

# Quality assessment of waterside river, ogbor hill, aba I: effect of three-point samples on some hematological parameters of wistar rats

## Abstract

This study determined some water quality indicators (biochemical oxygen demand, BOD, chemical oxygen demand, COD, dissolved oxygen, DO and total hardness, TH) of three-point samples of waterside River Ogbor hill Aba (WRO) and assessed the effect on rats hematology indices. The BOD, COD and OD (mg/L O<sub>2</sub>) respectively, of the Downstream-sample (21.52±0.01, 10.43±0.04, 31.41±0.01) was higher (p<0.05) than that for the Upstream (3.79±0.01, 7.61±0.01, 16.29±0.01), Midstream (3.20±0.00, 7.61±0.01, 14.84±0.02) and standard (3.00, NS, NS) while the TH (mg/L CaCO<sub>3</sub>) for the Downstream (20.04±0.01) was higher (p<0.05) than that for the Midstream (0.99±0.00) and Upstream (0.61±0.00) but lower (p<0.05) than that for the standard (500.00). The PVC (%) of the rats exposed to the various samples was lower (p<0.05) compared to the control but not significantly different (p>0.05) between the sample-treated groups. A similar trend was observed for the RBC ( $\times 10^9/L$ ) and HGB (g/L) counts. The PVC ( $\times 10^9/L$ ) of the groups exposed to the various samples was significantly different (p<0.05). However, while the platelet count of the rat groups exposed to Upstream and Downstream samples was lower (p<0.05), that of the Midstream sample was similar, compared to the control. The lower (p<0.05) WBC ( $\times 10^3/L$ ) of the rats in the various sample-treated groups compared to the control was higher in the Midstream-treated rats followed by Upstream-treated while least in Downstream-treated rats. Thus, the variously compromised quality of the three-point water samples from waterside River, Aba variously affected the studied hematological parameters of rats. Inherent self purification of river water may have resulted to the inconsistent observation which could be a pointer to underlying interplay of complex adverse biochemical responses, warranting further studies and interim urgent intervention measures to improve the quality status of WRO that serves as sole water source for a teaming residence.

**Keywords:** Haematological indices, Biological oxygen demand, Chemical oxygen demand, Dissolved oxygen

## Introduction

Out of the three natural water forms (liquid, solid and gaseous forms), liquid water is the commonly used and can be sourced from streams, lakes, rivers, ponds and wells.<sup>1</sup> Rivers are considered accessible and free sources of water. However, without regulation on use, rivers are predisposed to abuse and attendant overbearing challenge of contamination and pollution that could compromise water quality. Such danger of contamination, pollution and compromised water quality are the apparent fate of waterside River, Ogbor hill Aba, Abia State Nigeria (WRO). Waterside River in Ogbor hill area of Aba serves teaming settlers of cosmopolitan nature. And, the major activities in either bank of WRO that could predispose WRO to contamination and compromised water quality include hide and skin burning and related activities expected in abattoir, auto mechanic activities that could expose the river to lead and other heavy metals contamination, human wastes (faeces, urine) and general refuse disposal site as well as effluents from many factories (breweries, soap factories, soft drink factories) within the area. Sadly, WRO serves the teaming settlers who depend solely on it as water source for various water purposes. Such dependency by the inhabitants on WRO as sole water source is usually high during the dry season when the purpose for water sourced from WRO may include drinking apparently quite oblivious of the quality status and inherent health implications. And

little seems to have been done in terms of studying and documenting the quality status *vis a vis* highlighting the possible health implications following human use of WRO-sourced water. These warranted this study aimed at determining some principal indicators of water quality (biochemical oxygen demand, BOD, chemical oxygen demand, COD, dissolved oxygen, DO and total hardness, TH) of three-point samples from WRO and assessing the effects on some hematological parameters in Wistar rats. These indicators of water quality are basic to assessing the quality status of water<sup>2</sup> in relation to the significant these indicators play in assessing the composition, distribution, and abundance of aquatic organism.<sup>3,4</sup> Haematological indices are known bioindicators of health status hence useful in assessing physiological functions and/or dysfunctions in animals, including humans.

## Materials and methods

**Sample collection:** Three-point water samples were collected from the waterside River thus. The first point, Upstream, was at the bank under the bridge and close to the road. The second point, Downstream, was at the other bank far off the bridge and road but close to the residential and factory walls. The third point, Midstream, was at the mid point of the Upstream and Downstream. The water samples were collected using a properly labeled plastic containers and used without any treatment.

