

Full Length Research Paper

Effects of selected well water sources at Ishiagu quarry site, Ebonyi State, Nigeria on basic haematological parameters in peripheral blood of albino rats

Victor Chibueze Ude¹, Uhegbu Friday Obinwa¹, Emmanuel Iroha Akubugwo¹, Ositadinma Chinyere Ugbo² and Amadike Eziuche Ugbo^{1*}

¹Department of Biochemistry, Abia State University Uturu, Nigeria.

²Department of Microbiology, Abia State University Uturu, Nigeria.

Received 07 July, 2012; Accepted 06 September, 2012

The haematological studies on selected well-water sources within two kilometers from Ishiagu quarry site were evaluated using albino rats. Haematological parameters were determined using standard methods. Results shows that haemoglobin concentration, haematocrit value, red blood cell count and neutrophytes decreased, while platelet count increased significantly at ($p < 0.05$) in test groups compared to control. Mean corpuscular volume and corpuscular haemoglobin concentration decreased non-significantly at ($p < 0.05$). Total white blood count, percentage lymphocytes, monocytes, and eosinophytes were not influenced by the long term intake of the well-water. These findings show that consumption of well-water around the quarry site in Ishiagu may lead to anaemia in the experimental animals and therefore may not be safe for human consumption. The toxicological implications of these findings are discussed.

Key words: Haemoglobin, ill-health, haematocrit value, anaemia, thrombosis.

INTRODUCTION

Groundwater is an important water resource in both urban and rural areas of Nigeria. Rural dwellers rely mostly on hand-dug wells for potable water supply as streams usually dry up in dry seasons (Adekunle et al., 2007). These water sources are under threat from human activities such as low level hygiene, industrial waste, agricultural practices and mining (Akujieze et al., 2003; Ikem et al., 2002; Livingstone, 2001; Nwaedozie, 2000). Sharma et al. (2004) reported that groundwater can be polluted by many hazardous pollutants like coloured dyes, nitrates, heavy metals, pesticides and fluorides. The river systems may also be excessively contaminated with heavy metals released from domestic, industrial, mining and agricultural effluents (Vander Oost et al., 2003).

Heavy metals such as lead, mercury, chromium, copper and cadmium together with other household chemicals and poisons can be concentrated in

groundwater supplies beneath landfills (Wenger and Rhyner, 1984). These contaminants have been reported to possibly cause growth retardation, and haemoglobin abnormalities (Hogson, 2004). Furthermore, blood cell responses are important indicator of changes in the internal and /or external environment of animals (Adeyemo, 2007). Witeska (1998) and Kanu et al. (2006) reported that the major symptom associated with ingestion of high quantity of copper, lead, cadmium and chromium is destruction of blood cells which is followed by anaemia.

Well waters in Ishiagu have been reported to be contaminated with heavy metals such as cadmium, lead, chromium, aluminum, copper, iron and zinc (Akubugwo et al., 2012). Some other contaminants detected in the well water above World Health Organization (WHO) permissible limits include nitrate, sulphate, fluoride and phosphate (Akubugwo et al., 2012). However, there has not been any report on effects of selected well water sources at Ishiagu quarry site Ebonyi State-Nigeria on basic haematological parameters or other infectious diseases in peripheral blood of albino rats.

*Corresponding author. E-mail: amasryal@yahoo.com. Tel: +2348053574147.

The presence of these contaminants in Ishiagu well water, the major sources of water for drinking and domestic uses necessitated this research. The aim of this study was to determine the effect of the well water sources on basic haematological parameters of albino rats.

MATERIALS AND METHODS

Ishiagu, the study area is located at Ivo Local Government Area of Ebonyi State, South-East Nigeria. Lead/zinc mines and quarry sites are cited in the area with the settlers predominantly peasant farmers. They experience dry season between October and April, and the rural dwellers in the area depend on hand-dug wells for the sources of water for drinking and domestic purposes during dry season because in dry season most of their streams dry up.

For this study, water samples used to feed the rats were collected at SETRACO quarry site, one kilometer from the quarry site and two kilometers from the quarry site while sachet water from Camp of Faith Okigwe was used as control.

