

FEDERAL UNIVERSITY OF LAFIA INAUGURAL LECTURE SERIES NO. 25

FACULTY OF SCIENCE, DEPARTMENT OF CHEMISTRY



"DIRT" EATING (GEOPHAGY) AND CHEMICAL KINETICS OF GEOPHAGIC CONSTITUENTS IN HEALTHY TISSUES AND ORGANS

Prof. Suleiman Philip Ivom Ogah Professor of Analytical Chemistry

December 3, 2024



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DEDICATION

This publication is dedicated to my late mother, Madam Orogwu Ogah (nwoku nahuru egbe nwa Oria Anyigo Isa).

THE PRESENTER



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PROTOCOL

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- The Head of Department of Chemistry, Prof. Usman Abdullahi;
- Heads of other Departments and Units;
- The Chairman, Academic Staff Union of Universities (ASUU);
- Members of Academic Staff;

- Chairmen of other Unions here present;
- Members of Non-Teaching Staff;
- Your Royal Highness, Eze Sheikh Abdulfatah Chimaeze Emetumah III, Ofor of Umuofor and the Grand Imam of Igbo Land;
- Your Royal Highness Igwe Nathaniel Nduba Udeh, The Eze Igbo I of Nasarawa State and His Carbinet;
- Other Royal Highnessess here present;
- The President and members of Igbo Community in Lafia;
- The Chairman & members of Ikwo Dev. Union, Lafia Branch;
- The Chairman & members of Ebonyi State Community in Lafia;
- Honourable members here present;
- My wife, Hajia Mariam Ogah, and my sons, Muhammad Ivom Ogah and Umar Ivom Ogah; my daughters Adama, Fatima and Hajju; my grand children, Hidaya, Jasmine, Khaleel, Hanifa and Hamida; my wonderful sons in-law, Alhaji Suleiman Obinna Udo and Dr Alhaji Faisal Chinonso Emetummah;
- My former and present students;
- Members of the University Media team;
- Gentlemen of the Press;
- Ladies and Gentle men.

You are all welcome to my Inaugural Lecture.

BENEDICTION:

B ismillahi Al-Rahman Al-Raheem. In the name of Allah, the Beneficent, the Merciful. All praises be unto Allah the Lord of the World and Heavens. Praises be unto Him who created the world and gave man dominion over all things. Praises be unto Him who sees the hearts of men and prevents men from seeing the hearts and future of each other. May the Peace and Mercy of Allah be upon Muhammad (S.A.W), the noblest of all Prophets.

PREAMBLE:

et me begin this part with the words of one of my late senior brothers Chief Onwe Ogah Nweke, who asked me and I quote "Did you ever know that you would see the walls of a University?" my answer was no, he went further to say, "forgive whoever that offended you in the family" he continued, "the best journey of life for a man is for his *uwa* to be in front so that if the man is slow his *uwa* will pull him forward and the worst journey is that of man walking in front of his uwa such that his uwa would pull him back for whatever reason during the journey". The *uwa* here can be translated to mean god that guides every action and inaction of a human being as everyone is believed to have his own personal god. It may also mean what some people call guardian angel. We had this discursion sometime in 1986 when I got admission for diploma in science education. My senior brother was actually making reference to the situation that arose in the family when I made a decision as a small boy to go to school (primary school) in 1972 in a polygamous family of thirteen wives and over fifty children for which I am the last in the absence of the father as the head who died about ten years back. I grew up not to know my father because he died when I was less than six months old. The beginning was not easy and should be a story for another day and for a different audience. I was lucky that as I was concluding my primary education in 1977, one of my senior brothers, prof. F. U. Ogah, who just returned from overseas, took over to sponsor my secondary education.

My University education started in 1986 with admission into diploma programme in University of Maiduguri after all efforts to go to school of nursing failed. In fact, it was in one of the interviews that one member asked what I was looking for in school of nursing with my type of O'level result. He asked why I couldn't go to University. I can only state here that my education from primary school to PhD was self-sponsored except secondary school which I was partly sponsored by my senior brother Prof. F.U. Ogah. It may also surprise my audience to hear that I did not apply to read science in the University. My first choice was Arabic and Islamic studies, the second choice was Sharia Law and third choice was Language Education. I found my name under diploma in science education which I passed with distinction. One of my chemistry lecturers Mr. Adam called me and said; "Ogah, look I would advise you to go into Chemistry major after this your diploma. You have enough knowledge of education to teach, after all I did not pass through education but, I don't think I am doing badly in teaching" and in deed he was a mile stone in teaching. That was how I moved to Chemistry and now a professor in Chemistry, a professor of Analytical Chemistry.

My way to University as a lecturer started in 2001 with a call from the head of our family, Chief Nwazunku Ogah, that I should come home from the north (Maiduguri Borno State to Abakaliki Ebonyi State). I assumed duty as assistant lecturer in Ebonyi State University in 2002. The process of relocating from Maiduguri in Borno State to Abakaliki in Ebonyi State was not easy, but we leave the story for another day. However, it is worthy to note here that my appointment by Ebonyi State University was in December 2001, but my assumption of duty was almost a year later, August 2002.

The road to professorship in Federal University of Lafia started in October 2015. On a Thursday my senior brother prof. F. U. Ogah called and told me to get ready to leave for Lafia on Sunday against Monday. He further said that I should go with my credentials. He said that on Monday, I should go to the V-C's office and that I should call him when I got there. I did not ask questions other than to obey instructions because of our type of traditional training (who am I to ask questions except for clarity when seniors are talking?). That was how I met Prof. Ekanem Brade the rest is history. The nature of the road in FULafia is another chapter of a book for different audience.

Analytical Chemistry; Analytical Chemistry is a branch of chemistry that is concerned with separation and analyses of constituents of chemical substances. Chemical substances here may mean any material because we may ask; is there any substance that is not chemical? Analytical chemistry tries to find out two things about a substance:

- 1. What is in there in the substance, Qualitative Analyses;
- 2. How much of what is there is there, Quantitative Analyses.

Analytical Chemist is charged with the responsibility of providing answers to the two questions above and sometimes takes further steps to determine the structural constituent of the substance. Analytical Chemist is a general Chemist whose work transverses all branches of Chemistry just like our lecture today cuts across Inorganic Chemistry, Clinical Chemistry, environmental and Physical Chemistry.

"DIRT" EATING (GEOPHAGY) AND CHEMICAL KINETICS OF GEOPHAGIC CONSTITUENTS IN HEALTHY TISSUES AND ORGANS

INTRODUCTION:

t is generally agreed all over the world and from the beginning of time that man's basic needs on earth are food, shelter and L clothing. Every other thing man does is just supplementary to these. It is also undisputable that while man adapts to living without shelter and clothing, it is not possible to live without food. However, just availability or abundance of food does not guarantee existence of life for man. Food must satisfy certain requirement like free from microbes and lethal elements in addition to containing what are considered as essential for survival and development such as minerals and vitamins. In quest for these, man engages in various practices some of which might even endanger his life. Some other time the kind of thing man eats leaves one to begin to ask question as to what does man mean by food. The eating of "dirt" or earth called clay is one of such things. It is that part of the earth that man eats, processed or unprocessed, that this work calls edible clay.

Clay is a naturally occurring aluminum silicate (Guggenheim and Martin, 1995). It is principally composed of ultra fine grain mineral. Clays are distinguished from other fine grain soils by differences in size and mineralogy. Silt is a fine soil similar to clay. However, clay is finer (in fact ultra fine grain). It is so fine that it is almost impossible to identify crystal from it. The distinction between clay and silt grains varies by discipline. Geologist, soil scientist, sendimentologists and colloid chemist all draw their distinction based on particle size in µm which in all cases, clay has the finest particle. Geotechnical engineers distinguish between silts and clays based on soil plasticity (Velde, 1995).

Clay is widely eaten by children and pregnant women in eastern part of Nigeria. Some just pick up some clay particles from excavation sites and eat without any preparation while some others buy from the local market to eat. Some pregnant women can hardly do without it, while believing that it enhances the development of their babies and impart fair complexion to the babies.

Even when a baby is born the same clay is used as lotion on the new born baby. There are so many local applications of clay with so many believes attached to it.

Clay has been noted to contain aluminum as principal metal. Since clay in most cases are formed as a result of sedimentation and weathering of rocks it is most likely that some other metals might be embedded in the clay that are consumed by same people that eat clay. It is of interest because while some metals at certain concentrations may be useful as part of diet others may be harmful and accumulative effect pose danger to health.

No one has ever bordered to ask questions about the constituents of the clay eat by pregnant women in south eastern part of Nigeria, neither has the kinetics of any of such metal, been questioned.

In this research we analyze edible clay obtained from Enyigba village in Abakaliki Ebonyi State, Nigeria for some elements: Pb, Cd, Cu, Zn, Ba, Mn, Ca, Ni, Na, Mg, K, Fe, As, Co, and Cr.

Some tissues such as brain, heart, liver, kidney, lungs, skeletal muscle and blood from rabbits were analyzed for some of the elements following oral administration of clay to the animals.

The kinetics of some of the elements in the edible clay in the tissues and blood from the experimental rabbits were also determined.

ORIGIN OF CLAY

Clay form from weathering and secondary sedimentation processes with only few forming from primary igneous or metamorphic environment (Amythst, 1995). Clay are also formed generally by gradual chemical weathering of silicate bearing rock over long period of time by low concentration of carbonic acid and other diluted solvent. Clays are formed from hydrothermal activities. Clay deposit may be formed as residual deposit in soil, but thick deposits are usually formed as a result of secondary sedimentary deposition process after having been eroded and transported from their original location of formation (Ehlers and Blatt, 1982).

This implies that there are two primary sources of clay: **residual clay and transported clay.** Transported clays are more extensive than residuals having been removed from their original place by weathering and erosion and deposited at new location (Frolking, 1982).

THE CLAY MINERALS

Clay minerals have common set of structural and chemical properties (Costanzo, 2001). They are members of phylosilicates that contain large percentage of water trapped within the silicate sheets (Amethyst, 2011). There are four groups of clay minerals:

(i) The Kaolinite Group:

This is also known as white cosmetic clay (Natural, 2011). It has the formula of $Al_2 Si_2O_5$ (OH)₄.

(ii) The montimorillonite/smectite group.

This group is also known as Bentonite clay. It is edible clay from naturally occurring volcanic ash sediments (Natural, 2011). They have the general chemical formula (Ca, Na, H)(Al, Mg, Fe, Zn) $_2$ (Si, Al) $_4$ O $_{10}$ (OH) $_2$ XH $_2$ O where X is variable amount of water (Amethyst,2011).

(iii) The Illite or the clay mica group

This group is also known as **French Green clay or sea clay** (Natural, 2011). It is rock forming mineral with the general formula (K, H) Al_2 (Si, Al) $_4$ O₁₀ (OH) $_2$ - XH₂O. X is variable amount of water (Amethyst, 2011).

(iv) The Chlorite Group: This group is relatively large and common. The general formula is X_4 - $6Y_4 O_{10} (OH, O)_8$, the X is either Al, Fe, Li, Mg, Mn, Ni, Zn, or Cr, Y is either Al, Si, B, or Fe (Amethyst, 2011).

GEOPHAGY

Eating of "dirt" clay earth, soil or related material is generally referred to as *geophagy*, and such material referred to as *geophagic* material (Ekosse and Ngole, 2012). *Geophagy* is a worldwide practice (Aufreiter *et al*, 1997; Woywodt and Kiss, 2012). It is practiced by humans of all gender irrespective of age or race (Geissler *et al*, 1998). *Geophagy* practices cuts across African continent from east Africa to West Africa including Nigeria, Cameroon and Ghana and North Africa to South Africa (Abrahams and Parsons, 1997, Ekosse *et al*, 2010). Diamond (1998) concluded that animals eat "dirt" clay to act as detoxins to protect them from poison arising from eating poisonous plants.

Geophagy is also known as soil paca or simply pica. It is a tradition in places like Philippians, New Guinea, Coasta Rica, the Amazon and Orinoco basins of South America to use clay as part of food (John, 1990). Gerald Callahan called Pica a disease different from pollio or smallpox (Callahan, 2003). *Geophagy* is more pronounced among pregnant women especially in sub-saharan Africa including United States of America and children worldwide. The US Agency for Toxic Substances Committee (2000) identified 500mg of soil per day to be pathologic (ATSDR, 2000). Children under the age of 2 eat different kinds of soil from different sources Callahan (2003). While adult may be selective and consume clay from deeper part of the earth, children are less selective and eat top part of the soil.

