

**INTERNATIONAL JOURNAL OF ADVANCES IN
PHARMACY, BIOLOGY AND CHEMISTRY****Research Article****Assessment of heavy metals (As, Cd, Cr, Cu, Ni, Pb
and Zn) in blood samples of Sheep and Rabbits from
Jimeta-yola, adamawa State, Nigeria****Charles Milam, Mohammed Buba one, Rifkatu Kambel Dogara and
Emmanuel Yusuf Yila**Department of chemistry, Modibbo Adama University of Technology Yola,
Adamawa State Nigeria.**ABSTRACT**

The aim of this study was to determine the concentration of heavy metals in blood samples of Sheep and rabbits from some selected villages of Jimete-yola, Adamawa State, Nigeria. Samples of blood were collected and prepared using standard procedures. The concentrations of Arsenic (As), Cadmium (Cd), Chromium (Cr), Copper (Cu), Nickel (Ni), Lead (Pb) and Zinc (Zn) were measured using Flame Atomic Absorption Spectrophotometry (AAS, Buck Scientific VGP 210 Model). The results showed that, metal concentrations in blood samples of both animals was in the order Zn > Cu > Cd > Cr > Ni > Pb > As. Mean concentration of cadmium in blood samples of Sheep exceeded the maximum tolerable limit set by the European Union. Also from the results of this study, high concentrations of metals were observed to accumulate in the blood samples of Sheep than rabbits. However, the statistical analysis of variance did not show any significant variation ($P < 0.05$). But prolonged exposure to these pollutants in the environment by the animals, it will pose a serious health hazards to animals and may lead to accumulation of this metals in humans and cause metal toxicity.

Keywords: Heavy metals; Blood; AAS.**INTRODUCTION**

The term pollution refers to the occurrence of an unwanted change in the environment caused by the introduction of harmful substances or production of harmful conditions. Many chemical toxins such as heavy metals are released into the atmosphere by many natural processes and human activities³¹.

As a source of meat and for economic benefit Sheep and rabbits plays an important role for human diets as well as an important source of a wide range of nutrients, but it may also carry certain toxic substances. Although the levels of these toxic substances in blood are generally low in some sheep and rabbits, chemical residues in blood may present a hidden but they represent a serious threat to public health. Because residues generally cannot be seen, smelled or tasted, they are difficult to detect²⁰. All heavy metals are toxic at certain levels of intake, as

lead and cadmium play no useful role and pose a risk for animals and human health. Heavy metals detected in muscle, liver and kidney of cattle by various studies.^{4, 6, 9, 10, 21} Many investigators were detected the levels of heavy metals in meat and edible offal (Liver and kidney) in camel^{4, 5, 9, 15}. Heavy metal causes many symptoms as hepato toxic effects and increase in liver function test parameters in lead⁽²⁾, while various cardiovascular disorders such as hypertension and cardio myopathy and both carcinogenic and anti-carcinogenic activities in cadmium were reported by Dong Z et al.¹⁴. The copper in high doses causes oxidative damage in liver in forms of granular degeneration, necrosis of hepatocytes¹⁶. Zinc also in high doses causes vomiting, cough, salivation, headache, malaise, fever and depression¹. Heavy metal contamination can be

transferred to animals through direct exposure, polluted water, and crops grown on irrigated sewage, industrial effluents, vehicle emission and dirty slaughter houses¹⁹. In recent times, there has been considerable interest in the levels of heavy metallic elements in foods because of their deleterious effect on human health. Apart from those communities exposed to high levels of pollution by industrial effluent or emission rich in heavy metals, it is evident that, for most individuals, food and diet are the most common source of these potentially toxic elements. These elements in food or drinking water amount to approximately 80% for cadmium, 40% for lead and 8% for mercury⁽¹²⁾. Metal contaminations in foods, especially in meat have been broadly investigated²⁷. These metals are known to bio accumulate in soil and have long persistence time through plants or animals²⁴. The risk associated with the exposure to heavy metals present in food product had aroused widespread concern in human health. The goals of animal production to produce quality products for increasingly health-conscious consumers and improving the quality of human life tend to be deterred by the presence and bioaccumulation of heavy metals in animal products.