Animal feeding and studies

Twenty (20) male adult albino rats (*Rattus norvegicus*) weighing between 70 to 80 g were purchased from the Animal House of Abia State University Uturu, Abia State. They were divided into four (4) groups (A - D) of eight rats each and were acclimatized for 14 days. The first group (group A) was placed on well water from Ishiagu granite quarry site, while the second group (group B) was placed on well water from a distance of one kilometer from the quarry site. The third group (group C) was placed on well water from a distance of two kilometers from the quarry site and the fourth group (group D) was placed on sachet water purchased from COFO waters along Enugu Port Harcourt Expressway, Okigwe Imo State which was used as control.

The experimental animals were maintained on commercial feeds (growers mash) purchased from Vital Feeds in Okigwe with water from the wells fetched from Ishiagu and COFO sachet water for sixty five (65) days *Ad libitum*. At the end of the feeding experiment the rats were sacrificed by anaesthetizing in a jar containing cotton wool soaked in chloroform. The blood samples were collected through cardiac puncture and dispensed into heparinized bottles for red blood cell (RBC) count, haematocrit (PCV), haemoglobin (Hb) white blood cell (WBC) count and differentials analysis.

The haemoglobin concentration was determined using cyanomethaemoglobin method as was described by Ramnik (1994) while haematocrit was determined using microhaematocrit method as was described by

Cheesbrough (2004). The red blood cell and white blood cell count were counted under light microscope with an improved Neubauer haematocytometry and differential leucocytes counts were carried out using Leishman stained blood smear. Platelets were determined using standard method as was described by Cheesbrough (2004).

Mean corpuscular haemoglobin (MCH), mean corpuscular volume (MCV) and mean corpuscular haemoglobin concentration (MCHC) were derived from the RBC, PCV and Hb as was described by Jain (1986).

MCV was calculated in femtoliters = $PCV/RBC \times 10$
 MCH was calculated in picograms = $Hb/RBC \times 10$ and
 MCHC was calculated in gram per deciliter = $(Hb \text{ in } 100 \text{ mg blood}/PCV) \times 100$

Data analysis

The data was analyzed using Analysis of Variance (ANOVA) and Duncan Multiple Range Test (DMRT) at $p < 0.05$ (Sokal and Rohlf, 1969).

RESULTS

Table 1 shows the changes in basic haematological parameters in peripheral blood of albino rats as a result of rats on daily consumption of well water from Ishiagu for 65 days. The haemoglobin concentration, haematocrit level and RBC count of the test group decreased significantly ($p < 0.05$) compared to the control. Consumption of well water samples from Ishiagu did not influence the number of total white blood as well as values of mean corpuscular volume, mean corpuscular haemoglobin and mean corpuscular haemoglobin concentration. The platelets ranged from $105.00 \pm 4.80 \times 10^9$ (in control group) to $127.00 \pm 47.61 \times 10^9$ cell/l (in the group supplemented with well-water fetched directly at Ishigau query site). The observed effect on the haematological parameters was detected even when water samples were taken two kilometres away of Ishiagu quarry site.

Table 2 show the changes in differential WBC count as a result of rats on daily consumption of well water samples from Ishiagu for 65 days *Ad libitum*. Percentage of neutrophils decreased only in animals which consumed well-water fetched at the Ishiagu quarry site. Consumption of well water fetched from one and two kilometre distance from Ishiagu quarry site did not influence neutrophiles number. The lymphocytes ranged between 56.00 ± 2.58 and $61.00 \pm 2.20\%$. The highest concentration observed in monocytes was $3.75 \pm 1.71\%$ in group A while group B had the lowest concentration of $2.75 \pm 0.5\%$. Eosinophiles range from $1.75 \pm 0.97\%$ to $2.50 \pm 0.58\%$ and basophiles were not detected in all the

Table 1. Changes in basic haematological parameters in peripheral blood of albino rats as a result of rats on daily consumption of well water from Ishiagu for 65 days.

