	Mean ± *S.D.*	*r*	Mean ± *S.D.*	*r*
Ni	Shell	0.055±0.012	0.853**	0.016±0.006	0.243n.s.	0.040±0.019	0.853**
	Muscle	0.150±0.022		0.033±0.018		0.140±0.020	
Pb	Shell	0.070±0.016	0.748**	0.015±0.013	0.230n.s.	0.116±0.012	0.748**
	Muscle	0.120±0.029		0.070±0.016		0.190±0.024	
Zn	Shell	2.390±2.728	0.641*	1.280±0.463	0.064n.s.	3.135±0.990	0.641*
	Muscle	6.370±1.879		3.840±1.006		8.615±1.391	
Cr	Shell	0.004±0.002	0.819**	N.D. ±N.D.	0.000n.s.	0.040±0.019	0.819**
	Muscle	0.485±0.226		0.113±0.046		0.625±0.157	

* : significant; ** : very significant; n.s. : not significant

The correlation coefficients (*r*) between levels of these metals in shells and muscles in the samples from respective sites are given in Table 1. Fig.2 shows comparative mean levels of these metals in muscles on site basis gives a better appreciation of the coefficients of variation on site basis.

All the metals except Cr in shells of NTU samples were present in all the samples, albeit at different levels and their respective mean levels in the shells and muscles on site basis were: NTB (0.055 μg/g, 0.150 μg/g); NTU (0.016 μg/g, 0.033 μg/g) and UYM (0.040 μg/g, 0.140 μg/g) for Ni. The mean levels for Pb in shells and muscles were: NTB (0.070 μg/g, 0.120 μg/g); NTU (0.015 μg/g, 0.070 μg/g) and UYM (0.116 μg/g, 0.190 μg/g). Similarly, the Zn levels in shells and muscles were: NTB (2.390 μg/g, 6.370 μg/g); NTU (1.280 μg/g, 3.840 μg/g) and UYM (3.135 μg/g, 8.615 μg/g) while for Cr, the levels were: NTB (0.004 μg/g, 0.485 μg/g); NTU (N.D., 0.113 μg/g) and UYM (0.040 μg/g, 0.625 μg/g). The levels of all these metals in the muscles of all the samples including NTU samples were comparatively higher than those in shells with Zn displaying the highest levels (Fig.2). This was probably so because unlike the muscles, the shells of this animal have low metal absorption potential. A comparative look at the coefficients of variation shows that Ni (*C. V.* = 21.016%) in shells of NTB samples was more stable than Ni in NTU and UYM samples with respective *C. Vs* of 41.277% and 48.597%. On the other hand Ni in muscles of UYM samples (*C. V.* = 14.365%) was more stable than Ni in NTB samples (*C. V.* = 14.944%). This in turn was more stable than Ni in NTU samples (*C. V.* = 54.778%). All the metals except Cr in NTB samples and Ni in NTU samples (*C. Vs* > 45.000%) were very stable in the muscles. However, whereas Pb seems to have been the most unstable metal in the shells of NTB samples (*C. V.* = 114.136%), followed by Pb in shells of NTU samples (*C. V.* = 86.709%), it is Pb that also appears to have been the most stable metal in the muscles of UYM samples (*C. V.* = 12.844%) followed by Ni in muscles of UYM samples (*C. V.* = 14.365%) and Ni in muscles of NTB samples (*C. V.* = 14.944%). Stability of these metals in the samples implies the ability of the muscles to accumulate metals over a period of time until they become bioconcentrated in the tissues and subsequently get biotransferred to the consumer. As man continues to feed on such metal-contaminated species, the levels of these metals could become biomagnified until they reach toxic levels which may result in various health symptoms that may ultimatimately lead to death of the consumer. The stability of these metals in the muscles is disturbing when it is realized that muscles are the edible portions of the animal.

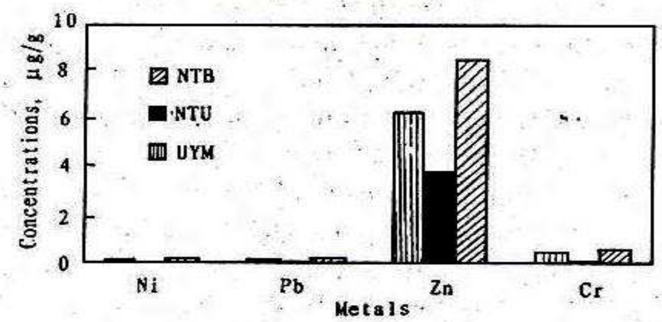

Fig.2 Comparative mean metal levels (μg/g) in muscles of samples

At 95% confidence level, the Pb and Zn levels in shells and muscles of NTB samples correlated very significantly (r = 0.871 and r = 0.732, respectively). In the same way, the levels of Ni, Pb and Cr in shells and muscles of UYM samples also correlated very significantly while the correlation between Cr levels in shells and muscles of NTB samples and between Zn levels in shells and muscles of UYM samples were merely significant. On the other hand, there was no significant correlation at all between levels of all the metals in shells and muscles of NTU samples (Table 2). Significant correlation between the levels of the metals in shells and muscles gives an indication of the dependence of the metal levels in shells on those in the muscles. This agrees with the findings of Laskowski and Hopkin (Laskowski, 1996). The levels of these metals in NTB and UYM samples were relatively much higher than those in NTU