Also due to the current trend of industrialization and urbanization, heavy metal pollution is increasing at alarming rates to reach disturbing levels thus leading to the contamination and deterioration of the environment. Since contamination with heavy metals is a serious threat because of their toxicity, bioaccumulation and bio magnifications in the food chain¹³, it becomes necessary to study the concentrations of toxic heavy metals in blood samples of Sheep and rabbits in order to assess the levels of exposure of the consumers to toxic metals and henceforth, maintain an ongoing knowledge on the levels of these metals both in the environment and in meat. The reported cases of heavy metal contamination in meat and other animal products is of great concern for both food safety and human health because of the toxic nature of some of these heavy metals at relatively minute concentrations.

Heavy metal is a general collective term which applies to the group of metals and metalloids with atomic density greater than 4 g/cm³ or that are 5 times greater than water¹⁸. Heavy metals are also called trace element due to their presence in trace (mg kg⁻¹) or in ultra-trace (1µg kg⁻¹) quantities in the environmental matrices. They are basically recovered from their ores by mineral processing operations^{24, 29}. Heavy metals such as iron, copper, manganese vanadium and tin occur naturally in the environment and could serve as plant nutrients depending on their concentrations. Mercury, silver,

cadmium, lead, chromium and many others that are indirectly distributed as a result of human activities could be very toxic even at low concentrations. These metals are non-biodegradable and can undergo global ecological circles.

Virtually, all metals can produce toxicity when ingested in sufficient quantities but the heavy metals are especially important because either they are so pervasive or they produce toxicity at such low concentrations. In general, heavy metals produce their toxicity by forming complexes or 'ligands' with organic compounds. These modified biological molecules lose their ability to function properly and results in malfunction or death of the affected cells. The most common groups involved in ligand formation are oxygen, sulfur, and nitrogen. When metals bind to these groups, they may inactivate important enzyme systems or affect protein structure.

MATERIALS AND METHODS

Blood collection

5.0 ml of blood samples of Sheep and rabbits were collected from their jugular vein, within the five (5) selected villages of Jimeta-yola, Adamawa State, Nigeria. Standard methods of collection were used as described by⁽²⁸⁾. Blood samples of 5 Sheep and 5 rabbits were collected from each of the villages in Jimeta-yola, making a total of 50 blood samples was collected, blood samples were put into 5ml capacity EDTA plastic bottles.

Digestion of blood samples

Blood samples were digested by the Conventional Wet Acid Method by adopting the method²³. With some modification. Accurately 1.0 mL of whole blood was taken into Pyrex flask separately. To this was added 6 mL of freshly prepared mixture of concentrated nitric acid and hydrogen peroxide [HNO₃ - H₂O₂] (4:2 V/V) and stood for 15 minutes. The flasks were covered with watch glass and then digested at 60 - 70 °C for 1 – 2 hours. The digests were then treated with 3 mL nitric acid and few drops of H₂O₂, while heating continued on hot plate at about 80 °C until a clear digested solution was obtained. The excess acid mixture was evaporated to semi-dry mass, cooled and diluted with 0.2 mL nitric acid. These were transferred into 100 mL volumetric flask and diluted to mark using triply distilled water. The worked-up samples were stored in polyethylene containers in a refrigerator at 4 °C prior to AAS analysis.

Elemental Analysis of Samples

The heavy metals concentrations in the digested samples were determined using the Buck scientific

VGP 210 model Atomic Absorption Spectrophotometer.

The principle of the Atomic Absorption Spectrophotometer involves absorption of light at a wavelength specific to that element by free atoms of the element. The Atomic Absorption Spectrophotometer was calibrated with standard solutions for each element then the sample solution was aspirated into the equipment and the reading was shown on the Atomic Absorption Spectrophotometer.