Parameter	Group A	Group B	Group C	Group C
Haemoglobin (g/dl)	12.81 ± 1.13 ^a	13.69 ± 1.03 ^b	14.25 ± 1.44 ^c	14.88 ± 0.32 ^d
Haematocrit (%)	31.75 ± 0.96 ^a	33.00 ± 1.15 ^a	34.00 ± 1.41 ^a	35.50 ± 1.29 ^b
Platelets (×10 ⁹ cell/l)	127.00 ± 47.61 ^b	120.75 ± 46.80 ^b	116.25 ± 7.31 ^b	105.00 ± 4.80 ^a
WBC (×10 ⁹ cell/l)	8.18 ± 1.10 ^a	8.33 ± 0.78 ^a	7.18 ± 0.57 ^a	7.10 ± 0.80 ^a
RBC (×10 ¹² cell/l)	2.74 ± 0.22 ^a	3.00 ± 0.36 ^b	3.02 ± 0.09 ^b	3.21 ± 0.03 ^c
MCV (fl)	112.08 ± 3.20 ^a	111.01 ± 1.89 ^a	112.82 ± 3.11 ^a	113.59 ± 2.32 ^a
MCH (pg)	46.75 ± 1.50 ^a	45.63 ± 0.98 ^a	47.09 ± 1.20 ^a	48.36 ± 1.00 ^a
MCHC (g/dl)	40.35 ± 1.10 ^a	41.48 ± 0.68 ^a	41.11 ± 1.32 ^a	41.91 ± 0.88 ^a

The values represent mean ±SD (n=4). Values bearing different superscript in row vary significantly (p<0.05).

Legend: Group A = Rats on daily consumption of well water fetched from Ishiagu quarry site compound. Group B = Rats on daily consumption of well water fetched from a distance of one kilometer from Ishiagu quarry site compound. Group C = Rats on daily consumption of well water fetched from a distance of two kilometers from Ishiagu quarry site compound. Group D = Rats on daily consumption of COFO sachet water (control).

Table 2. Changes in differential white blood cell (WBC) counts in peripheral blood of albino rats as a result of rats on daily consumption of well water from Ishiagu for 65 days measured in percentage (%).

Parameter	Group A	Group B	Group C	Group C
Neutrophyles	33.25 ± 2.00 ^a	37.50 ± 1.92 ^b	38.88 ± 1.00 ^b	38.50 ± 2.91 ^b
Lymphocytes	61.00 ± 2.20 ^a	58.00 ± 2.16 ^a	57.00 ± 2.45 ^a	56.00 ± 2.58 ^a
Monocytes	3.75 ± 1.71 ^a	2.75 ± 0.58 ^a	3.00 ± 1.55 ^a	3.00 ± 0.82 ^a
Eosinophyles	2.00 ± 1.63 ^a	1.75 ± 0.97 ^a	2.00 ± 0.58 ^a	2.50 ± 0.58 ^a
Basophyles	0.00	0.00	0.00	0.00

The values represent mean ±SD (n=4). Values bearing different superscript in row vary significantly (p<0.05). Legend: Group A = Rats on daily consumption of well water fetched from Ishiagu quarry site compound. Group B = Rats on daily consumption of well water fetched from a distance of one kilometer from Ishiagu quarry site compound. Group C = Rats on daily consumption of well water fetched from a distance of two kilometers from Ishiagu quarry site compound. Group D = Rats on daily consumption of COFO sachet water (control).

blood films.

DISCUSSION

Changes in basic haematological parameters such as haemoglobin (Hb), haematocrit value, red blood cell (RBC) count, white blood cell (WBC) count, platelets and differentials are routinely used to determine stress associated with environmental, nutritional and/or pathological factors (Islam et al., 2004). Witeska and Kosciuk (2003) reported that heavy metals such as cadmium, chromium, nickel and lead might alter the properties of haemoglobin by decreasing their affinity towards oxygen binding capacity rendering the erythrocytes more fragile and permeable which probably may result in cell swelling, deformation and damage. More so, lead inhibits two enzymes, δ-aminolevulinatase dehydratase (δ-ALA-D) and haem synthetase (or ferrochelatase) that catalyzes the reaction for the

biosynthesis of haem group of haemoglobin (Ogwuegbu and Muhanga, 2005).