Table 2 Correlation between levels of Ni, Pb, Zn and Cr in shells and muscles of *Achatina achatina*

Sites/samples	Trace metals	r-values
NTB	Ni	0.276$^{n.s}$
	Pb	0.871**
	Zn	0.732**
	Cr	0.536*
NTU	Ni	0.243$^{n.s}$
	Pb	0.230$^{n.s}$
	Zn	0.064$^{n.s}$
	Cr	0.000$^{n.s}$
UYM	Ni	0.853**
	Pb	0.748**
	Zn	0.641*
	Cr	0.819**

* = significant; * * = very significant; n.s = not significant

samples probably because NTB and UYM samples were randomly collected and were likely to have been exposed to trace metal contamination resulting from anthropogenic activities such as uncontrolled discharge of domestic sewage and industrial waste water into farmlands, plantations and abandoned plots of uncultivable land where this species of snails are often found in large numbers. It could also have resulted from the careless and indiscriminate disposal of solid wastes in the environments. Moreover, NTB and UYM sites were respectively within a Local Government Headquarters and a State Capital and were therefore more likely to have experienced more anthropogenic activities that resulted in more pollution of the sites than the "enclosed and restricted" Snail Farm of NTU site. In the case of NTU samples, it is suspected that Pb levels must have increased as a result of the spilled leaded-petrol and other lubricants used in the running and maintenance of the farm's power generating plant. The relative high levels of Pb and Cr in NTU samples are believed to have also resulted from the use of gloss paints containing these transition metals for the internal and external decorations of the farm's buildings and surroundings. Moreover, since NTU samples were nourished with artificial feedstuffs, the possibility of any contaminants including toxic metals present in the feedstuffs being transferred to the organisms cannot be ruled out. Furthermore, being specially nourished, the snails must have grown unusually larger in size than the ones often reared naturally and according to Bryan and Uysal (Bryan, 1978), levels of Pb metal in animals often increases with size of the animal. This they attributed to increase in tissue growth rather than to a reduction in body burden. This could also be said of the other metals.

Lead (Pb) and Cr are known to present environmental pollution problems worldwide because of their wide applicability in industries. In fact, the input of anthropogenically derived Pb to the environment has now outweighed all natural sources and may likely remain so for many years. Unfortunately, the practice of snail hunting is very common among children of school age who also happen to consume a lot of this animal. Results of toxicological studies on Pb have shown that this metal is not only a neurotoxin that has been linked up with several symptoms such as fatigue, loses of appetite, constipation, colic anemia, neuritis, seizures, general weakness, insomnia, hypertension, renal dysfunction, sperm count suppression and death, it is also found to be responsible for the decline in the intelligence of children exposed to quite low concentrations. According to Moore (Moore, 1991), chronic neuropathy in children is evident at a blood level of 70 to 100 $\mu g/dL$ while colic and other gastrointestinal symptoms are evident at 60 $\mu g/dL$. Similarly, anemia may appear at PbB of 70 $\mu g/dL$ and reduced haemoglobin synthesis at 40 $\mu g/dL$. These values are said to be lower than those reported for adults. There is also an evidence that Pb$<$30 $\mu g/dL$ induce cognitive and behavioral disorders in children (Ernhart, 1988). Moreover, since children generally have higher relative food intake capacity than the adults, they are more prone to Pb poisoning than the adults. Eventhough Cr was not detected in the shells of NTU samples, appreciable concentrations were recorded in the muscles. In most cases, the toxicity of Cr depend on its oxidation state. For instance, although there is yet no strong data base to draw a definitive conclusion on the carcinogenicity of Cr compounds, Langard (Langard, 1988) and Langfranchi *et al*. (Langfranchi, 1988) are of the opinion that the more soluble Cr^{6+} salts are more toxic than Cr^{3+} in humans and are often mutagenic and sometimes carcinogenic. Moreover, sorption of Cr is said to increase generally with temperature and any factor capable of increasing metabolic rate particularly in young animals. However, V-Balogh *et al*. (V-Balogh, 1988) observed a negative relationship for total Cr and body size in the gastropod *Lymnaea stagnalis* and attributed this to differing levels of exposure as the animal grew. The higher Pb and Cr levels in

UYM samples compared to those of NTB samples may have resulted from the higher degree of industrial activities in Uyo municipality than in Nsit Ibom Local Government Area.