RESULTS

The mean concentrations of heavy metals in blood samples of Sheep subject with respect to various locations in Jimeta-yola, Adamawa State, are as presented in Figure 1. The levels of cadmium range from 0.017 to 0.072 mg/l; 0.014 to 0.041 mg/l Cr; 0.142 to 0.262 mg/l Cu; 0.013 to 0.035 mg/l Ni; 0.024 to 0.036 mg/l Pb and 0.575 to 1.603 mg/l Zn. whereas the Arsenic (As) was not detected in any of the samples analyzed.

Figure 2. shows the mean concentrations of the heavy metals in blood sample of rabbits with respect to different locations in Jimeta-yola, Adamawa State. The level of cadmium (Cd) ranges from 0.023 to 0.053 mg/l; 0.013 to 0.027 mg/l Cr; 0.072 to 0.123 mg/l Cu; 0.012 to 0.023 mg/l Ni; 0.012 to 0.036 mg/l Pb; 0.127 to 0.833 mg/l Zn, similarly the Arsenic (As) was not detected.

Figure 3. shows a comparison in the mean concentrations of heavy metals in blood samples of Sheep and rabbits. The cadmium (Cd) in blood samples of Sheep is 0.042 mg/l, while in the blood samples of rabbits a total concentrations of 0.04 mg/l was recorded. The mean concentration of Chromium (Cr) in blood samples of Sheep was 0.025 mg/l while 0.019 mg/l was observed in blood samples of rabbits. Similarly the mean level of Copper (Cu) in blood samples of Sheep is 0.185 mg/l was recorded and 0.101 mg/l were observed in blood samples of rabbits. Also the mean concentration of Nickel (Ni) is 0.025 mg/l in Sheep and 0.018 mg/l was recorded in blood samples of rabbits. For Lead (Pb), the mean concentration in blood samples of Sheep is 0.027 mg/l and 0.017 mg/l was recorded in blood samples of rabbits. And the mean level of Zinc (Zn) in blood samples of Sheep is 1.115 mg/l and 0.551 mg/l were recorded in blood samples of rabbits.

DISCUSSION

Heavy metals residues (As, Cd, Cr, Cu, Ni, Pb and Zn) in blood samples of Sheep and rabbits from selected villages of Jimeta-yola, Adamawa State, Nigeria.

The results of this study shows that heavy metals in blood samples of Sheep and rabbits from different villages of Jimeta-yola accumulate differently based on the exposure. However, the statistical analysis of variance indicated that there is no significant difference ($p < 0.05$). It was observed that Arsenic (As) was not detected in any of the blood samples analyzed, this is because the body clears arsenic from the blood within a few hours as reported³. The concentrations of metals were observed to be higher in the blood samples of Sheep subjects when compared to the concentrations of metals in the blood samples of rabbits. Such variation is because Sheep grazed freely on contaminated environment and drink water from ponds, stream, rivers and other possible contaminated water source. Sheep in the process are exposed to the high levels of heavy metals in the environment, these agree with that reported by⁽²⁵⁾. Cadmium (Cd) is a natural element in the earth's crust. It combines with other elements such as oxygen (cadmium oxide), Chlorine (Cadmium Chloride) or Sulfur (Cadmium Sulfate, Cadmium Sulfide). All soil and rocks, including coal and mineral fertilizers, contain some Cadmium. Production of Cu, Pb and Zn also produces Cadmium. Cadmium does not corrode easily and has many uses, such as in batteries, pigments, metal coatings and plastics⁷. Cadmium stays in the body for a very long time and can build up from many years of exposure to low levels. A balance diet can reduce the amount of Cadmium taken into the body from food and drink³. Blood Cadmium levels are mainly used for determining recent exposure to Cadmium rather than whole body burdens. According to the World Health Organization, blood concentrations $< 10 \mu\text{g}/\text{dl}$ are considered acceptable. The concentrations of cadmium in blood samples of Sheep were higher than this limit.