## Animal study:

- Animal procurement and exposure groups:** Twenty adult male Wistar rats (*Rattus norvegicus*) used in this study were procured from the animal house of the Faculty of Biological Sciences, University of Nigeria and Nsukka. The animals were acclimatized for 2 weeks and then randomized (based on weight) to four experimentation groups with sample size of five rats. Rats in the Group A (Control) were sham-exposed to 0.5 ml of clean sachet water while rats in Groups B, C and D were respectively exposed to 0.5 ml of WRO samples collected Upstream, Midstream and Downstream. The exposure was *per* oral, using a gavage, and daily for 4 weeks (twenty-eight days). Rats in all the groups were allowed free access to water and rat chow (commercial growers mesh feeds (Vital Feeds, Jos Nigeria).
- Sacrifice, blood sample collection and preparation:** After 4 weeks (28 days) exposure, the rats were sacrificed the next day after overnight fast by cardiac puncture technique<sup>5</sup> after cervical dislocation and the blood sample of the respective rats was collected individually into clean heparin tubes for hematological assessment.
- Ethical consideration:** This study considered and adhered to the standard ethical use of experimental animals. Throughout out the experimentation (acclimatization and exposure periods), all rats were housed at 25°C in stainless steel cages under normal daylight/dark cycle and humid tropical conditions. The rats were allowed free access to rat feed (Vital Feeds, Jos Nigeria) and tap water, and generally received humane care in accordance with the guidelines of the National Institute of Health, NIH, USA for ethical treatment of laboratory animals as approved by the various (departmental and college) ethical committees of Michael Okpara University of Agriculture Umudike, Nigeria.

**Determination of hematological indices:** The haematological indices of the rats exposed as in the study design were determined with Diatron Abacus Junior Haematology Analyzer that operated based on the principle of impedance to measure or count the volume and number of cells as described in a previous study.<sup>6</sup> Briefly, to a known dilution volume drawn through a small aperture a constant current was passed through the aperture from one side to the other to cause a change in resistance which generates a voltage pulse with the voltage amplitude (of the voltage pulse) proportional to the ratio of cell volume per aperture volume while the number of pulses represent the cells count.

**Determination of some water quality indicators:** The determination of DO, BOD and COD of the samples was essentially according to Winkler method as described in UNEP.<sup>7</sup> In particular, the determination of dissolved oxygen (DO) entailed allowing DO to react with iodine and form iodide and then calculating the DO after titration with a standard  $\text{Na}_2\text{S}_2\text{O}_3$  solution using the relation:

$$DO \left( \frac{\text{mg}}{\text{L } \text{O}_2} \right) = \frac{\text{Volume of } \text{Na}_2\text{S}_2\text{O}_3 \text{ used} \times \text{Normality} \times 8 \times 1000}{\text{Volume of sample}}$$

The determination of biochemical oxygen demand (BOD) involved measuring the difference in the oxygen concentration of the sample between zero day and after incubation at 20°C for 5 days.

$$BOD \left( \frac{\text{mg}}{\text{L } \text{O}_2} \right) = A_2 - A_1$$

where  $A_1$  is the Oxygen concentration of the sample before incubation while  $A_2$  is the Oxygen concentration of the sample after incubation. The determination of chemical oxygen demand (COD) involved digesting potassium dichromate, based on the principle that the COD of a sample that refluxed in high acidic solution with a known excess of potassium dichromate could be calculated on titrating the unreduced  $\text{K}_2\text{Cr}_2\text{O}_7$  (that remained after digestion) with ferrous ammonium sulphate using the relation:

$$COD \left( \frac{\text{mg}}{\text{L } \text{O}_2} \right) = A - B \times \text{No of FAS} \times 20 \text{ ml of sample}$$

where FAS, Ferrous ammonium sulphate; B, FAS used for sample and A, FAS used for the blank

However, the total hardness (TH) of the samples was determined using ethylenediaminetetraacetate (EDTA) titrimetric method.<sup>8</sup> This method was based on the principle that small amount of Dye Erichrome BlackT in aqueous solution containing calcium and magnesium ion at a pH of 10.00±0.01 turn wine red on addition of EDTA when all the magnesium and calcium had been completed, the dye turns blue. The total hardness was calculated using the relation:

$$TH \left( \frac{\text{mg}}{\text{L } \text{CaCO}_3} \right) = \frac{\text{Volume of titrant} \times 1000}{\text{Volume of sample}}$$