HEALTH USES OF CLAY

Generally, all the groups of clay have health uses as in medicine and cosmetics.

Different people may have different reasons for eating clay or soil as sometimes called. However, for whatever reason one has there are definitely merits and demerits in the practice. Clay detoxifies by binding alkalloids in some poisonous plants and plant materials (Diamond, 1995).

Geophagy is mostly common among pregnant women and nursing mothers who have high demand for minerals and mineral supplements. Other Americans such as the indigenous Pomo of Northern California used clay in their diet mixed with ground acorn to neutralize the acid of the acorn (Hunter, 1993). Clay or soil consumption is believed to have relieving effects from ailments which includes supplementation of minerals and nutrients (Ekosse and Ngole, 2012).

Geophagic materials are used orally to heal common ailments of gastrointestinal tract because of the medicinal properties (Carretero, 2002). Clay or other earthy material is consumed to relieve hunger (Brand *et al*, 2010). Iron rich clay has been found to be a very effective antibiotic (online). Just as almost natural in life that everything has merits and demerits, *geophagy* has its own demerits. There are reports of lead poisoning and other toxicities in children eating contaminated soils (Callahan, 2003). Torvik *et al* (1990) found millions of species of microorganisms in soil.

Kent and Triplett (2002) found even more species of *prokaryotic* microorganism in the soil. American centers for Disease control and prevention (2002) reported infection of two children at separate sites with *raccoon* roundworm, *Baylisacaris procyoris,* due to eating of infected soil material. One of the victims died of severe neurologic damage (Callahan, 2003).

Geophagy can have dire consequences. Most of the disease encountered through *geophagy* is childhood related because infected top soil is involved rather than deep clays (Torvik *et al*, 1990). Infection from geophagic material was reported in the United States with *toxocariasis* as the most common due to ingestion of soil contaminated with dog or cat feces (Laufer, 2002). Ozumba and Ozumba (2002) reported that the most common parasitic infection associated with *geophagy* or dirt eating among Nigerian children is ascariasis.

Clay is widely eaten across the world particularly by pregnant women, a habit known as *Geophagy*. The native pregnant women who eat clay claim that it helps to keep the baby in the womb healthy. Some say that it works as anti-vomiting in the early stage of pregnancy. Some also eat it because of its taste and others say they eat it because of its scent. All these are oral claims but most importantly some clays are rich in essential nutrient such as sulphour and phosphorous. The practice of clay eating is widely spread among animals and humans (Peter, 2003).

Geophagy is common in rural or pre-industrial societies among children and pregnant women (Callahan, 2003). It has been recorded that sick or injured animals such as mammals, birds, butterflies and even reptiles use clay for medicinal purpose (Cooper, 2000). *Geophagy* is practiced by members of all races, social classes, ages and sexes. In many parts of the developing world clay for consumption are displayed for sale. In some parts of Africa, rural areas of the United States and villages in India clay consumption is associated with pregnancy and some women eat clay to eliminate nausea, possibly because the clay coats the gastro intestinal tract and may absorb dangerous toxin. The clay rich in calcium may provide calcium for fetal development (Vermeer, 1975). People seem to use geophagy to protect themselves from plant toxins. Indians that eat bitter and toxic wild potatoes capable of producing stomach pains and vomiting, usually eat them with clay to make them safe by binding the alkaloids (Diamond, 1998).

In fact a lot of works are well documented on animal and human geophagy either for nutritious reasons or medicinal reason. It is recommended that a *paraquat* poisoned victim should swallow "dirt" clay even at the risk of salmonella, because *paraquat* will be deactivated upon contact with soil (Wong, 1993). Chimpanzees in Kibale National Park, Uganda have been observed to consume soil rich in kaolinite clay shortly before or after consuming plants including *Trichilia rubescens*, which has anti-malarial properties. Simulated mastication and digestion has shown that clay helps to release active anti-malarial component from the leaves (Ziegler, 1997). There are health risks associated with clay consumptions. Contamination by animal or human feces, parasite eggs, such as round worm that can stay dormant for years can present a problem. Tetanus poses another risk (Abraham,2003). Some clay may possibly contain toxic metals.

PROPERTIES OF CLAY

The atomic structure and physical properties of clay minerals cannot be easily understood experimentally because of poor crystallinity and ultra fine particles (Bock *et al*, 1995). Clay minerals all have great affinity for water, some swell easily and may double in thickness when wet (Velde, 1995). This property is utilized by the paint industry to disperse pigment evenly throughout the paint.

Clay mineral tends to form microscopic to sub-microscopic crystals. They can absorb or lose water from simple humidity changes.

There are two main parts or properties that give clay wide applications.

- (a) The large surface area arising from the tiny size of the particles. This property is utilized in ion exchange in detoxification process (USGS, 1999).
- (b) The electrically charged nature of the clay particles results in very strong electrostatic interactions. According to Johnson (1996) there are six different active sites in clay mineral as (i)"broken edge" site (ii) Isomorphic substitution site (iii) Exchangeable cation site. (iv) Hydrophobic surface (v) Hydration shell of exchangeable cation and (vi) Hydrophobic sites on adsorbed organic molecules.

Naturally clay is negatively charged and cat ions can easily bind to their natural surface while negative or non-polar hydrophobic chemicals will be rejected (Lahav, 1983). Adsorption of organic cat ions to the clay charged sites exchanging the original inorganic cat ions, might exhibit hydrophobic movement toward the outside surface of the organic clay composite. Such surface can bind non-polar chemicals. In some cases it may give rise to interaction with negatively charged chemicals due to charge reversal on the clay surface as a result of over load in organic cat ions (Rytwo *et al*, 1998).

SAMPLE AREAAND SAMPLE COLLECTION

Samples of clay were collected from Enyigba village mining site in Abakaliki Ebonyi State of Nigeria, **Figures 1 and 2.** The samples were bought from different sampling points at the mining gate as shown figures **3 and 4**.



Figure 1. Map of Nigeria showing Ebonyi state in deep yellow.



Figure 2. Map of Ebonyi state Nigeria showing sample collection Areas.



Figure 3a. Lead mining Site in Enyigba Ebonyi State Nigeria where clay was collected as a 'byproduct of mining'



Figure 3b. Clay Mining Site in Enyigba Ebonyi State Nigeria



Figure 3c: Edible Industrial Site

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Figure 3d: Displayed Edible Clay

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PREPARATION OF CLAY SAMPLE FOR ANALYSIS

The sample from the different sampling points were ground together and thoroughly mixed. The fine powder was dried in an oven at a temperature of 110°C and then cooled and heated to a constant weight. From this bulk sample 2000 mg of the dried sample was weighed. The two grams was a representative sample of the bulk. This sample was placed in a beaker and 10 cm³ of distilled water added just to make paste of the clay. Sixty (60) cm^3 of freshly mixed aqua regia (mixture of Nitric acid and Hydrochloric acid in the of 1:3 y volume) was added little at a time to dissolve the material. The mixture was allowed to stand overnight for complete digestion of the clay sample (Ogah eta al; 2016). Then the mixture was diluted with distilled water, and filtered. The filtrate was kept in refrigerator at 4°C and analyzed for the elements using Atomic Absorption Spectroscopy (AAS). The samples were run in triplicate. The elements whose concentration levels were determined include Pb, Cd, Cu, Zn, Ba, Mn, Ca, Ni, Na, Mg, K, Fe, As, Co and Cr at the appropriate wavelength and lamp current for each analyte.

EXPERIMENTALANIMALS

Fifty nine clinically healthy rabbits of adult age of both sexes were used. They were kept in laboratory for 14 days in Veterinary Teaching Hospital Federal University of Agriculture Makurdi to acclimatize, (**Figure 4**). The weight of each animal was also determined by use of scale balance. The animals were clearly labeled and each body weight recorded. The animals were fed with grass and vegetables.

ACUTE TOXICITY STUDY:

The acute toxicity of the clay on the rabbits was studied using up and down method as revised by Dixon, (1991). The rabbits were fasted over night but allowed to drink water prior to the study. Five adult rabbits of known weight were randomly selected and used for the study. The up and down method aims to estimate the LD_{50} value by testing individual animal sequentially the dose for each animal being adjusted up or down, depending upon the outcome of the previous animal (USEPA,1998).

Principle of limit test: The limit test is a sequential test that uses a maximum of 5 animals. A test of 2000, or exceptionally 5000 mg/kg body weight may be used. It is primarily used in situations where the experimenter has information indicating that the material is likely to be non-toxic (DE CD, 1987). 5000mg /kg body weight of freshly made paste of clay in distilled water was given orally to one of the rabbits using stomach tube. The dosed animal was observed for 48h for signs of toxicity or death. The animal survived. The same procedure was adopted until all the five animals were treated. There was no loss of animal but the surviving animals were observed for 14 days.

TISSUE KINETICS OF THE CLAY CONSTITUENT IN RABBITS:

Fifty four (54) rabbits were used for this study. They were separated in three equal groups, and placed in different cages. Group A was administered 2000mg/kg body weight of freshly made paste of clay while Group B was administered 4000mg/kg body weight. Group C, was control and was not dosed but fed with grasses and vegetables. The high and low doses were necessary to see if absorption and distribution into tissues would depend on the concentration of the constituent of the clay and hence whether the rate of disappearance in the tissue would depend on the concentration.

All the groups were fasted prior to the dosing but water was provided *ad-libitum* in plastic bowels. The clay was in the form close enough to that which it is eaten by humans. The fine weighed powder was made into paste by adding 10 mls of distilled water just enough to allow easy administration. After dosing all the animals were continuously fed with grass and vegetable and water *ad libitum*, but in different containers in different cages. Group C was administered with distilled water only from the source used for paste making of the clay sample. The animals were observed for any clinical behaviour.



COLLECTION OF TISSUE AND BLOOD SAMPLES

Three animals from each group were sacrificed each day of sample collection. Samples of brain, heart, liver, kidney, lungs, skeletal muscle and blood were collected from the sacrificed animals. Samples were collected on the following days post treatment with clay; 1^{st} day, 2^{nd} day, 4^{th} day, 6^{th} day, 8^{th} day, and 10^{th} day. The work area and instruments were thoroughly cleaned between sacrifice to avoid contamination. The tissue samples were put in plastic bags and stored in refrigerator below -10° C until analysed. Heparin was used as anticoagulant for blood sample (Ogah et al; 2016).

DIGESTION OF TISSUE SAMPLES

The tissues were dried to a constant weight in an oven at a temperature of 60° C. One gramme of the dried tissue was weighed into a conical flask. Fifteen (15) cm³ of freshly prepared aquaregia was added followed by gradual addition of 20 cm³ of 20% H₂O₂. The aqua-regia was employed to solublize the metal while the H₂O₂ was to oxidize the tissue. The mixture was placed on hot plate at 80°C for 2h. The solution was allowed to cool, then filtered and made up to 100 cm³ in a volumetric flask. The filtrate was refrigerated until further analysis.

DETERMINATIONS OF METALS IN TISSUE AND BLOOD SAMPLE USING AAS

The sample solutions were in turn aspirated for each suspected element in triplicate. The concentration of the analyte in the sample was recorded from read out devise of the bulk AAS.

BLANK PREPARATIONS:

The Blank were made by taking 15 mls of aqua-regia with 20 mls of 20% H₂O₂ in 100 cm³ volumetric flasks and made up to the mark with distilled water. This solution was aspirated into the flame of AAS and concentrations recorded at wave length of each suspected analyte.

Solutions of control were in turn run for each suspected element just like the sample. The concentrations were recorded. The values, where applicable were subtracted from the value for the corresponding sample.

CALCULATION OF KINETICS CONSTANTS

Blood or tissue kinetics is the mathematical description of concentration changes in the body with time. For the clay the decline in concentrations in blood and tissues with time after oral administration was examined under a single compartment open model (fig 5).



Fig 5. One compartment open kinetic model.

According to a single compartment model, the clay after being taken orally gains entry into the blood stream and is instantly and homogenously mixed, distributed and eliminated by excretion and/or metabolism from the body. This compartment is a mathematical entity with no physiological meaning but is useful in describing the disposition kinetics of a material (Mercer et-al, 1978). The kinetic analysis of experimental data obtained from blood and tissue was performed using a mean value by standard

procedures (Gibaldi and Perrier, 1982). A programme for linear regression analysis was used. The following kinetics parameters were determined.