Chromium was detected in all the blood samples of Sheep and rabbits subjects from the entire locations. The highest concentration of chromium was observed in blood samples of Sheep from Namtari location, while the lowest concentration was observed in blood samples of rabbits from doubeli location. Chromium is an essential element helping the body to use sugar, protein and fat, and at the same time carcinogenic for organisms. Excessive amounts may cause adverse health effects¹ and can reduce how effective insulin is at controlling blood sugar and cause irritation itching and flushing. The Chromium level in blood samples was found to be lower than the tolerable level of 1.0 mg/l.

Copper (Cu) was also detected in all the blood samples analyzed from the entire locations. The highest concentration was observed in blood samples

of Sheep from doubeli location, while the lowest was observed in the blood samples of rabbits in Wurojabbe location. This could be attributed to the large amount of refuge dumpsite along the doubeli axis. Although copper is an essential metal, the high concentration in blood samples of Sheep could pose significant health risks to animals and in turn humans, as this metal can be toxic at high concentrations. Consumption of excessive copper (through diet) can result in liver and kidney damage as well as anemia⁸.

Nickel (Ni) the major man-made sources of nickel release are the combustion of coal and heavy fuel oil. Ni is found naturally in the earth's crust (in various forms such as nickel sulfides and oxides), and is present in small quantities in soils, aquatic environments and vegetation³⁰. There is little evidence that nickel compounds accumulate in the food chain. Ni is not a cumulative toxin in animals or in humans. Almost all cases of acute nickel toxicity result from exposure to nickel carbonyl¹¹. Nickel can cause respiratory problems and it is carcinogenic⁸. Nickel levels in this study, was not much higher than those reported by Akan JC et al. for meat and organs of Sheep in Maiduguri Metropolitan⁴. None of the blood samples of Sheep and rabbits was found to contain nickel concentrations above the permissible limit recommend by standard guideline.

Lead (Pb) is a toxic metal that has no known vital or beneficial effect on organisms and its accumulation over time in the bodies of animals and humans can cause serious ailment²⁸. The highest concentration of lead was recorded in the blood samples of Sheep as compared to rabbits. However, all are below the tolerable limit of 0.1 mg/l. Lead may enter the

atmosphere during mining, smelting, refining, manufacturing processes and by the use of lead containing products¹. The sources of lead contamination of livestock come from the air, water they drink and food they eat.

Zinc (Zn) concentrations in the blood samples of Sheep and rabbits from all the locations were extremely high as compared to the concentrations of other micronutrients that were considered in this study. However, none of the examined blood samples of Sheep and rabbits exceeded the permissible limit (40 ppm)¹⁷. Nearly similar results were recorded by⁽²⁶⁾. Zinc is an essential element in animals and human diet. Too little Zinc can cause problems; however, too much Zinc is harmful to animals and human health⁸.

CONCLUSION

In conclusion, the heavy metals concentrations of the subject from blood samples of Sheep and rabbits were in all cases below the reference values except for levels of Cadmium in Sheep which is above the reference value. This may pose a serious health hazard to animals. From the above results, the concentrations of all the heavy metals in the blood samples of Sheep were higher than those of the blood sample of rabbits; this is possible because; Sheep grazed freely on the contaminated environment and drink water from ponds, stream, rivers and other possible contaminated water source. Sheep in the process are exposed to the high levels of heavy metals in the environment.