## Statistical analysis

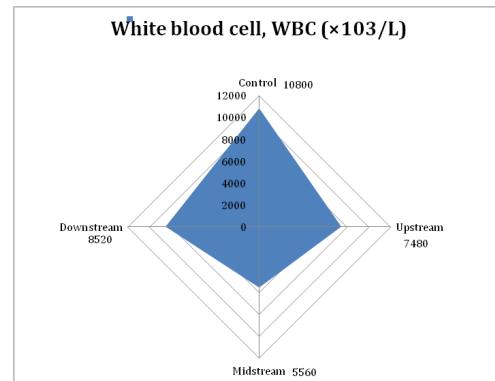
All collected data were analyzed by one way analysis of variance (ANOVA) using the statistical package for Social Science (SPSS version 17; SPSS Inc., Chicago, IL, USA). Results were presented as means ± standard deviation (Mean ± SD) at 95 % significance level ( $p < 0.05$ ).

## Results and discussion

The result of the determined water quality composition of the samples on Table 1 revealed that the BOD, COD and OD (mg/L  $\text{O}_2$ ) respectively, of the Downstream-sample (21.52±0.01, 10.43±0.04, 31.41±0.01) was higher ( $p < 0.05$ ) than that for the Upstream (3.79±0.01, 7.61±0.01, 16.29±0.01), Midstream (3.20±0.00, 7.61±0.01, 14.84±0.02) and standard (3.00, NS, NS) while the TH (mg/L  $\text{CaCO}_3$ ) for the Downstream (20.04±0.01) was higher ( $p < 0.05$ ) than that for the Midstream (0.99±0.00) and Upstream (0.61±0.00) but lower ( $p < 0.05$ ) than that for the standard (50.00). The effect of the three-point water samples on the hematological indices of the rats as shown on Table 2 revealed that the PVC (%) of the rats exposed to the various samples was lower ( $p < 0.05$ ) compared to the control but not significantly different ( $p > 0.05$ ) between the sample-treated groups. A similar trend was observed for the RBC ( $\times 10^9/\text{L}$ ) and HGB (g/L) counts. The PVC ( $\times 10^9/\text{L}$ ) of the groups exposed to the various samples was significantly different ( $p < 0.05$ ). However, while the platelet count of the rat groups exposed to Upstream and Downstream samples was lower ( $p < 0.05$ ), that of the Midstream sample was similar, compared to the control. The effect of the three-point water samples on the WBC count ( $\times 10^3/\text{L}$ ) of the rats as shown on Figure 1 revealed that the lower ( $p < 0.05$ ) WBC ( $\times 10^3/\text{L}$ ) of the rats in the various sample-treated groups compared to the control was higher in the Midstream-treated rats followed by Upstream-treated while least in Downstream-treated rats. This study determined some water quality indicators (biochemical oxygen demand, BOD, chemical oxygen demand, COD, dissolved oxygen, DO and total hardness, TH) of three-point samples of waterside River Ogbor hill Aba (WRO) and assessed the effect on rats hematological indices. The persistently