Elimination rate: regression analysis was performed on concentration time data and the slope (i.e. change in concentration/change in time) is the elimination rate constant k.

Half life, $t_{1/2}$: Half life was obtained using the formular

$$t_{1/2} = \frac{\ln 2}{K} = \frac{0.693}{K}$$

Where, k is the elimination rate constant and

 $t_{1/2}$ is the half life

STATISTICAL ANALYSIS

The data collected were presented as mean standard error mean. One way analysis of variance (ANOVA) was used to analyze the differences between the means. P values less than 0.05 were considered significant (Armitage, 1980). Graphpard instant^(R) version 3.0 (2003) statistical computer softwares was used.

RESULTS OF ELEMENTAL ANALYSES OF EDIBLE CLAYFROMENYIGBAEBONYI STATE NIGERIA:

The results of the analyses of the elements present in the edible clay obtained from Enyigba Abakaliki, Ebonyi State Nigeria is presented in Tables 1a, b and c.

The essential metallic elements including Na, K, Ca, and Mg occurred in high concentrations in the edible clay. The concentrations of these elements in the edible clay were 71.960 ± 3.63 , 84.330 ± 2.73 , 249.00 ± 1.73 and 198.570 ± 0.90 mg/g

respectively for Na, K, Ca, and Mg (Table 1a). The concentrations of trace metallic nutrient elements in the edible clay are shown in table 1b. Iron (Fe) $(151.920\pm 2.6 \text{ mg/g})$ occurred in a very high concentration, while Cr $(0.170\pm 0.04 \text{ mg/g})$ was obtained in a very low concentration. The concentrations of Ni, Cu and Mn were 89.340 ± 0.69 , 40.690 ± 0.67 and $65.200\pm 0.81 \text{ mg/g}$ respectively. Some toxic heavy metals were found in the edible clay (Table 1c). The concentrations of these toxic heavy metals in the edible clay were Pb, $0.570\pm 0.08 \text{ mg/g}$; Cd, $1.930\pm 1.23 \text{ mg/g}$; As, $3.920\pm 1.90 \text{ mg/g}$ and Ba, $8.680\pm 4.91 \text{ mg/g}$.

ACUTE TOXICITY STUDY

The administration of the edible clay to rabbits at the dose of 5000 mg/kg for LD_{50} determination did not produce any mortality in the treated rabbits using the up and down method.

Table 1a: Essential metallic nutrient elements in the edibleclay obtained from Enyigba Abakaliki Ebonyi State Nigeria(Ogah et al; 2016).

Metals	Mean (<u>+</u> SE) Concentrations mg/g
Na	71.960 <u>+</u> 3.63
К	84.330 <u>+</u> 2.73
Са	249.000 <u>+</u> 1.73
Mg	198.570 <u>+</u> 0.90

SE = standard error mean.

 Table 1b: Trace metallic nutrient elements in edible clay obtained from Enyigba Abakaliki Ebonyi state Nigeria (Ogah and Ikelle, 2015).

Metals	Mean (<u>+</u> SE) Concentrations mg/g
Cu	40.690 <u>+</u> 0.67
Zn	0.830 <u>+</u> 0.02
Mn	65.200 <u>+</u> 0.81
Ni	89.340 <u>+</u> 0.69
Fe	151.920 <u>+</u> 2.6
Со	1.190 <u>+</u> 0.51
Cr	0.170 <u>+</u> 0.04

SE = standard error mean.

Table 1c: Toxic metals in edible clay obtained fromEnyigba Abakaliki Ebonyi State Nigeria(Ogah and Ikelle, 2015).

Metals	Mean (\pm SE) Concentrations mg/g
Pb	0.570 <u>+</u> 0.08
Cd	1.930 <u>+</u> 1.23
As	3.920 <u>+</u> 1.90
Ba	8.680 <u>+</u> 4.91
Al	1.29 <u>+</u> 0.24

SE = standard error mean.

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DISTRIBUTIONS OF METALS IN TISSUES AND BLOOD

Table 2: Mean $(\pm SE)$ Ca Concentration ($\mu g/g$) in tissues of rabbits orally treated with doses of 2000 mg/kg body weight (low) and 4000 mg/kg body weight (high) (Ogah et al; 2016).

Blood	424.74±25.08 ^b	486.60±93.20 ¹	416.49±71.90 ^a	482.50±24.70 ¹	305.15±8.25ª	437.10±55.50 ²	280.41±71.54 ^a	392.00±111.00 ¹	243.30±39.34ª	268.00±28.90 ²	243.30±39.34 ^b	268.00±28.90 ²	190 05
Skeletal Muscle	296.91±121.84ª	408.20±70.30 ²	131.96±57.73 ^b	309.00±125.00 ²	127.83±90.72ª	219.00±157.00 ²	90.72±17.97ª	190.00±134.00 ²	78.35±4.12ª	127.80±43.60 ²	65.98±4.12 ^b	78.35±4.12 ²	J. J.
Lungs	227.00±173.00ª	260.00 ± 143.00^1	119.60±27.00 ^a	223.00 ± 118.00^1	107.22±27.00ª	156.70±51.70 ¹	107.22±27.04ª	148.50±35.70 ²	107.20±14.87ª	127.80±47.60 ¹	74.23±7.14ª	119.60±45.90 ²	- - - -
Liver	325.77±138.74 ^b	425.00±178.00 ²	152.58±46.47 ^b	210.00±130.00 ²	107.20±33.00 ^b	148.45±106.66 ²	90.72±14.87 ^b	107.20±10.90 ²	74.23±7.14 ⁿ	74.23±7.14 ²	70.10±4.12 ^b	70.10±8.25 ¹	
Kidney	185.57±111.57 ^a	235.00±161.00	107.22±10.91ª	206.00±152	103.09±10.91ª	198.00±112.00	86.60±18.90 ²	115.50±18.00	86.60±0.00ª	107.20±10.90	74.23±7.14ª	90.70±16.50	-
Heart	222.68±142.31 ^b	297.00±125.00	177.32±91.002 ^b	223.00±161.00	123.71±55.78 ^b	210.00±124.00	115.46±29.74ª	202.00±110.00	90.72±10.91ª	190.00±128.00	70.10±10.91ª	107.20±10.90	-
Brain	384.00± 101.00 ^b	436.00±215.00 ²	280.00±123.00 ^b	293.00±137.00 ²	223.60±99.10ª	239.00±114.00 ²	206.20±18.00ª	235.00±149.00 ²	169.10±82.80ª	206.00±147.00 ¹	138.80±35.80ª	186.00±115 ¹	- -
Days/ Group	1. low	High	2. low	High	4. low	High	6. low	High	8. low	High	10.low	High	-

Table 3: Mean $(\pm SE)$ Mg Concentration $(\mu g/g)$ in tissues of rabbits orally treated with doses of 2000 mg/kg body weight (low) and 4000mg/kg body weight (high) (Ogah et al; 2016).

Blood	179.28±58.41ª	230.80±37.001	171.88±8.59 ^b	215.50±16.70 ¹	159.54±3.79ª	203.80±46.50 ¹	138.32±3.83b	180.90±18.10 ¹	41.48±17.80ª	161.35±5.15²	35.86±11.68ª	47.30±12.30 ¹
Skeletal Muscle	188.49±35.61ª	268.40±27.60 ¹	149.67±1.65ª	249.00±19.50 ¹	120.07±3.29ª	167.80±15.90 ¹	112.34±8.04 ^b	160.20±14.70 ²	31.84±0.72 ^b	150.00±19.30 ²	31.22±5.42ª	134.20±37.001
Lungs	160.53±25.33ª	196.05±3.66 ¹	157.24±10.84ª	175.66±5.221	150.00±2.96ª	173.00±13.80 ¹	146.38±19.52 ^b	161.40±11.50 ²	132.24±13.03 ^b	146.40±15.40 ²	107.73±7.84	146.40±19.50 ¹
Liver	207.24±26.11ª	226.60±19.90 ²	185.51±5.69ª	192.40±15.90 ¹	166.45±17.77ª	180.92±0.872	152.30±17.07b	165.40±18.60 ¹	139.80±10.77ª	155.90±26.00 ¹	139.47±11.97 ^b	145.60±16.10 ¹
Kidney	241.63±27.86 ^b	250.30±15.10 ¹	171.05±21.38ª	204.30±14.30 ¹	164.47±29.14ª	198.68±8.00 ²	146.38±16.67ª	191.45±8.39 ²	144.41±18.11 ^b	160.86±8.62 ²	141.45±16.45 ^b	157.70±11.40 ¹
Heart	137.69±16.35ª	152.60±15.10 ²	131.25±16.79 ^b	146.40±21.00 ¹	126.32±5.06ª	145.72±6.96 ²	118.09±13.37ª	139.50±11.10 ¹	113.49±5.78ª	134.50±13.00 ¹	62.17±21.71ª	120.07±2.301
Brain	175.66±13.25ª	208.88±2.16 ¹	148.36±24.64ª	196.50±51.60 ¹	145.40±2.63ª	187.50±28.10 ²	136.18±10.91ª	155.43±5.18 ¹	129.93±8.55 ^b	152.30±4.78 ¹	110.53±5.92 ^b	151.00±25.00 ²
Days/ Group	1. Low	High	2. Low	High	4. Low	High	6. Low	High	8. Low	High	10.Low	High

a, b, values for low dose along the same column differently superscript, differ significantly (p<0.05). 1, 2 value for high dose along the same column differently superscript, differ significantly (p<0.05).
Table 4: Mean $(\pm SE)$ Fe Concentration $(\mu g/g)$ in tissues of rabbits orally treated with doses of 2000 mg/kg body weight (low) and 4000 mg/kg body weight (high).

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Days/	Brain	Heart	Kidney	Liver	Lungs	Skeletal	Blood
Group						Muscle	
1. Low	228.68±12.18ª	269.77±12.31ª	297.67±52.42ª	253.72±11.40ª	273.64±8.53ª	170.73±3.29ª	350.39±17.05ª
High	265.10±14.00 ¹	344.20±46.501	326.40±13.00 ¹	442.85±9.491	404.70±21.30 ¹	251.94 ± 8.091	384.10±17.70 ²
2. Low	190.70±9.30ª	249.61±9.43ª	148.84±9.30ª	240.31±7.63 ^a	246.42±18.56 ^a	159.11±4.15ª	133.55±4.15ª
High	232.60±25.90 ¹	293.22±1.27 ¹	261.24±3.881	375.36±1.391	366.70±24.00 ¹	182.17±0.78 ¹	382.50±33.40 ¹
4. Low	139.38±13.93ª	248.84±16.28 ^b	141.44±13.15ª	224.03±0.78ª	245.74±5.43ª	153.33±9.10 ^b	131.01 <u>+</u> 4.32 ^b
High	206.05±8.751	287.00±11.30 ²	241.86±4.65 ¹	347.30±27.10 ²	329.50±17.00 ¹	158.14 ± 4.65^{1}	$348.84{\pm}0.001$
6. Low	130.23±12.31ª	143.41±5.08ª	134.11±12.18 ^b	162.12±5.42 ^a	198.75±2.34ª	88.70±9.47ª	125.25±12.96 ^b
High	148.84 ± 4.65^{1}	256.60±44.701	233.90±17.70 ²	309.30±2.331	297.70±36.30 ¹	140.00 ± 14.20^{1}	334.10±14.70 ²
8. Low	75.09±4.32 ^b	134.88±4.65 ^b	130.39±2.41 ^b	138.02±12.39b	170.39±13.53ª	62.40±5.41ª	111.63±0.00ª
High	95.04±2.64 ¹	251.94±8.091	208.50±15.40 ¹	307.75±5.432	265.12 ± 0.00^{1}	83.72±0.00 ¹	144.19±4.65 ¹
10. Low	67.44±2.33ª	97.99±0.32ª	127.13±4.10 ^b	69.69±0.08ª	135.66±10.08ª	41.39±0.41ª	108.30±3.33ª
High	68.45±0.66 ¹	171.30±11.602	200.00±4.65 ¹	88.99±2.641	220.00±8.69 ¹	70.05±0.29 ¹	141.09±6.75 ¹
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a, b, values for low dose along the same column differently superscript, differ significantly (p<0.05). 1, 2 value for high dose along the same column differently superscript, differ significantly (p<0.05). The oral administration of the edible clay to rabbits at two varying doses of 2000 mg/kg body weight and 4000 mg/kg body weight, resulted in detectable levels of most of the elements found in the clay in various tissues and blood of the treated animals.