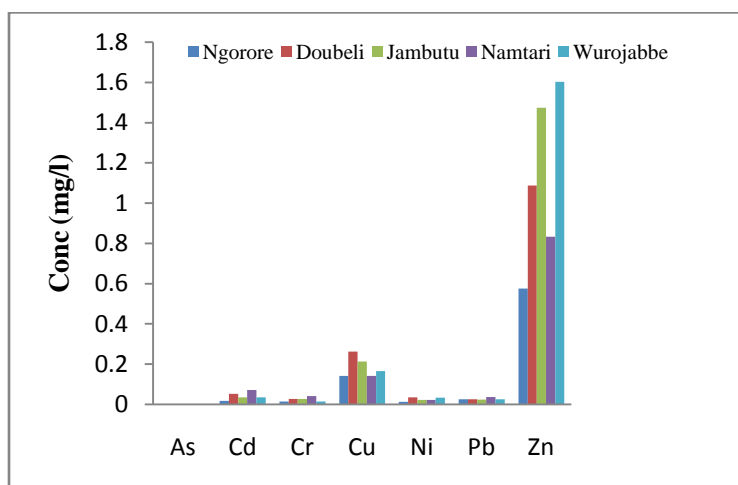


Figure 1

The mean concentrations (mg/l) of some heavy metals detected in blood samples of Sheep in some selected villages of Jimeta-yola, Adamawa State, Nigeria.

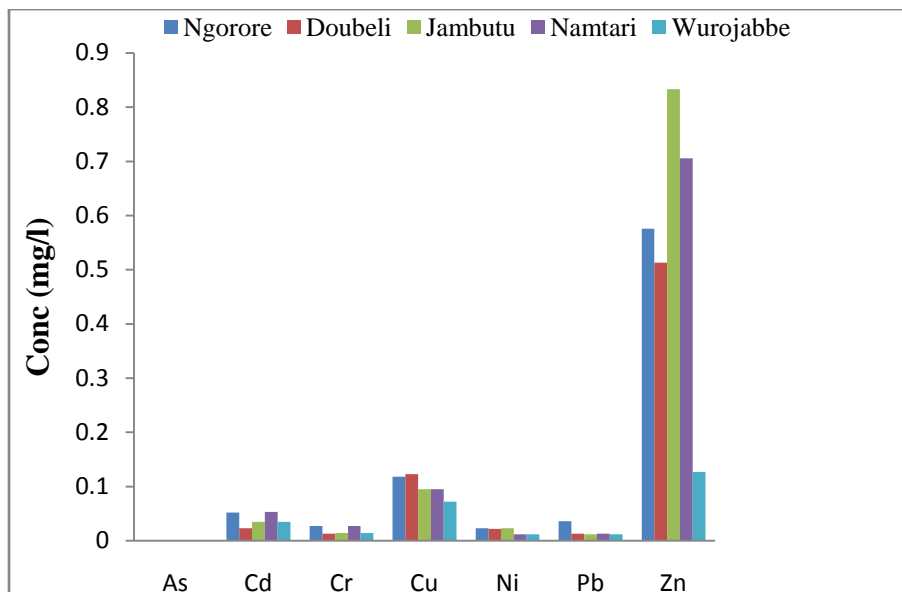


Figure 2

The mean concentrations (mg/l) of some heavy metals detected in blood sample of Rabbits in some selected villages of Jimeta-yola, Adamawa State, Nigeria.

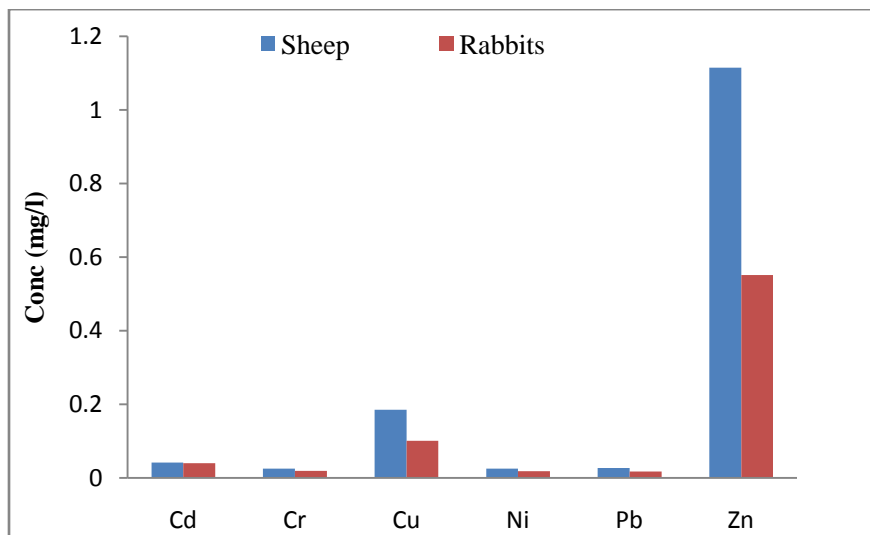


Figure 3

Comparison of heavy metal concentrations found in blood samples of Sheep and Rabbits from different locations in JimetaYola, Adamawa State, Nigeria.