The decreased level of haemoglobin concentration, haematocrit value and red blood cell count in the experimental animals compared to control suggests that the pollutants in the well water might have destroyed the red blood cells; hence lower the haemoglobin and haematocrit level. The decreased level of haemoglobin concentration, haematocrit value and red blood cell count may be as a result of toxic effect of the well water on haemotopoietic precursors. It has been shown that lead reduces proliferation/differentiation of erythroid (BFU-E) precursors *in vitro* (van den Heuvel et al., 1999). Low concentrations of red blood cells and haemoglobin have been associated with anaemia (Islam et al., 2004). Islam et al. (2004) also reported that anaemia is related to the health effect of consuming leachate-contaminated groundwater. Fluoride treatment to rats resulted in reduction of total erythrocyte, haemoglobin percentage and haematocrit value (Sharma et al., 2004).

Vinodhini and Narayanan (2009) reported that the perturbation in the blood indices may be attributed to a defense reaction against toxicity through the stimulation of erythropoiesis. They also reported that the decrease in haematological indices proved the toxic effect of heavy metals that affect both metabolic and haemopoietic activities of *Cyprinia carpio L.*, a type of fish. Oladiji et al. (2004) and Agbafor et al. (2007) reported the same trend of result in studies carried out using polluted water to feed rats.

Platelets count was higher in the test group relative to control and may be due to the presence of heavy metals amongst other pollutants in the contaminated well waters (Adeyemi et al., 2007). Platelets or thrombocytes are the blood cell fragments that leads to the formation of blood clot and high level of platelets may arise from ingestion of high level of lead (Datta et al., 1994).

Nutritional and/or pathological factors arising from ingestion of leachate-contaminated groundwater may lead to increase in white blood cells and differentials. The nutritional factors may be due to the presence of hazardous chemicals in the groundwater samples while the pathological factors may arise from the bacteria present in the leachate-contaminated water (Adeyemi et al., 2007). The release of epinephrine during stress may cause a decrease in leucocyte count, which shows the weakening of the immune system (Adeyemo, 2007). The decreased number of neutrophils in animals supplemented with well water from the quarry region shows that they may be under stress which results in the release of epinephrine. Epinephrine is known to induce decrease of neutrophil number. The observed slight increase in white blood cell and lymphocytes may be traced to nutritional and/or pathological factors from ingestion of contaminated well water sources. The present result is similar to the findings of Oladiji et al. (2004).

In conclusion the significant decrease in haemoglobin concentration, haematocrit value and red blood cell may be an indication of anaemia while the increased platelet level shows ill-health thrombosis. This implies that the inhabitants of Ishiagu should be made to know the implication of using the contaminated well water sources for drinking, cooking and other domestic purposes by the community health officers and should finally be advised to discontinue its use for the above mentioned purposes.