higher ( $p<0.05$ ) BOD, COD and OD (mg/L O<sub>2</sub>) composition in the Downstream-sample compared to those in the Upstream, Midstream and for standard was as expected since downstream seems far off to the perceived major points under the bridge and near the road. Higher BOD, COD and DO of the Downstream may indicate relatively good water quality of the Downstream sample. In particular, high DO as noted in the Downstream sample suggested lower pollution status.<sup>9,10</sup> Generally, BOD, the amount of dissolved oxygen required for the biochemical decomposition of organic compounds and the oxidation of inorganic materials, particularly for the Downstream sample was higher than the EPA standard and 2.13 mg/L O<sub>2</sub> for Sokoto River.<sup>11</sup> The higher ( $p<0.05$ ) TH (mg/L CaCO<sub>3</sub>) of the Downstream sample (20.04±0.01) compared to the Midstream (0.99±0.00) and Upstream (0.61±0.00) samples though lower ( $p<0.05$ ) than that for the standard (500.00) indicated that the samples were not, though the Downstream sample is more likely to be, hard. Hard water reduced mortality from heart diseases<sup>12</sup> thus apparently supporting the relatively lower contamination load of the downstream sample as noted with the BOD, COD and OD results of this study and the inference thereto. High DO as noted in the downstream sample suggested lower pollution status<sup>10</sup> hence better water quality. Nevertheless, organic matter contamination of the three-point samples of waterside River, Aba was suggested due to the relatively lower COD value observed in this study compared to that (30 mg/L O<sub>2</sub>) for Sokoto River.<sup>11</sup> Haematological studies are useful in the diagnosis of many diseases and in determining the physiological status of animals.<sup>6-13</sup> Decreased WBC, platelets, PVC are clinical indicators of acute toxicity.<sup>14</sup> In particular, while low platelet count, thrombocytopenia, generally indicates suppressed or reduced ability to fight pathogens hence reduced immunity, high platelet count recently predicts cancer.<sup>15</sup> In the present study, the PVC (%) of the rats exposed to the various samples was lower ( $p<0.05$ ) compared to the control but not significantly different ( $p>0.05$ ) between the sample-treated groups. A similar trend was observed for the RBC ( $\times 10^9/L$ ) and HGB (g/L) counts. The PVC ( $\times 10^9/L$ ) of the groups exposed to the various samples was significantly different ( $p<0.05$ ). However, while the platelet count of the rat groups exposed to Upstream and Downstream samples was lower ( $p<0.05$ ), that of the Midstream sample was similar, compared to the control. This may

suggest reduced immunity in either the Upstream or Downstream-treated rats. The lower ( $p<0.05$ ) WBC ( $\times 10^3/L$ ) of the rats in the various sample-treated groups compared to the control was higher in the Midstream-treated rats followed by Upstream-treated while least in Downstream-treated rats suggesting likelihood of underlying bone marrow suppression and dysfunction in the rats.<sup>16</sup> The result also suggested that Midstream rats may be anemic, at least in the long run. Elevated WBC, leukocytosis, and indicated anemic condition.<sup>17</sup> Groups exposed to the water samples had lower WBC compared to the control which may indicate underlying bone marrow suppression and dysfunction (Knight and Walter, 2001)<sup>16</sup>. Since all the samples significantly depleted the PVC, HGB, RBC and WBC counts whereas the Upstream and Downstream samples depleted ( $p<0.05$ ) the platelet count, the samples could be adjudged toxic on the hematological function of the rats. The apparent toxic status of particularly downstream sample irrespective of its relatively better water quality status as noted in this study may be attributed to the self-purification characteristic of rivers. Probably, the self-purification capacity of waterside River was able to achieve apparently better water quality status which did not altogether obliterate the toxic status on the hematological indices of the rats.



**Figure 1** Effect of three-point (Upstream, Midstream and Downstream) water samples from waterside river, Aba Abia State on the WBC count ( $\times 10^3/L$ ) of normal male albino rats.

**Table 1** Some water quality indicator compositions of three-point (Upstream, Midstream and Downstream) water samples from waterside river, Aba Abia state

Parameters	EPA standard <sup>#</sup>	Upstream	Midstream	Downstream
Biological Oxygen Demand, BOD (mg/L O <sub>2</sub> )	3	3.79±0.01	3.20±0.00	21.52±0.01
Chemical Oxygen Demand, COD (mg/L O <sub>2</sub> )	NS	7.61±0.01	7.61±0.01	10.43±0.04
Dissolved Oxygen, DO (mg/L O <sub>2</sub> )	NS	16.29±0.01	14.84±0.02	31.41±0.01
Total hardness, TH (mg/L CaCO <sub>3</sub> )	500	0.61±0.00	0.99±0.00	20.04±0.01

NS, Not stated; Value presented as mean±SD of triplicate determinations. Significant difference at  $p<0.05$ . #EPA.<sup>9</sup>

**Table 2** Effect of three-point (Upstream, Midstream and Downstream) water samples from waterside river, Aba Abia State on the haematological indices of normal male albino rats

Parameters	Control	Upstream	Midstream	Downstream
Packed cell volume, PVC (%)	53.00±1.41	51.20±1.92	50.20±1.78	51.20±1.30
Red blood cell, RBC ( $\times 10^9/L$ )	25.8.00±10.36	15.3.00±12.04	15.5.00±5.47	16.7.00±8.36
Haemoglobin, HGB (g/L)	17.36±0.73	15.06±0.37	14.18±0.26	14.68±0.25
Platelet count ( $\times 10^9/L$ )	250.00±7.07	236.00±15.16	250.00±10.00	216.00±15.16

Value presented as mean±SD of sample size, n=5 rats. Significant difference at  $p<0.05$

## Conclusion

Thus, the variously compromised quality of the three-point water samples from waterside River, Aba variously affected the studied hematological parameters of rats. Inherent self purification of river water may have resulted to the inconsistent observation which could be a pointer to underlying interplay of complex adverse biochemical responses, warranting further studies and interim urgent intervention measures to improve the quality status of WRO that serves as sole water source for a teaming residence.