Right from day 1 to day 10 post treatments with the clay (Table 2), Calcium occurred in the brain of the treated rabbits. The concentrations of calcium obtained at day 1 post treatment were $384.00+101.00 \mu g/g$ and $436.00+213.00 \mu g/g$ in animals treated with 2000 and 4000 mg/kg body weight respectively. There was a decline in Ca concentrations and, at 10 days post treatment the levels were $138.80 \pm 35.80 \ \mu g/g$ and $206.00 \pm 115.00 \ \mu g/g$ respectively in 2000 mg/kg body weight and 4000 mg/kg body weight of the treated animals. The concentrations of Ca observed to be present in the heart, kidney, liver, lungs, skeletal muscle and blood were highest at day 1 post treatment, and these were followed by constant decline in Ca levels in the various tissues and blood, with the levels in the various tissues and blood being the least on 10th day post treatment. The concentrations obtained with the 4000 mg/kg dose in various tissues and blood were significantly (P less than 0.05) higher than those obtained with the 2000 mg/kg dose (Table 2).

The concentrations of Mg present in the various tissues of rabbits treated orally with 2000 mg/kg and 4000 mg/kg body weight of the edible clay are shown in Tables 3. The highest concentration of this element was obtained at day 1 post treatment, while the least level was obtained at day 10 post treatment. The Mg concentrations were higher in the tissues and blood of animals treated with 4000 mg/kg dose when compared to the level present in the tissues and blood of 2000 mg/kg treated rabbits.

Table 5: Mean $(\pm SE)$ Cu Concentration ($\mu g/g$) in tissues of rabbits orally treated with doses of 2000 mg/kg body weight (low) and 4000 mg/kg body weight (high).

	Brain	Heart	Kidney	Liver	Lungs	Skeletal	Blood
						Muscle	
7.08±0.00)a	9.43±2.36ª	8.25±1.18ª	15.33 ± 1.18^{a}	6.90±0.18ª	3.54±0.00ª	0.00 ± 0.00
11.79±1.1	81	9.59±3.24 ²	11.79 ± 1.18^{2}	16.31 ± 1.09^{1}	10.16 ± 0.00^{1}	$7.08{\pm}0.00^{1}$	$3.54{\pm}0.00$
3.54±0.00	e	4.72±1.18ª	5.90±1.18ª	10.61 ± 0.00^{a}	3.54±0.00 ^b	3.54±0.00 ^b	0.00 ± 0.00
7.54±0.00	-1	$9.43{\pm}1.18^{1}$	9.43 ± 1.18^{1}	15.31 ± 2.35^{2}	5.90 ± 1.18^{1}	5.90 ± 1.18^{2}	0.00 ± 0.00
3.54 ± 0.00	q	4.72±1.18 ^b	3.54±0.00 ^a	4.72±2.36 ^a	0.00±0.00 ^b	1.18 ± 1.18^{a}	0.00 ± 0.00
7.08±0.00	5	$7.08{\pm}0.00^{1}$	8.28 ± 3.10^{2}	10.61 ± 2.04^{1}	4.72 ± 1.18^{2}	3.54 ± 2.04^{1}	0.00±0.00
3.54±0.00	q	3.74±0.20 ^b	3.54±0.00 ^a	4.87±1.33	0.00±0.00	0.00 ± 0.00	0.00 ± 0.00
7.08±0.00	5	$7.08{\pm}0.00^{2}$	8.25±1.18 ¹	9.43 ± 1.18^{1}	3.54 ± 0.00	$3.54{\pm}0.00$	0.00±0.00
2.36±1.18	ત્વ	3.54±0.00 ^b	2.36±1.18 ^b	2.36±1.18ª	0.00±0.00	0.00 ± 0.00	0.00 ± 0.00
4.72±1.18	-	7.08 ± 2.04^{1}	$3.54{\pm}0.00^{2}$	5.90±1.18	3.54 ± 0.00	$3.54{\pm}0.00$	0.00 ± 0.00
0.0±0.00	e	3.54±2.04 ^b	2.36±1.18 ^b	0.00±0.00	0.00±0.00	0.00 ± 0.00	0.00 ± 0.00
0.54 ± 0.00	1	4.72 ± 1.18^{1}	$3.54{\pm}2.04^{1}$	$3.54{\pm}0.00$	3.54 ± 0.00	$3.54{\pm}0.00$	0.00±0.00

a, b, values for low dose along the same column differently superscript, differ significantly (p<0.05). 1, 2 value for high dose along the same column differently superscript, differ significantly (p<0.05).

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Table 6: Mean $(\pm SE)$ Ni Concentration $(\mu g/g)$ in tissues of rabbits orally treated with doses of 2000 mg/kg body weight (low) and 4000 mg/kg body weight (high).

Days/ Froup	Brain	Heart	Kidney	Liver	Lungs	Skeletal Muscle	Blood
MO	114.40±4.92ª	125.00±11.36	130.68±15.03 ^a	136.36±0.00 ^b	113.64±15.03 ^a	153.41±19.68 ^a	221.59±17.05 ^a
High	147.70±15.00 ¹	147.70±22.70 ²	153.41±9.84 ¹	153.40±17.00 ¹	158.33 ± 6.10^{1}	182.49 ± 6.04^{1}	231.20±14.10 ¹
MO	90.91±5.68ª	125.00±11.36 ^a	96.59±5.68ª	130.68±5.68 ^a	107.95±5.68ª	113.64±15.03 ^b	204.55±35.48 ^b
High	147.70±15.00 ²	142.05±5.68 ²	153.40±17.00 ¹	147.70±24.80 ¹	141.30 ± 11.00^{1}	169.61±0.72 ¹	215.90 ± 24.80^{1}
Low	79.55±5.68ª	85.23±0.00 ^b	90.91±5.68b ^a	125.00±15.03 ^a	90.91±15.03ª	113.64±5.68 ^b	198.86±11.36 ^a
High	142.00±22.70 ¹	136.40±17.00 ¹	142.05±5.68 ¹	136.36 ± 0.00^{2}	113.60±11.40 ¹	142.00 ± 20.50^{1}	210.23±5.68 ¹
Low	79.55±5.68ª	85.23±0.00ª	85.23±0.00 ^b	119.312±17.05 ^a	79.55±5.68ª	85.23±0.00 ^b	159.85±14.47 ^a
High	130.70±31.601	130.68 ± 5.68^{2}	113.64 ± 5.68^{1}	130.70±15.00 ¹	113.60±11.40 ¹	119.32±0.00 ²	203.80±26.80 ¹
Low	73.86±5.68 ^b	73.86±5.68 ^b	73.86±5.68ª	107.62±14.72 ^a	68.18±0.00 ^b	68.18±0.00ª	142.05±5.68ª
High	113.62±5.70 ²	119,32±9.842	96.60±11.40 ¹	130.70±15.00 ¹	108.00 ± 11.40^{1}	119.32 ± 0.00^{2}	$198.90{\pm}11.40^{1}$
.Low	73.86±5.68 ^b	68.18±0.00ª	68.56±0.38 ^b	68.18±0.00 ^b	51.14±9.84ª	56.82±5.68 ^a	125.76±14.91 ^a
High	96.60±11.40 ¹	90.90±11.40 ²	90.91±5.68 ¹	130.68±5.68 ¹	102.27 ± 0.00^{2}	85.23±0.00 ²	163.50±12.80 ¹

a, b, values for low dose along the same column differently superscript, differ significantly (p<0.05). 1, 2 value for high dose along the same column differently superscript, differ significantly (p<0.05). **Table 7:** Mean $(\pm SE)$ Mn Concentration $(\mu g/g)$ in tissues of rabbits orally treated with doses of 2000 mg/kg body weight (low) and 4000 mg/kg body weight (high).

Days/ Group	Brain	Heart	Kidney	Liver	Lungs	Skeletal Muscle	Blood
1. Low	7.53±0.87ª	7.46±4.94ª	7.80±1.14ª	13.99±4.85ª	5.60±2.80ª	7.46±4.07ª	3.73±0.93 ^b
High	12.13 ± 3.73^{1}	10.26 ± 1.87^{1}	13.99±1.62	19.59±5.83	15.86±0.93	9.73±2.07 ¹	7.46±0.93 ²
2. Low	7.46±0.93ª	5.60±2.80ª	7.46±3.36ª	12.66±2.42 ^a	3.73±0.93 ^b	6.53±3.36 ^a	3.73±0.93 ^b
High	9.33±2.47 ¹	10.26 ± 5.19^{1}	12.13 ± 3.36^{1}	15.86 ± 3.36^2	14.93 ± 2.47^{1}	9.33±2.47 ²	7.46±0.93 ²
4. Low	4.66±1.87ª	4.66±1.87ª	4.66±2.47 ^a	12.26±0.87 ^b	3.73±1.87ª	2.80±0.00 ^b	2.80±1.62ª
High	9.33±0.93 ²	8.40±1.62 ²	7.46±0.93 ²	14.93 ± 0.93^{1}	12.13 ± 3.73^{1}	$8.40{\pm}1.62^{1}$	6.53 ± 0.93^{2}
6. Low	1.87±0.93ª	2.80±1.62ª	1.87 ± 0.93^{b}	6.53±1.87ª	1.87±1.87ª	0.93±0.93 ^b	0.00±0.00
High	5.60±4.27 ¹	3.73 ± 1.87^{2}	5.60 ± 3.23^{1}	8.40±1.62 ²	7.46±2.47 ¹	7.46±0.93 ²	6.53 ± 0.93^{2}
8. Low	0.00±0.00	0.93±0.93ª	0.00±0.00	5.60±1.62ª	0.93±0.93 ^b	0.00±0.00	0.00±0.00
High	$2.80{\pm}1.80^{1}$	2.80±0.00	4.66 ± 4.66^{1}	7.46±2.47 ²	$5.60{\pm}1.60^{2}$	2.80±1.62 ¹	0.00±0.00
10 Low	0.00±0.00	0.00±0.00	0.00±0.00	1.87±1.87ª	0.00±0.00	0.00±0.00	0.00±0.00
High	2.80±0.00	2.80±1.62 ¹	1.87 ± 0.93^{2}	4.66±2.47 ²	3.73±2.47 ²	$2.80{\pm}2.80$	0.00±0.00
a, b, '	values for low	dose along the	same column d	ifferently super	script, differ si	gnificantly (p	<0.05).

1, 2 value for high dose along the same column differently superscript, differ significantly (p<0.05).

The concentrations of Cu present in the edible tissues and blood of rabbits treated with the edible clay were comparatively low compared to elements like Ca, Mg and Fe. At day 1 post treatment the Cu levels were 7.08 ± 0.00 $\mu g/g$, 9.43+ 2.36 $\mu g/g$, 8.25+ 1.18 $\mu g/g$, 15.33+ 1.18, 6.90+ 0.18 $\mu g/g$, $3.54\pm0.00 \,\mu\text{g/g}$ and $0.00\pm100 \,\mu\text{g/g}$ in the brain, heart, kidney, liver, lungs, skeletal muscle and blood respectively in the animals treated with 2000 mg/kg ody weight dose. The animals treated with 4000 mg/kg body weight dose had the following levels brain, $11.79 \pm 1.18 \ \mu g/g$; heart, 9.59 ± 3.24 $\mu g/g$; kidney, 11.79+1.18 $\mu g/g$; liver, 16.31+1.09 $\mu g/g$; lungs, 10.16+0.00 μ g/g; skeletal muscle, 7.08 \pm 0.00 μ g/g and blood, 3.54 \pm 0.00 μ g/g (Table 5). Ten days post treatment there was absence of Cu in the brain, liver, lungs, skeletal muscle and blood in the rabbits treated with 2000 mg/kg body weight dose. In the case of blood, Cu was only obtained at day 1 post treatment following the administration of the edible clay at 4000 mg/kg body weight. The concentration of Ni in various tissues and blood following treatment with edible clay in rabbits is shown in table 6. The blood had the highest concentrations of Ni in both the 2000 mg/kg body weight and 4000 mg/kg body weight doses.