Zn levels in both shells and muscles of all the samples were generally higher than the levels of the other metals in the samples (Table 1; Fig.2). This was probably so because of the abundance of Zn in the earth's crust as well as in effluent and wastes discharged into the environments. Thus activities which tend to generate more Zn into the environments should be carried out with caution. This is necessary since Zn is not a completely non-toxic metal as many people often believe. In fact, excess Zn resulting from ingestion of an amount$>$2g is said to be capable of producing symptoms such as fever, diarrhea, vomiting and gastrointestinal tract irritation in humans (Moore, 1991). Excess Zn in food thus poses a threat to life for eventhough sorption of Zn from food is relatively low, maximum residues often occur in the muscle, liver and kidney (Carson, 1987).

Nickel (Ni) is an important ingredient in many industrial processes because of its lustre, corrosion resistance, alloying ability, high strength and durability, good thermal and electrical conductivity. Therefore Ni is often present in industrial and municipal wastewater and sludge. Little attention has always been given to Ni toxicity and according to Moore (Moore, 1991), no evidence of carcinogenicity exists for ingestion of Ni in either food or water. Contrarily, Coogan *et al*. (Coogan, 1989) had earlier contended that Ni in some forms is likely to be carcinogenic in humans eventhough clinical or epidemiological studies were not available on the effects of oral ingestion of Ni. According to them in addition to evidence of statistically significant elevations of respiratory cancers among Ni refinery workers, very high concentrations of this metal have also been recorded in the liver, kidney and brains of humans. This presupposes that Ni could accumulate in tissues of organisms with high metabolic activity wherever it occurs in excess. Therefore just like any other toxic metal, Ni-laden wastes should not be disposed of indiscriminately particularly in urban cities and densely populated rural areas.

3 Conclusion

Since there are many factors which have been claimed by various authors to influence the accumulation of trace metals by organisms, it would be unreasonable to attribute the levels of the toxic metals in the samples to any particular factor(s). On the other hand, it is perhaps more rational to suggest that whereas levels of the metals in NTB and UYM samples were influenced by geographical location, age/period of exposure of the organisms to the metals, essentiality or non essentiality of the metals to the organisms, seasonal variation and perhaps temperature and pH, the levels of NTU sample must have been largely influenced by the location of the Snail Farm, source and quality of feeds as well as size and tissue of the organisms considered. Moreover, since the study was conducted during the rainy season, it is possible that metals available, particularly in NTB and UYM sites must have become speciated depending on temperature, pH and the presence of other elements. This must have increased the chances of these metals reacting with organic substances present in the environments to form organometallic compounds. Since Akwa Ibom State is an oil producing state, several of such compounds including those of Ni, Pb, Zn and Cr are likely to be present. These organometallic compounds possess lyophilic properties which make them readily penetrable to the brain, thus making them more neurotoxic than inorganic compounds. The consumption of snail contaminated by these metals could therefore be dangerous and as revealed by results of this study, these metals are more stable in the edible muscles. Being one of the cheapen sources of protein in Nigeria, many people may continue to consume this organism regardless of where it is obtained. This is worrisome since the effects of toxic metals are often cumulative. Such cumulative effects are capable of leading to ailments that may defy any medication. This could result in several disabilities, incapacitation, infantile and/or premature deaths.

Therefore since these metals are already present naturally in the environments, there is need to monitor the quality of meat including snails sold out for public consumption. Efforts should also be intensified by the Akwa Ibom State Protection Agency at ensuring compliance with laws on effluent and solid waste discharge. Although it is difficult to completely prevent environmental pollution, it may not be difficult to control environmental conditions in animal farms if contaminant-free animals are to be bred for human consumption. In other to achieve this, the government should assist in establishing animal farms and encouraging other people to do so according to specifications and environmental guidelines. This calls for enlightenment programmes by the government and the environmental protection agency to inform the public of the danger of indiscriminate collection and consumption of animals including snails from pollution prone environments such as cities, towns and densely populated rural areas. Farm managers should be specially advised to exercise prudence in the choice of feedstuffs as well as in the handling of the feeds since any contaminant(s) ingested by the animal could subsequently be transferred to the consumers. Therefore the relative high levels of these metals particularly Pb and Cr in the muscles of NTU samples notwithstanding it could be said that the snails bred in an "enclosed and restricted" environment devoid of much anthropogenic activities are safer for human consumption than those often collected randomly from "open and unrestricted" environments.

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