## Acknowledgment

None.

## Conflict of interest

None.

## References

1. Okonko IO, Adejoye OD, Ogunmisi TA, et al. Microbial and physicochemical analysis of different water samples used for domestic purposes in Abeokuta and Ojota, Lagos State, Nigeria. *African Journal of Biotechnology*. 2008;7(5):617–621.
2. Adekunle IM, Adetunji MT, Gbadebo AM, et al. Assessment of groundwater quality in a typical rural settlement in southwest Nigeria. *Int J Environ Res Public Health*. 2007;4(4):307–318.
3. Mustapha MK, Omotosho S. An assessment of the physico-chemical properties of mow lake, Kwara State, Nigeria. *African Journal of Applied Zoology and Environmental Biology*. 2005;7:73–77.
4. Ikem A, Osibanjo O, Sridhar MKC, et al. Evaluation of groundwater quality characteristics near two wastes sites in Ibadan and Lagos, Nigeria. *Water Air and Soil Pollution*. 2002;140(1–4):307–333.
5. Lucas RL, Lentz KD, Hale AS. Collection and preparation of blood products. *Clin Tech Small Anim Pract*. 2004;19(2):55–62.
6. Uchendu OM, Egbuonu ACC. Ameliorative potentials of egg plant (*Solanum melongena* Linn) fruit ethanolic extract on monosodium glutamate-intoxicated rats' lipid profile, haematology and heart histology. *International Journal of Biochemistry Research and Review*. 2017;18(3):1–10.
7. United Nations Environmental Programme. United Nations Environmental Programme. *Analytical Journal of Epidemiology* 2004;153:453–464.
8. American Public Health Association. Standard methods of examination of water and wastewater 20th edn, USA: APHA; 1998.p. 348–350.
9. EPA. Parameters of water quality: Interpretation and standards. *Environmental Protection Agency*. Ireland; 2012. p. 52–57.
10. Ladokun O, Oni S. Physico-chemical and microbiological analysis of potable water in Jericho and Molete areas of Ibadan metropolis. *Advances in Biological Chemistry*. 2005;5:197–202.
11. Raji MIO, Ibrahim YKE, Tyler BA, et al. Physicochemical characteristics of water samples collected from River Sokoto, northwestern Nigeria. *ACS*. 2015;5(3):194–199.
12. EPA. Water quality in Ireland 2010–2012. Published by the Environmental Protection Agency, Ireland; 2015. p. 52–86.
13. Egbuonu ACC, Ogbu AE, Ijeh II, et al. Sub-chronic esculetin (6,7-Dihydroxy-Coumarin)-induced alteration in some haematological and serum parameters in normal male wistar rats. *Asian Journal of Biochemistry*. 2015;10(6):306–311.
14. Arsal SS, Esa NM, Hazilawati H, et al. Evaluation of acute, subacute and subchronic oral toxicity of Rhaphidophora decursiva (Roxb) Schott extract in male Sprague Dawley rats. *Journal of Medicinal Plant Research*. 2013;7(41):3030–3040.
15. Sylman JL, Mitrugno A, Tormoen GW, et al. Platelet count as a predictor of metastasis and venous thromboembolism in patients with cancer. *Convergent Science Physical Oncology*. 2017;3:1–13.
16. Knight AP, Walter RG. Plants affecting blood bracken fern poisoning. In: Knight AP, Walter RG editors. *A guide to plant poisoning of animals in North America*. USA: Teton New Media; 2001. p. 195–197.
17. Dinauer MC, Crates TD. Disorders of phagocytes function and number. In: Hoffman R, et al. (Eds.), *Hoffman Hematology: Basic Principles and Practice*. 5th edn, Churchill Livingstone, USA; 2008. p. 1–2560.