The concentrations of Ni present in the blood at day 1 post treatment were $221.59\pm17.05 \ \mu g/g$ and $231.20\pm14.10 \ \mu g/g$ for 2000 g/kg body weight and 4000 mg/kg body weight doses respectively. The concentrations of Ni in the various tissues were highest at day 1 post treatment. These concentrations were observed to decrease with time and at 10 days post treatment the levels were still detectable in the various tissues.

Detectable levels of Mn were observed in the various tissues and blood of treated rabbits except in the blood from day 6 post treatment for animals treated with 2000 mg/kg body weight and from day 8 in those treated with 4000 mg/kg body weight (Table 7). The levels of Mn in the various tissues and blood were not as high as those obtained for Ni, Ca, Fe and Mg. The concentrations of Mn in the rabbits treated with 4000 mg/kg body weight were higher than those treated with 2000 mg/kg body weight at p less than 0.05 level of significant.

Table 8: Mean (\pm SE) K Concentration (μ g/g) in tissues of rabbits orally treated with doses of 2000 mg/kg body weight (low) and 4000 mg/kg body weight (high) (Ogah et al; 2016).

I. Low 141.00 ± 36.76^a 78.00 ± 15.93^a 94.67 ± 7.26 114.00 ± 10.50 109.00 ± 18.33^a 177.67 ± 12.72^a High 152.70 ± 19.10 124.70 ± 16.20 106.30 ± 12.50 114.30 ± 10.20 1181.67 ± 8.28^1 L. Low 152.70 ± 19.10 124.70 ± 16.20 106.30 ± 12.50 $114.30\pm10.20^{\circ}$ $181.67\pm8.28^{\circ}$ L. Low 128.00 ± 21.73^a $72.33\pm7.69a$ 88.67 ± 14.75^a $89.67\pm2.90^{\circ}$ $94.00\pm6.00^{\circ}$ 153.33 ± 8.67^a A. Low 115.67 ± 7.12^a 69.00 ± 15.601 105.00 ± 4.73^1 $96.00\pm12.50^{\circ}$ $117.00\pm10.40^{\circ}$ 153.33 ± 8.67^a A. Low 115.67 ± 7.12^a 68.00 ± 8.74^a $88.33\pm4.63^{\circ}$ 81.33 ± 14.84^a $93.33\pm6.33^{\circ}$ $142.00\pm2.31^{\circ}$ A. Low 115.67 ± 7.12^a 68.00 ± 8.74^a $88.33\pm4.63^{\circ}$ 81.33 ± 14.84^a $93.33\pm6.33^{\circ}$ $142.00\pm2.31^{\circ}$ A. Low $115.67\pm7.71^{\circ}$ 68.00 ± 8.74^a $88.33\pm4.63^{\circ}$ 81.33 ± 14.84^a $93.33\pm6.33^{\circ}$ $142.00\pm2.31^{\circ}$ High $141.67\pm7.51^{\circ}$ 68.00 ± 8.74^a $88.33\pm4.63^{\circ}$ 81.33 ± 14.84^a $93.33\pm6.33^{\circ}$ $142.00\pm2.31^{\circ}$ G. Low 92.67 ± 11.68^a $54.67\pm1.45^{\circ}$ $84.33\pm1.76^{\circ}$ $77.67\pm1.86^{\circ}$ $76.33\pm5.90^{\circ}$ $141.33\pm6.00^{\circ}$ High $120.00\pm1.73^{\circ}$ $57.33\pm9.02^{\circ}$ $84.33\pm1.76^{\circ}$ $76.67\pm2.31^{\circ}$ $92.70\pm15.50^{\circ}$ $141.33\pm6.00^{\circ}$ No $41.06\pm1.73^{\circ}$ $57.33\pm9.02^{\circ}$ $57.67\pm6.17^{\circ}$ $57.67\pm6.12^{\circ}$ 57.72° $159.30\pm10.60^{\circ}$ No $114.70\pm13.40^{\circ}$	Days/ Group	Brain	Heart	Kidney	Liver	Lungs	Skeletal Muscle	Blood
High 152.70 ± 19.10 124.70 ± 16.20 106.30 ± 12.50 114.30 ± 10.20 113.30 ± 10.30^1 181.67 ± 8.28^1 2. Low 128.00 ± 21.73^a $72.33\pm7.69a$ $88.67\pm14.75a$ 89.67 ± 2.90^b 94.00 ± 6.00^b 153.33 ± 8.67^a High 143.70 ± 37.80^1 69.00 ± 15.601 105.00 ± 4.73^1 96.00 ± 12.50^1 110.00 ± 7.21^1 179.00 ± 10.40^1 4. Low 115.67 ± 7.12^a 68.00 ± 8.74^a 88.33 ± 4.63^b 81.33 ± 14.84^a 93.33 ± 6.33^b 142.00 ± 2.31^b High 115.67 ± 7.12^a 68.00 ± 8.74^a 88.33 ± 4.63^b 81.33 ± 14.84^a 93.33 ± 6.33^b 142.00 ± 2.31^b 6. Low 92.67 ± 11.68^a 54.67 ± 1.45^b 84.33 ± 1.76^b 77.67 ± 1.86^b 76.33 ± 5.90^b 141.33 ± 6.00^b High 120.00 ± 1.73^1 57.33 ± 9.02^2 $89.33\pm7/69^1$ 90.67 ± 7.31^1 93.70 ± 15.50^1 159.30 ± 11.60^1 8. Low 42.33 ± 22.52^a 49.00 ± 4.00^b 75.67 ± 6.17^b 75.00 ± 8.96^a 69.67 ± 12.55^a 120.00 ± 14.73^a High 114.70 ± 13.40^1 57.33 ± 9.02^2 $89.33\pm7/69^1$ 90.67 ± 7.31^1 93.70 ± 15.50^1 159.30 ± 10.60^2 9. Low 42.33 ± 22.52^a 49.00 ± 4.00^b 75.67 ± 6.17^b 75.00 ± 8.96^a 69.67 ± 12.53^a 120.00 ± 14.73^a High 114.70 ± 13.40^1 57.33 ± 9.02^2 89.00 ± 6.02^2 90.00 ± 5.57^2 153.30 ± 10.10^a 10 Low 19.33 ± 11.57^a 8.67 ± 6.17^a 67.67 ± 0.88^2 85.30 ± 12.70^1 70.33 ± 6.97^2 151.00 ± 5.77^2 High 100.00 ± 10.00^1 48.67 ± 1.45^2 86.67 ± 0.88^2 </td <td>1. Low</td> <td>141.00±36.76^a</td> <td>78.00±15.93ª</td> <td>94.67±7.26</td> <td>114.00±10.50</td> <td>109.00±18.33^a</td> <td>177.67±12.72ª</td> <td>46.90±5.18ª</td>	1. Low	141.00±36.76 ^a	78.00±15.93ª	94.67±7.26	114.00±10.50	109.00±18.33 ^a	177.67±12.72ª	46.90±5.18ª
2. Low 128.00±21.73 ^a 72.33±7.69a 88.67±14.75 ^a 89.67±2.90 ^b 94.00±6.00 ^b 153.33±8.67 ^a High 143.70±37.80 ¹ 69.00±15.601 105.00±4.73 ¹ 96.00±12.50 ¹ 110.00±7.21 ¹ 179.00±10.40 ¹ 4. Low 115.67±7.12 ^a 68.00±8.74 ^a 88.33±4.63 ^b 81.33±14.84 ^a 93.33±6.33 ^b 142.00±2.31 ^b 6. Low 92.67±11.68 ^a 54.67±1.45 ^b 84.33±1.76 ^b 77.67±1.86 ^b 76.33±5.90 ^b 141.33±6.00 ^b fligh 120.00±1.73 ¹ 57.33±9.02 ² 89.33±7/69 ¹ 90.67±7.31 ¹ 93.70±15.50 ¹ 159.30±11.60 ¹ 8. Low 92.67±11.68 ^a 54.67±1.45 ^b 84.33±1.76 ^b 76.53±5.90 ^b 141.33±6.00 ^b 114.1.070±1.73 ¹ 57.33±9.02 ² 89.33±7/69 ¹ 90.67±7.31 ¹ 93.70±15.50 ¹ 159.30±11.60 ¹ 8. Low 42.33±22.52 ^a 49.00±4.00 ^b 75.67±6.17 ^b 75.00±8.96 ^a 69.67±1.5.5 ^a 120.00±14.73 ^a 9. Low 114.70±13.40 ¹ 54.00±5.69 ² 87.69±3.28 ¹ 90.00±5.57 ² 159.33±10.10 ^a High	High	152.70±19.10	124.70±16.20	106.30±12.50	114.30±10.20	113.30 ± 10.30^{1}	181.67 ± 8.28^{1}	51.50±6.34
High 143.70 ± 37.80^1 69.00 ± 15.601 105.00 ± 4.73^1 96.00 ± 12.50^1 110.00 ± 7.21^1 179.00 ± 10.40^1 4. Low 115.67 ± 7.12^a 68.00 ± 8.74^a 88.33 ± 4.63^b 81.33 ± 14.84^a 93.33 ± 6.33^b 142.00 ± 2.31^b High 141.67 ± 7.51^1 64.70 ± 15.90^1 95.30 ± 13.40^2 94.00 ± 6.81^1 97.33 ± 6.33^b 142.00 ± 2.31^b 6. Low 92.67 ± 11.68^a 54.67 ± 1.45^b 84.33 ± 1.76^b 77.67 ± 1.86^b 76.33 ± 5.90^b 141.33 ± 6.00^b High 120.00 ± 1.73^1 57.33 ± 9.02^2 $89.33\pm7/69^1$ 90.67 ± 7.31^1 93.70 ± 15.50^1 159.30 ± 11.60^1 8. Low 42.33 ± 22.52^a 49.00 ± 4.00^b 75.67 ± 6.17^b 75.00 ± 8.96^a 69.67 ± 12.55^a 120.00 ± 14.73^a High 114.70 ± 13.40^1 54.00 ± 5.69^2 87.69 ± 3.28^1 89.00 ± 6.03^2 90.00 ± 5.57^2 153.30 ± 10.50^1 10 Low 19.33 ± 11.57^a 38.67 ± 6.17^a 67.67 ± 6.23^a 69.00 ± 10.07^a 66.67 ± 2.33^b 115.33 ± 19.10^a High 100.00 ± 10.00^1 48.67 ± 1.45^2 86.57 ± 0.88^2 85.30 ± 12.70^1 70.33 ± 6.97^2 151.00 ± 5.77^2	2. Low	128.00±21.73ª	72.33±7.69a	88.67±14.75ª	89.67±2.90 ^b	94.00±6.00 ^b	153.33±8.67 ^a	37.07±7.09ª
4. Low 115.67 ± 7.12^a 68.00 ± 8.74^a 88.33 ± 4.63^b 81.33 ± 14.84^a 93.33 ± 6.33^b 142.00 ± 2.31^b High 141.67 ± 7.51^1 64.70 ± 15.90^1 95.30 ± 13.40^2 94.00 ± 6.81^1 97.33 ± 8.33^1 159.30 ± 32.00^1 6. Low 92.67 ± 11.68^a 54.67 ± 1.45^b 84.33 ± 1.76^b 77.67 ± 1.86^b 76.33 ± 5.90^b 141.33 ± 6.00^b High 120.00 ± 1.73^1 57.33 ± 9.02^2 $89.33\pm7/69^1$ 90.67 ± 7.31^1 93.70 ± 15.50^1 159.30 ± 11.60^1 8. Low 42.33 ± 22.52^a 49.00 ± 4.00^b 75.67 ± 6.17^b 75.00 ± 8.96^a 69.67 ± 12.55^a 120.00 ± 14.73^a High 114.70 ± 13.40^1 54.00 ± 5.69^2 87.69 ± 3.28^1 89.00 ± 6.03^2 90.00 ± 5.57^2 153.30 ± 10.50^1 High 100.00 ± 10.00^1 54.00 ± 5.69^2 87.69 ± 3.28^1 89.00 ± 6.03^2 90.00 ± 5.57^2 153.30 ± 10.50^1 High 100.00 ± 10.00^1 8.67 ± 1.45^2 86.67 ± 0.88^2 85.30 ± 12.70^1 70.33 ± 6.97^2 151.00 ± 5.77^2	High	143.70 ± 37.80^{1}	69.00±15.601	105.00±4.731	96.00±12.50 ¹	110.00 ± 7.21^{1}	179.00 ± 10.40^{1}	50.12±5.531
High141.67±7.51164.70±15.90195.30±13.40294.00±6.81197.33±8.331159.30±32.0016. Low92.67±11.68a54.67±1.45b84.33±1.76b77.67±1.86b76.33±5.90b141.33±6.00bHigh120.00±1.73157.33±9.02289.33±7/69190.67±7.31193.70±15.501159.30±11.6018. Low42.33±22.52a49.00±4.00b75.67±6.17b75.00±8.96a69.67±12.55a120.00±14.73aHigh114.70±13.40154.00±5.69287.69±3.28189.00±6.03290.00±5.572153.30±10.50110 Low19.33±11.57a38.67±6.17a67.67±6.23a69.00±10.07a66.67±2.33b115.33±19.10aHigh100.00±10.00148.67±1.45286.67±0.88285.30±12.70170.33±6.972151.00±5.772	4. Low	115.67±7.12 ^a	68.00±8.74ª	88.33±4.63 ^b	81.33±14.84 ^a	93.33±6.33 ^b	142.00±2.31 ^b	36.97±7.24ª
6. Low 92.67±11.68 ^a 54.67±1.45 ^b 84.33±1.76 ^b 77.67±1.86 ^b 76.33±5.90 ^b 141.33±6.00 ^b High 120.00±1.73 ¹ 57.33±9.02 ² 89.33±7/69 ¹ 90.67±7.31 ¹ 93.70±15.50 ¹ 159.30±11.60 ¹ 8. Low 42.33±22.52 ^a 49.00±4.00 ^b 75.67±6.17 ^b 75.00±8.96 ^a 69.67±12.55 ^a 120.00±14.73 ^a High 114.70±13.40 ¹ 54.00±5.69 ² 87.69±3.28 ¹ 89.00±6.03 ² 90.00±5.57 ² 153.30±10.50 ¹ High 19.33±11.57 ^a 38.67±6.17 ^a 67.67±6.23 ^a 69.00±10.07 ^a 66.67±2.33 ^b 115.33±19.10 ^a High 100.00±10.00 ¹ 48.67±1.45 ² 86.67±0.88 ² 85.30±12.70 ¹ 70.33±6.97 ² 151.00±5.77 ²	High	141.67±7.51 ¹	64.70±15.90 ¹	95.30±13.40 ²	94.00±6.81 ¹	97.33±8.33 ¹	159.30 ± 32.00^1	48.77±6.25 ¹
High120.00±1.73157.33±9.02289.33±7/69190.67±7.31193.70±15.501159.30±11.6018. Low42.33±22.52a49.00±4.00b75.67±6.17b75.00±8.96a69.67±12.55a120.00±14.73aHigh114.70±13.40154.00±5.69287.69±3.28189.00±6.03290.00±5.572153.30±10.50110 Low19.33±11.57a38.67±6.17a67.67±6.23a69.00±10.07a66.67±2.33b115.33±19.10aHigh100.00±10.00148.67±1.45286.67±0.88285.30±12.70170.33±6.972151.00±5.772	6. Low	92.67±11.68ª	54.67±1.45 ^b	84.33±1.76 ^b	77.67±1.86 ^b	76.33±5.90 ^b	141.33±6.00 ^b	31.63±5.68 ^a
8. Low 42.33±22.52 ^a 49.00±4.00 ^b 75.67±6.17 ^b 75.00±8.96 ^a 69.67±12.55 ^a 120.00±14.73 ^a High 114.70±13.40 ¹ 54.00±5.69 ² 87.69±3.28 ¹ 89.00±6.03 ² 90.00±5.57 ² 153.30±10.50 ¹ 10 Low 19.33±11.57 ^a 38.67±6.17 ^a 67.67±6.23 ^a 69.00±10.07 ^a 66.67±2.33 ^b 115.33±19.10 ^a High 100.00±10.00 ¹ 48.67±1.45 ² 86.67±0.88 ² 85.30±12.70 ¹ 70.33±6.97 ² 151.00±5.77 ²	High	120.00±1.73 ¹	<i>5</i> 7.33±9.02 ²	89.33±7/691	90.67±7.31 ¹	93.70±15.50 ¹	159.30 ± 11.60^{1}	$37.80{\pm}1.30^{2}$
High 114.70 ± 13.40^1 54.00 ± 5.69^2 87.69 ± 3.28^1 89.00 ± 6.03^2 90.00 ± 5.57^2 153.30 ± 10.50^1 10 Low 19.33 ± 11.57^a 38.67 ± 6.17^a 67.67 ± 6.23^a 69.00 ± 10.07^a 66.67 ± 2.33^b 115.33 ± 19.10^a High 100.00 ± 10.00^1 48.67 ± 1.45^2 86.67 ± 0.88^2 85.30 ± 12.70^1 70.33 ± 6.97^2 151.00 ± 5.77^2	8. Low	42.33±22.52 ^a	49.00±4.00 ^b	75.67±6.17 ^b	75.00±8.96ª	69.67±12.55 ^a	120.00±14.73ª	31.43±6.77 ^a
10 Low 19.33±11.57 ^a 38.67±6.17 ^a 67.67±6.23 ^a 69.00±10.07 ^a 66.67±2.33 ^b 115.33±19.10 ^a High 100.00±10.00 ¹ 48.67±1.45 ² 86.67±0.88 ² 85.30±12.70 ¹ 70.33±6.97 ² 151.00±5.77 ²	High	114.70 ± 13.40^{1}	54.00±5.69 ²	87.69±3.28 ¹	89.00±6.03 ²	90.00±5.57 ²	153.30 ± 10.50^{1}	37.30 ± 0.36^{2}
High 100.00±10.00 ¹ 48.67±1.45 ² 86.67±0.88 ² 85.30±12.70 ¹ 70.33±6.97 ² 151.00±5.77 ²	10 Low	19.33±11.57 ^a	38.67±6.17ª	67.67±6.23ª	69.00±10.07 ^a	66.67±2.33 ^b	115.33±19.10 ^a	17.87±1.41 ^b
	High	100.00 ± 10.00^{1}	48.67±1.45 ²	86.67±0.88 ²	85.30±12.70 ¹	70.33±6.97 ²	151.00±5.77 ²	35.13±2.81 ²