REFERENCES

- Adekunle IM, Adetunji MT, Gbadebo AM, Banjoko OB (2007). Assessment of groundwater quality in a typical rural settlement in South West Nigeria. *Int. J. Environ. Res. Public Health.*, 4(4): 307-318.
- Adeyemi O, Oloyede OB, Oladiji AT (2007). Effect of leachate- contaminated groundwater on the growth and blood of Albino rats. *Internet J. Haematol.*, 3(2): 1-5.
- Adeyemo OK (2007). Haematological profile of *Clarias gariepinus* (Burchell, 1822) exposed to lead. *Turk. J. Fish. Aquat. Sci.*, 7: 163-168.
- Agbafor KN, Akubugwo EI, Ugbogu AE (2007). Toxicological evaluation of Okposi and Uburu salt Lake, Nigeria. *Int. J. Biotechnol. Allied Sci.*, 2(2): 186-189.
- Akubugwo EI, Ude VC, Uhegbu FO, Ugbogu OC (2012). Physicochemical properties and heavy metal content of selected water sources in Ishiagu Ebonyi State-Nigeria. *J. Bio. Environ. Sci.*, 2(2): 21-27.
- Akujieze CN, Coker SJ, Oteze GE (2003). Groundwater in Nigeria- a millennium experience- distribution, practice, problems and solution, *Hydrogeol. J.*, 1: 259-274.
- Cheesbrough M (2004). *District Laboratory practice in tropical counties*. University Press UK pp. 299-327.
- Data DJ, Mahenta JD, Dutta A (1994). Haematological values of local Duck of Assian in relation to sex. *Ind. Vet. J.*, 71: 1082-1084.
- Hogson, E. (2004). Water and soil pollutants. In textbook of modern toxicology. 3th Eds. John Wiley and sons inc. New Jersey. USA. p. 42-57.
- Ikem A, Osibanjo O, Sridhar MKC, Sobande A (2002). Evaluation of groundwater quality characteristics near two wastes sites in Ibadan and Lagos, Nigeria. *Water, Air Soil.*, 140 (1-4): 307-333.
- Islam MC, Lucky NS, Islam MR, Ahad A, Das BR, Raham MM, Siddivi MSI (2004). Haematological parameters of Fayoumi, Assil and Local chicken reared in Sychat region in Bangladesh. *Int. J. Poult. Sci.*, 3(2): 144-147.
- Jain NC (1986). *Schalm's veterinary haematology*. 4th Eds. Les au Febiger, Philadelphia, p. 1221.
- Kanu I, Achi OK, Ezeronye OU, Anyanwu EC (2006). Seasonal variation in bacterial heavy metal biosorption in water samples from Eziama River soap and brewery industries and the environmental health implication. *Int. J. Environ Sci. Tech.*, 3(1): 95-102.
- Livingstone DR (2001). Contaminant-stimulated reactive oxygen species production and oxidative damage in aquatic organisms *Mar. Pollut. Bull.*, 42: 656-666.
- Nwaedozi JM (2000). Environmental pollution- A case study of wastewater effluent parameters of some industries in Kaduna. *Afr. J. Environ. Stud.*, 1(1&2): 84-89.
- Ogwuegbu MOC, Muhanga W (2005). Investigation of lead concentration in the blood of people in the copperbelt province of Zambia. *J. Environ.*, 1(1): 66-73.
- Oladiji AT, Adeyemi O, Abiola OO (2004). Toxicological evaluation of the surface water of Amilegbe River using rats. *J. Nig. Soc. Exper. Biol.*, 16(2): 94-101.
- Ramnik S (1994). *Methods and interpretations in medical laboratory technology* 4th eds medical publishers Ltd India, pp.184-199.
- Sharma JD, Sharma MK, Agrwal P (2004). Effect of fluoride contaminated drinking water on Albino rats (*Rattus norvegicus*). *Asian J. Exp. Sci.*, 18(182): 37-46.
- Sokal RR, Rholf FJ (1969). *Practice of Statistics in*

Research San Francisco Freeman.

Van Den Heuvel RL, Leppens H, Schoeters GE (1999). Lead and catechol hematotoxicity *in vitro* using human and murine hematopoietic progenitor cells. *Cell. Biol. Toxicol.*, 15(2): 101-110.

Vander Oost R, Beyer J, Verneykebin NPE (2003). Fish bioaccumulation and biomarkers in environmental risk assessment. *Rev. Environ. Toxicol. Pharmacol.*, 13: 57-149.

Vinodhini R, Narayanan M (2009). The impact of toxic heavy metals on the haematological parameters in common carp (*cyprinus carpio*), Iran. *J. Environ. Health Sci. Eng.*, 6(1): 23-28.

Wenger RB, Ryner CR (1984). Optimal service region for solid waste facilities. *Waste Mgt. Res.*, 2(1): 1-15.

Witeska M (1998). Changes in selected blood indices of common carp after acute exposure to cadmium. *Acta. Vet. Brno.*, 63: 289-293.

Witeska M, Kosciuk B (2003). Changes in common carp blood after short term zinc exposure. *Environ. Sci. Pollut. Res.*, 3: 15-24.