REFERENCES

1. Abd El-Salam NM, Ahmad S, Basir A, Rais AK, Bibi A, UllahR, Shad AA, Muhammad Z, Hussain I. Distribution of heavy metals in the liver, kidney, heart, pancreas and meat of cow, buffalo, goat, sheep and chicken from Kohat market Pakistan. *J.Global Vet*, 2013; 2: 280-284.
2. Adeyemi O, Ajayi JO, Olajuyin AM, Oloyede OB, Oladiji AT, Oluba OM, Adeyemi O, Ololade IA, Adebayo EA. Toxicological evaluation of the effect of water contaminated with lead, phenol and benzene on liver, kidney and colon of Albino rats. *Food Chem Toxicol*, 2009; 47: 885-7.
3. Akan JC, Sodipo OA, Liman Y, Chellube ZM. Determination of Heavy Metals in Blood, Urine and Water Samples by Inductively Coupled Plasma Atomic Emission Spectrophotometer and Fluoride Using Ion-Selective Electrode. *J. Anal Bioanal Tech*, 2014; 5: 47-56
4. Akan JC, Abdulrahman FI, Sodipo OA, Chiroma YA. Distribution of Heavy Metals in the Liver, Kidney and Meat of Beef, Mutton, Caprine and Chicken from KasuwanShanu Market in Maiduguri Metropolis, Borno State, Nigeria. *J. Appl. Sci. Eng. Technol*, 2010; 2(8): 743-748
5. Akoto O, Bortey-Sam N, Nakayama SMM, Ikenaka Y, Baidoo E, Yohannes YB, Mizukawa H, Ishizuka M. Distribution of Heavy Metals in Organs of Sheep and Goat Reared in Obuasi: A Gold Mining Town in Ghana. *Int. J. Environ. Sci. Toxic. Res*, 2014; 2(2):81-89.
6. Ambushe AA, Hlongwane MM, McCrindle RI, McCrindle CME. Assessment of Levels of V, Cr, Mn, Sr, Cd, Pb and U in Bovine Meat. *S. Afr. J. Chem*, 2012; 65: 159-164.
7. ATSDR (2012): Toxicological profile for Cadmium. Agency for Toxic Substances and Disease Registry, Atlanta: US Department of Health and Human Services.
8. ATSDR, (2004): Agency for Toxic Substances and Disease Registry, Division of Toxicology, NE, Atlanta
9. Badis B, Rachid Z, EsmaB. Levels of selected heavy metals in fresh meat from cattle, sheep, chicken and camel produced in Algeria. *Ann. Res. Rev. in Bio*, 2014; 4: 1260-1267
10. Bala A, Saulawa MA, Junaidu AU, Salihu MD, Onifade KI, Magaji AA, Anzaku SA, Faleke OO, Musawa AI, Mohammed M, Muhammad LU, Ubangari MM, Ifende V. Detection of cadmium (Cd) residue in kidney and liver of slaughtered cattle in Sokoto Central Abattoir, Sokoto State, Nigeria. *J. Vet Ady*, 2012; 2: 168-172.
11. Barceloux DG, Barceloux D. Nickel. *Clin. Toxicol*, 1999; 37(2): 239-258.
12. Bennet FG. Modeling Exposure Routes of Trace Metals from Sources to Animal and Bothkins. *Environmental science earth as a living planet* (2nd edition) New York Longman Inc. Human Health. Springer-Verlag, Berlin, 1995; 2: 345-356.
13. Demirezen OK, Uruc H. Comparative study of trace elements in certain fish, meat and meat products, *Food Chemistry*. 2005; 32: 215-222
14. Dong Z, Wang L, Xu J, Li Y, Zhang Y, Zhang S, Miao J. Promotion of autophagy and inhibition of apoptosis by low concentrations of cadmium in vascular endothelial cells. *Tox. In Vit*, 2009; 23(1):105-108
15. Eltahir YE, Ali HM, Mansour MH, Mahgoub O. Serum mineral contents of the Omani racing Arabian camels (*Camelus dromedaries*). *J. Anim. Vet. Adv*, 2010; 9:764- 770
16. Emin OO, Hayati Y, Yasar E, Cevik TA, Gunfer T. The Effects of Copper Sulfate on Liver Histology and Biochemical Parameters of Term Ross Broiler Chicks. *Biological Trace Element Research*, 2010; 133: 335-341.
17. FAO/WHO. Joint FAO/WHO food standards program codex committee on contaminants in foods, 2011; 5(1): 64-89
18. Hawkes JK. Assessment of Health Risk from Exposure to Contaminated Soil. *J. Risk Anal*, 1997; 5(4):289-302.
19. Jukna C, Juckna V, Siugzdaite J. Determination of heavy metals in viscera and muscles of cattle. *Bulg J. Vet Med*, 2006; 9 (1): 35-41.
20. Khalafalla FA, Abdel-Atty NS, Mariam A, Abdel-Wahab MA, Ali FH, Omima I, Rofaida B Abo- Elsoud. Assessment of heavy metal residues in retail meat and offals. *J Am sci*, 2015; 11(5):50-54
21. Khalafalla FA, Ali, FH, Schwagede F, Abd-El-Wahab MA. Heavy metal residues in beef carcasses in Beni-Suef abattoir, Egypt. *Veterinaria Italiana*, 2011; 47: 351-361
22. Lenntech (2004): Water Treatment and Air Purification. Water Treatment, Publish by Lenntech, Rotterdams, Netherlands.
23. Memon AR, Tasneem GK, Hassan, IA, Nasreen S. Evaluation of Zinc status in whole blood and scalp hair of female cancer patients. *Clinica.Chimica.Acta*, 2007;10:59-64
24. Miranda M, Lopez-Alonso M, Castillo C, Hernandez J, Benedito JL. Effect of Moderate