a, b, values for low dose along the same column differently superscript, differ significantly (p<0.05). 1, 2 value for high dose along the same column differently superscript, differ significantly (p<0.05). **Table 9:** Mean $(\pm SE)$ Na Concentration $(\mu g/g)$ in tissues of rabbits orally treated with doses of 2000 mg/kg body weight (low) and 4000 mg/kg body weight (high) (Ogah et al; 2016).

				;	,		
Days/ Group	Brain	Heart	Kidney	LIVer	Lungs	Skeletal Muscle	Blood
1. Low	110.00±28.87 ^a	66.67±12.02ª	116.67±17.64 ^a	86.67±6.67ª	96.67±14.53ª	97.00±18.68ª	85.67±6.30 ^b
High	110.70 ± 20.30^{1}	85.70±34.90 ¹	116.70 ± 24.00^{1}	90.00±11.50 ¹	130.00 ± 10.00^{1}	120.30 ± 42.00^{1}	89.30±7.42 ¹
2. Low	110.00±15.28ª	66.67±18.56 ^a	113.33±12.02ª	73.33±12.02ª	94.67±17.94ª	95.00±13.23ª	84.40±3.65 ^b
High	110.00 ± 28.90^{1}	77.00±8.50 ²	110.00 ± 11.50^2	80.00±0.00	116.67±8.82 ¹	113.30±16.70 ²	89.00 ± 6.00^{1}
4. Low	90.00±11.55ª	63.33±28.48ª	110.00±15.28 ^a	66.67±3.33 ^b	90.00±23.09ª	93.33±33.33ª	83.57±5.70 ^b
High	103.33 ± 3.33^2	73.30±12.00 ¹	100.00 ± 15.30^{1}	80.00±11.50 ¹	110.00 ± 10.00^{1}	106.70 ± 23.30^{1}	88.83±8.991
6. Low	80.00±55.08ª	63.33±6.67 ^b	80.00±0.00	66.67±6.67ª	86.67±8.82 ^b	90.00±5.77 ^b	76.17±7.00 ^a
High	90.00±5.77 ²	73.30±20.30 ¹	100.00 ± 10.00^{1}	76.70±17.60 ¹	103.30 ± 18.60^{1}	100.00 ± 15.30^2	86.60±10.70 ¹
8. Low	76.67±8.82 ^b	60.00±17.32ª	76.67±12.02 ^b	63.33±18.56 ^a	70.00±10.00 ^a	86.67±6.67 ^b	63.17±11.59ª
High	90.00±5.77 ²	63.33 ± 8.82^{2}	93.67±3.18 ¹	66.67±6.67 ²	100.00 ± 17.30^{1}	100.00±17.30 ²	76.73±1.32 ²
10 Low	30.00±10.00 ^b	36.67±3.33 ^b	70.33±11.84 ^b	45.00±5.00 ^b	69.00±5.86 ^b	66.687±12.02ª	61.60±15.15 ^a
High	66.67±3.33 ²	46.70±12.00 ¹	83.33±3.331	63.30 ± 14.50^{1}	83.33±8.82 ²	96.70±20.30 ¹	69.30±4.84 ²
a, b,	values for low	v dose along th	e same column	differently suj	berscript, differ a	significantly (p	<0.05).

1, 2 value for high dose along the same column differently superscript, differ significantly (p<0.05).

The levels of K and Na in the various tissues and blood of rabbits treated orally with edible clay are presented in Tables 8 and 9 respectively. The brain, heart, kidney, liver, lungs, skeletal muscle and blood contained 141.00+ 36.76 µg/g, 78.00+ 15.93 µg/g, 94.67± 7.26 µg/g, 114.00± 10.50 µg/g, 109.00± 18.33 µg/g, $177.67+12.72 \ \mu g/g$ and $46.90+5.18 \ \mu g/g$ of K in rabbits treated with 2000 mg/kg body weight of the edible clay at day 1 post treatment. The rabbits treated with 4000 mg/kg body weight of the clay were observed to contain $152.70+19.10 \mu g/g$, 124.70+16.20μg/g, 106.32±12.50 μg/g, 114.30 ±10.20 μg/g, 113.30±10.30 $\mu g/g$, 181.67+8.28 $\mu g/g$ and 51.50+6.34 $\mu g/g$ of K at day 1 in the brain, heart, kidney, liver, lungs, skeletal muscle and blood respectively. The concentrations obtained in these tissues in the animals treated with 4000 mg/kg body weight were significantly higher than those from the animals treated with 2000 mg/kg body weight of the clay. These initial high concentrations obtained on day 1 were observed to decrease with time and on day 10 post treatment substantial amounts were still present in the various tissues and blood (Table 8).

Sodium concentrations were observed to be high in the tissues of rabbits treated with 2000 mg/kg body weight and 4000 mg/kg body weight of the clay (Table 9). In general the Na levels in the tissues and blood of the animals treated with 4000 mg/kg body weight of clay were higher than in those treated with 2000 mg/kg body weight dose for p less than 0.05 level of significant.

Lead and Cr were not detected in the tissues and blood of the rabbits given the clay orally at 2000 mg/kg and 4000 mg/kg body weight.



Fig. 1. Elimination rate constant of Ca in tissues of rabbits treated with doses of 2000 mg/kg body weight (low) and 4000 mg/kg body weight (high).



Fig. 2. Elimination rate constant of Mg in tissues of rabbits treated with doses of 2000 mg/kg body weight (low) and 4000mg/kg body weight (high).

ELIMINATION RATE OF METALS IN VARIOUS TISSUES (Ogah et al; 2016).



Fig. 4. Elimination rate of Fe in tissues of rabbits treated with doses of 2000 mg/kg body weight (low) and 4000 mg/kg body weight (high).



Fig. 5. Elimination rate of Cu in tissues of rabbits treated with doses of 2000 mg/kg body weight (low) and 4000 mg/kg body weight (high).



Fig. 6. Elimination rate of Ni in tissues of rabbits treated with doses of 2000 mg/kg body weight (low) and 4000mg/kg body weight (high).



Fig. 7. Elimination rate of Mn in tissues of rabbits treated with doses of 2000 mg/kg body weight (low) and 4000mg/kg body weight (high).



Fig. 8. Elimination rate of K in tissues of rabbits treated with doses of 2000 mg/kg body weight (low) and 4000 mg/kg body weight (high).



Fig. 9. Elimination rate of Na in tissues of rabbits treated with doses of 2000 mg/kg body weight (low) and 4000 mg/kg body weight (high).

The elimination rate of the elements present in the edible clay in the sampled organs and tissues are presented in figures 1 to 8. The rate of elimination of Ca from the brain of rabbits treated with the 2000 mg/kg body weight and 4000 mg/kg body weight of edible clay was higher than the rate of its elimination from other tissues and blood (Figure 1). The kidney had the lowest rate of Ca elimination with elimination rate constant of 9.15 and 16.80 ug/day in animals treated with 2000 mg/kg body weight and 4000 mg/kg body weight of edible clay respectively. Magnesium was slowly eliminated from the tissues and blood of the clay treated animals. The rates of elimination from the brain of rabbits treated with the clay at 2000 mg/kg body eight and 4000 mg/kg body weight were 5.53 μ g/day and 6.85 μ g/day respectively, (Figure 2). The elimination rate of eliminations for the blood were 17.6 μ g/day and 17.2 μ g/day in rabbits given the clay orally at 2000 mg/kg body weight and 4000 mg/kg body weight.

The rate of elimination of Fe were higher **at p<0.05** in the tissues of rabbits treated with 4000 mg/kg edible clay compared to those treated with 2000 mg/kg (Figure 3), except for the heart and the kidney. The elimination rate constant of Fe in the liver was 20.00 μ g per day in rabbits treated with 2000 mg/kg dose and 30.90 μ g per day in those treated with 4000 mg/kg.

The highest elimination rate of Cu in the liver was $1.52 \ \mu g \text{ per day}$ in the rabbits given 2000 mg/kg edible clay, and $1.43 \ \mu g \text{ per day}$ in those treated with 4000 mg/kg edible clay (Figure 4). The rate of elimination of Cu from the heart, liver, lungs, and skeletal muscle were higher **at p<0.05** in the animals treated with 2000 mg/kg compared to those treated with 4000 mg/kg dose. Cu was absent from the blood of rabbits given 2000 mg/kg of clay, the elimination rate constant, therefore could not be calculated.

Ni (Figure 5) shows the rate constant of elimination of Ni from the various tissues and blood. The skeletal muscle had the highest **at p<0.05** elimination rate constants of 9.74 μ g/day and 10.10 μ g per day respectively in animals given 2000 mg/kg and 4000 mg/kg edible clay.

The elimination rate constants of Mn in the various tissues of animals treated with the edible clay at 4000 mg/kg were higher **at p<0.05** than in those treated with the clay at 2000 mg/kg except the skeletal muscle (Figure 6) with the rate constants of elimination of 0.893 μ g/day and 0.858 μ g per day respectively.

The rate constant of elimination of K and Na are presented in figures 7 and 8. The rate constants of elimination of these two metals were higher **at p<0.05** in 2000 mg/kg treated rabbits for the following tissues-kidney, liver, lungs, skeletal muscle and blood, compared to those treated with 4000 mg/kg.

THE HALF LIFE (t_{1/2}) OF ELIMINATION OF METALS PRESENT IN THE EDIBLE CLAY IN THE VARIOUS ORGANS AND TISSUES OF TREATED RABBITS



Fig. 10. Elimination half-life of Ca in days in tissues of rabbits treated with doses of 2000 mg/kg body weight (low) and 4000mg/kg body weight (high) (Ogah et al; 2016).



Fig. 11. Elimination half life of Mg in days in tissues of rabbits treated with doses of 2000 mg/kg body weight (low) and 4000mg/kg body weight (high) (Ogah et al; 2016).



Fig. 12. Elimination half life of Fe in days in tissues of rabbits treated with doses of 2000 mg/kg body weight (low) and 4000mg/kg body weight (high).



Fig. 13. Elimination half life of Cu in days in tissues of rabbits treated with doses of 2000 mg/kg body weight (low) and 4000mg/kg body weight (high).



Fig. 14. Elimination half life of Ni in days in tissues of rabbits treated with dose of 2000 mg/kg body weight (low) and 4000 mg/kg body weight (high).



Fig. 15. Elimination half life of Mn in days in tissues of rabbits treated with doses of 2000 mg/kg body weight (low) and 4000 mg/kg body weight (high).



Fig. 16. Elimination half life of K in days in tissues of rabbits treated with doses of 2000 mg/kg body weight (low) and 4000 mg/kg body weight (high) (Ogah et al; 2016).



Fig. 17. Elimination half life of Na in days in tissues of rabbits treated with doses of 2000 mg/kg body weight (low) and 4000mg/kg body weight (high) (Ogah et al; 2016).

The half life $(t_{1/2})$ of elimination of metals present in the edible clay in the various organs and tissues of treated rabbits are presented in Figures 9 to 16. The half life of elimination of Ca was higher **at p<0.05** in the tissues of animals given the clay at 2000 mg/kg compared to those given the clay at 4000 mg/kg, except in the lungs (Figure 9). Figure 10 shows the half lives of Mg in the various tissues of treated rabbits. Skeletal muscle, lungs and heart have higher half lives in the animals treated with 4000 mg/kg dose, than those treated with 2000 mg/kg dose.

The kidney of the clay treated animals has the highest life **at** p<0.05 for Fe compared to the other sampled tissues (Figure 11). The half lives of Fe in the kidney were 0.054days and 0.059days for 2000 mg/kg and 4000 mg/kg doses respectively. Copper(Cu) generally appears to have the highest half lives in the various tissues of the metals analyzed (Figure 12).

The half lives of Ni in the heart, liver, lungs and blood of rabbits treated with the edible clay at 4000 mg/kg were higher than the half lives of those treated with 2000 mg/kg dose (Figure 13). The highest half life of Ni was recorded in the liver of 4000 mg/kg treated rabbits. The highest half life for Mn was obtained from the blood of rabbits treated with edible clay at 2000 mg/kg (Figure 14). This was followed by the half life in the lungs of rabbits given clay at 2000 mg/kg body weight.

The half lives of K and Na in the various sampled tissues of rabbits treated with the edible clay are shown in figures 15 and 16. The half lives of these two metals were higher in the kidney, liver, lungs, skeletal muscle and blood of animals given the clay at 4000 mg/kg compared to those given at 2000 mg/kg. The brain of rabbits treated with the clay at 2000 mg/kg had the least half life of Na.

DISCUSSION

The edible clay from Enyigba in Abakaliki, Ebonyi State from the observations made in the study contains different types of metals elements. The analyses showed that the elements found in the clay occurred in varying concentrations. These elements may be separated into three groups based on human nutritional requirements or health implications. The groups are:

- (i) essential metallic nutrients in animal and human nutrition. This group contains such elements as Na, K, Ca and Mg. The elements are required for growth and maintenance of health (Tietz *et al*, 1986).
- (ii) The trace elements in human and animal nutrition. This second group is known to consist of the following elements: Cu, Zn, Mn, Ni, Fe, Co and Cr.
- (iii) Trace heavy metals. This particular group is known to induce toxicity to both man and animals even in minute quantities. The elements placed in this group include Pb, Cd, As, Ba and Al (Tietz *et al*, 1986).

The essential trace elements were identified from human clinical experience and animal experiments. Those elements such as Ca, Mg, Na and K play essential roles in human and animal health and diseases have been previously highlighted (Khan, 1996; Ogugbuaja *et al*, 1997; Moses *et al*, 2002). Calcium apart from its importance in the maintenance of the skeletal structure of the body is also involved in blood coagulation, functioning of the heart, muscle and nerves and permeability of cell membranes (Odutola, 1992). Sodium is the major cation of the extracellular fluid and is responsible for 50% of the osmolality of the plasma or serum.

Potassium is the major cation of the intracellular fluid, and is very important in the maintenance of life (Radostits *et al*, 1997). Nutritionists have long recognized magnesium as an essential nutrient. Severe deficiencies of this element result in neuromuscular signs similar to those of eclampsia. Magnesium is also known to regulate calcium transport and so can play a significant role in bone metabolism (Sojka and Weaver, 1995). The concentrations of these essential trace elements found in the edible clay are within the safety limits reported by WHO (1996). The high levels of Ca and Mg in the clay may suggest that it may be compounds of calcite (CaCO₃) or dolomite CaMg(CO₃)₂ or a combination of the two compound to form lime stone.

The presence of Cu, Zn, Mn, Ni, Fe, Co and Cr which are trace elements in the clay are note worthy. Trace elements describe those elements present in the biological systems at very low concentrations. The functional role of trace elements is described in terms of their essential roles in nutrition and or toxicity (Kaplan and Pesche, 1989). Generally the concentrations of the trace elements obtained in this study are within the safety limit that has been reported by WHO (1996).

Many trace elements are known to influence various body functions and are extensively used both in chemotherapy and radiotherapy. Manganese is involved in normal skeletal growth and development, lipid metabolism, prevention of sterility, protein and nucleic acid metabolism and activation of enzyme functions (Blaurock-Busch, 1997). Elemental iron occurred in high concentration in the edible clay, although the concentration was within (WHO, 1996) acceptable level. Iron is important in haemoglobin production and is used to treat iron deficiency anaemia (Laurence *et al*, 1997). The concentration of Ni found in the clay appears to be high and worthy of note, since the human dietary requirement of Ni is 0.05 mg/day (Tietz *et al*, 1986).

Cobalt is essential component of vitamin B12, and is present in the adult mainly in the bone. Copper is important in some enzyme systems and also in the production of red blood cells. Copper deficiency may result in anaemia. It is also involved in the normal production of wool or fur (Tietz *et al*, 1986). Zinc is an essential element in nutrition of humans, animals and plants. It is required in the genetic make-up of every cell and is an absolute requirement for all biological reproduction (O'Dell, 2000). Its accumulation in the body is harmful. The concentrations of the above trace elements found in the clay are within the WHO (1996) acceptable levels for human and animal nutrition.

The heavy metals that occurred in the clay are within tolerable concentrations (Blanke and Walter, 1986; Pietroski and Coleman, 1980). These heavy metals are capable of inducing toxic effects even in small concentrations or doses. The toxic inorganic element form a much larger group and over 80 inorganic compounds have established toxicity which has been observed in industrial and environmental studies. The commonly investigated elements are lead, cadmium, mercury and arsenic, which are toxic even in low concentrations and induce serious problems of environmental pollution.

Furthermore, iron, nickel, manganese, zinc, cobalt, selenium and chromium may be toxic in high concentrations (Blaurock-Busch, 1997). The toxic inorganic elements found in the clay such as lead, arsenic, cadmium, barium and aluminium were found in low concentrations. This may be an indication of low degree of pollution in the area. Ebonyi state is not an industrialized area, hence it is expected that the production and disposal of these toxic metals will be too minimal to contribute to environmental pollution. The low clay concentration of toxic elements may also be due to the low deposits of these elements in the soil.

The results of the tissue kinetics study showed that most of the elements present in the edible clay obtained from Envigba, were readily absorbed following oral administration - of the clay. The mean peak blood and tissues concentrations of these elements occurred twenty four hours (one day) after the clay administration to rabbit at 2000 mg/Kg and 4000 mg/Kg doses. The elements obtained in high concentrations in the sampled tissues include Ca, Mg, Ni, Fe, K, and Na. Copper and Mn, occurred in low concentrations in the tissues and blood while Pb and Cr were absent. The high amounts of Ca, Mg, Ni, K and Na present in these tissues and blood may be due to increased uptake of these elements by organs and tissues involved. The high concentrations of these elements in tissues may also be due to the levels of these elements in the edible clay sample. The elemental analyses of the clay showed that Ca, Mg, Ni, Fe, K and Na were present in the clay at the following concentrations 249.00 ± 1.73 , 198.57 ± 0.90 , 89.34+ 0.69, 151.92+ 2.60, 84.33+ 2.73 and 71.96+ 3.63 mg/g respectively. It is therefore not surprising that those elements occurred in high concentrations in the sampled tissues. The low levels of Cu and Mn found in the various tissues following oral administration may be an indication of their low concentrations in the clay.