- pollution on toxic and trace metal levels in calves from a polluted area of Northern Spain. *Environ Int*, 2005; 31:543- 548
25. Nwude DO, Okoye PAC, Babayemi JO. Heavy metal level in animal muscle tissue. A case study of Nigeria raised cattle. *Res. J. Appl. Sci*, 2010; 5:146-150
26. Purnama A, Zakaria F, Kusumaningrum HD, Hasan S. Selected minerals in meat of cattle grazing in mine revegetation Areas and safe consumption for human. *Food Science and Quality Management*, 2014; 40:100-106
27. Sharif LM, Dalal'eh R, Hassan M. Copper and mercury levels in Local Jordanian and Imported Sheep meat and organs. *Bulg. J. Vet Med*, 2005; 8 (4): 255-265
28. Solomon IP, Oyebadejo SA, Uyanga VA. Effect of Heavy Metals in feed in Dumpsite for age (Calapo – *Calopogonium mucunoides*) on Heamatological profile of Rabbits (*Oryctolagus cuniculus*). *Asian Journal of Biomedical and Pharmaceutical Sciences*, 2014; 4 (36): 35-43
29. United Nations Environmental Protection/Global Program of Action. 2004; why the Marine Environment needs Protection UNEP/GPA Coordination Office
30. USEPA (2000). Guidance for Assessing Chemical Contaminant Data for use in Fish Advisories: EPA 823-B-00-007. 3rd Ed. vol. 1, Office of Science and Technology and Office of water, USEPA, Washington, DC., USA
31. Bothkins D.B Keller (1995). Environmental science earth as a living planet (2nd edition) New York Longman Inc.