Biological systems are known to possess mechanisms for absorption, transportation, storage and excretion of elements. The administration of the edible clay orally may have resulted in the following within the gastro-intestinal tract. (i) The dissolution of the clay, (ii) Absorption of elements into the systemic circulation and (iii) Transfer of particles via the lymphatic system to lymph nodes or blood (Srivastava and Misra,1984). Elements transported by the blood are deposited in the body organs and tissues. The liver, kidney, brain, heart, skeletal muscle, and lungs constitute likely recipients of such elements (Moses *et al*, 2008).

This study has shown that the liver acted as the main accumulator of the elements. The observed high accumulations of these elements in the liver are probably related to their effective absorption from the gastro-intestinal tract and the ability of the liver to store them. Other studies have reported the capabilities of most organs, notably liver and kidney, to store large quantities of elements in the biological systems. The mechanisms entailed interactions between these elements with the intrinsic proteins in the organs and tissues, (King and Keen, 1999). The high concentrations obtained in the liver and kidney should be expected since the kidney is the primary organ of elimination and the liver the main organ of biotransformation (Baggot, 1977).

The presence of the sampled elements in the brain, heart, and skeletal muscle is interesting. Extensive amounts of elements such as Ca, Mg, Ni, K, Na and Fe were obtained in these tissues. The presence of these elements in the brain and skeletal muscle may be an indication of the ability of these elements to cross physiological barriers. The organs of the body, which are moderately supplied with blood such as heart and lungs, also contain high levels of Ca, Mg, Ni, Na, K, and Fe. This may be an indication of rapid distribution of these elements to the tissues which resulted in the increased uptake of these elements by these organs.

The presence of Ca, Mg, Ni, Fe, Na and K in the blood on day one post administration of the edible clay may be due to absorption of the elements from the stomach and or small intestine (Aliu, 2007). However, absorption may not explain the continued presence of these elements in the blood at day ten post administration.

The elements after absorption into the blood were distributed to organs and tissues of the body. Re-distribution of these elements may account for the prolonged presence of the elements in the blood, since the blood is the medium of transport of these elements in the body either to the tissues or to the excretory organs.

Plasma protein binding of the elements may also contribute to the prolonged presence of these metals in the blood. Although the study did not involve the plasma protein binding of these metals, the elements are known to bind to plasma albumin. High degree of plasma protein binding generally makes substances long acting because the bound fractions are not available for elimination, unless actively extracted in the liver or kidney. *Entero_hepatic* circulation of these elements distributed to the liver may be metabolized or eliminated through the bile into the intestine and thereafter reabsorbed into the blood thereby prolonging the duration of presence of the elements in the blood. (Aliu, 2007).

The high concentration of calcium in the brain of clay treated rabbits is worthy of note since in an earlier study it has been shown that elevated level of calcium in the brain decreases learning ability and also decreases brain serotonin turn over (Trulson *et al*,1986). However, with the recommended dietary calcium level of 1000 mg/day for human adults, the concentration of Ca in the clay may be considered to be low when given at the doses 2000 mg/kg and 4000 mg/kg this agrees with the result obtained by Michelle, (2011).

Copper and manganese were observed in this study to occur in low concentrations in the various sampled organs and tissues of rabbits given the edible clay at 2000 mg/kg and 4000 mg/kg doses. Twenty four hours (one day) after treatment with 2000 mg/kg Cu was not detected in the blood, while in the animals treated with 4000 mg/kg Cu was present at the concentration of 3.54+ 0.00 μ g/g. The highest concentration of Cu occurred in the liver of the rabbits given the clay at the doses of 2000 mg/kg and 4000 mg/kg. The high presence of Cu in the liver compared to other sampled tissues is in agreement with the observations of Braide and Anika (2007), who observed that very high concentrations of Cu may accumulate in the liver normally with no untoward effect on animals. Ochei and Kolhathar (2008) also reported increased Cu level in the liver in Cerrhosis and in Wilson's disease. Emmanuel (1989) was of the opinion that although high level of Cu occurred in the liver, substantial amount also occurred in the brain, heart and kidneys. The low concentration of Cu in the organs and tissues of the treated rabbits compared to elements such as Ca, Mg, Ni, Fe, Na and K may be as a result of limited absorption of Cu from the gastro-intestinal tract, due to low concentration of Cu in the edible clay. The absence of Cu in the blood of rabbits treated with 2000 mg/kg edible clay on day 1 post treatment and its absence from the blood of animals treated with 4000 mg/kg on day 2 post treatments may be an indication of rapid distribution of this element to the various organs and tissues of the body.

All the sampled organs and tissues in the present study were observed to contain manganese. The presence of this element in these tissues may be due to increased uptake of the element by organs and tissues involved. The low level of manganese in the various organs and tissues of treated rabbits compared to the elements like Ca, Mg, Ni, Fe, Na and K may be due to the level of the element in the edible clay. Manganese absorption from the gastrointestinal tract is known to be low and depends on the level of intake (Weigand and Kirchgessner, 1985).

Lead, chromium and aluminium from the result of this study were not detected in the sampled organs and tissues, which may be due to the low level of these elements in the edible clay, which affected their absorption from the gastrointestinal tract.

The half-lives of the elements found in the clay sample were indications of the lengths of time each of these elements will be retained in a particular organ or tissue. The higher the half-life of an element, the longer the persistence of that element in a particular tissue (Renwick, 1996), for example Ca with a half-life of 0.076day in kidney of rabbits treated with 2000 mg/kg of the edible clay orally will persist in the kidney more than Ca found in the liver of same rabbits with a half-life of 0.03days.

The slow elimination of Cu from the sampled tissues is substantiated by the fact that the half-life of elimination of Cu ranged between 0.594 and 1.543days after 2000 mg/kg clay administration, and 0.485 to 2.864days in tissues of animals given 4000 mg/kg edible clay. The enhanced half-life of elements may be a reflection of increased tissue sequestration, prolonged absorption and or presence of complexing compound in the clay. It also indicates how widely the elements were distributed or increased serum protein or tissue binding of the elements which limits their distribution to excretory organs.

CONCLUSIONS

Analyses of the clay obtained from Enyigba, Ebonyi State showed the presence of essential metallic elements, trace elements and toxic heavy metals in the clay. The edible clay was observed to have low toxicity when given orally, since the administration of 5000 mg/kg to rabbits orally produced no ill-effect. The administration of the edible clay to rabbits orally resulted in the absorption and distribution of Ca, Mg, K, Na, Cu, Ni, Fe and Mn to the various organs and tissues of the body. The absorbed elements were still present in the organs and tissues of the body 10 days post administration, showing that the elements were widely distributed in the body, with increased half-lives. Except for Ni the concentrations of all the other absorbed elements were within the WHO acceptable levels for human consumption per day. The levels of the trace heavy metals in the edible clay were low hence they were not absorbed into the body.

RECOMMENDATIONS

- (i) Histopathological (clinical) parameters is worthy of investigation.
- (ii) Multiple dose therapy to be applied with respect to possible accumulative effect of the identified metals.
- (iii) Ebonyi State Government and indeed the Federal Government of Nigeria should exploit the Enyigba clay deposit for the economic benefit of the state and the country at large.
- (iv) Screening of organic pollutants in the clay is of study interest.
- (v) Bioavailability of the mineral elements in the clay should be determined.
- (vi)The mineral constituent of other clay deposits across the country should be profiled

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CITATIONS OF PROF. SULEIMAN PHILIP IVOM OGAH

Professor Sule Philip Ivoms Ogah was born to Chief Ogah Nweke Ongele and Madam Orogwu Ogah on 15th July 1959 as the last male child of the family of 13 wives (his mother being the 12th wife) and 60 children in Obuegu Ohatekwe Amagu, Ikwo Local Government Area of Ebonyi State Nigeria.

He was less than 6 months old when his father died. He was entrusted into the care of his elder half-brother Alike Ogah. He started primary school rather late for his age in 1972. He was however, very brilliant in school and maintained first and second positions in his class throughout his primary education. He passed common entrance examination into Ezzikwo High School Amuzu, now Ezza High School where he passed General Certificate of Education in 1982. He worked briefly in Nigeria Defence Academy Kaduna as a very junior staff. He decided to relocate to Maiduguri to continue his education. He tried to go to school of nursing between 1984 and 1985 without success, even as he had been taking 1st places in all the entrance examinations into the schools of nursing. The reasons advanced for not taking him into school of nursing was that he has a limping limb which according to them would not allow him smooth handling of patients.

In 1986 he gained admission into the University of Maiduguri to read diploma in Science Education, a programme he completed with distinction in 1988. Immediately he finished his diploma, he got married to a beautiful girl, Hajia Mariam Ogah, a woman that gave him five wonderful children, two boys and three girls. He immediately went back through direct entry into the University of Maiduguri in 1989 for B.Sc. Chemistry which he completed in 1992. In 1995, Prof. S.P.I Ogah, as he is fondly called, went back to University of Maiduguri for M.Sc. Analytical Chemistry and completed the programme in 1998. His quest for Education and love for University Education took him back for his Ph.D. in Analytical Chemistry. He completed his Ph.D. programme in 2013. He taught Chemistry and Physics in University of Maiduguri Teaching Hospital.

Prof. S.P.I Ogah served as a Chemistry teacher and Principal of Mairi Islamic Secondary School, Maiduguri from 1991 to 2002, a job that saw him through his University Education as he was working and schooling at the same time. He left Mairi Islamic Secondary School in 2002 and joined Ebonyi State University as an assistant Lecturer where he rose to the rank of a senior lecturer. He left Ebonyi State University in December 2015 and joined Federal University of Lafia on merger of service having spent thirteen years of meritorious service with Ebonyi State University Abakaliki.

Prof. S. P. I. Ogah is a widely travelled person having visited some major cities in the world including London, Manchester, Newcastle, Addis Ababa, Cairo, France, Dubai and Makka.

He held various positions in the University, His Local Community, Ebonyi State and also at the National level. Among the positions held are:

- 1. Chairman Ebonyi State Muslim Pilgrims' Welfare Board Chairman Ebonyi State Council for Islamic Affairs date;
- 2. Member Representing South East in National Hajj Commission of Nigeria Committee for Madina Accommodation;
- 3. Member representing South East in 2015 National Hajj Commission of Nigeria feeding and accommodation Committee Makka and Madinna, Saudi Arabia;

- 4. Chairman South East forum of Chairmen and Secretaries of Muslim Pilgrims Welfare Boards;
- 5. Member National Hajj Commission of Nigeria Zonal Committee on Hajj Development Levy;
- 6. Member Nigeria Supreme Council for Islamic Affairs Education Committee;
- 7. Member Halal Certification Committee of Nigeria;
- 8. National Treasurer Amagu General Assembly;
- 9. Served as Chairman and member of various University Committees both in EBSU and FULafia;
- 10. He served as Head of Departments and most notably the first tenure staff to head Department of Chemistry in Federal University of Lafia.

Community Services:

Award: Prof. S. P. I. Ogah won many awards among which include:

- MTN Ebonyi State University 2014 best lecturer prize in "Surprise your Professor "campaign organized by MTN Nigeria Communications Limited ("MTN"), 11th February 2015.
- ii. Corporate and Media Africa Communications Limited; the Meritorious Diamond Award for National Development.

External examination:

Postgraduate external Examiner A. B. U. Zaria.

Visiting appointment:

Visiting Senior Lecturer University of Jos.

Accreditation/observation:

Prof. S. P. I. Ogah has participated in accreditation of academic programs.

Membership of professional bodies: He is a member of:

- 1. Chemical Society of Nigeria,
- 2. Fellow Institute of Chartered Chemists of Nigeria (FICCON).

He has over sixty publications renown in journals and attended a number of seminars, conferences and workshops. Prof. S. P. I. Ogah is actively involved in community services which among others include:

- 1. Scholarship at different levels of education to the needy in his community and beyond.
- 2. Completion of cement work and Electrification of the pavilion at the civic center at Obuegu Ohatekwe Amagu Playground.
- 3. Putting of street light along Ndechi, School, Azuokpuru roads in Obuegu Ohatekwe Amagu community.
- 4. Drilling of motorized borehole at Obuegu Ohatekwe.
- 5. Drilling of hand pump boreholes at:
 - i. Edukwu Agbakoro playground (ancestral home) of Amagu people;
 - ii. Anozie Nwandugo Ohatekwe playground; and
 - iii. Eguekpa Junction.
- 6. Attracted the construction of Akpina Bridge by Honourable Chinedu Ogah, OON.

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