International Journal of Fisheries and Aquatic Studies 2015; 3(1): 09-14

ISSN: 2347-5129
(ICV-Poland) Impact Value: 5.62
(GIF) Impact Factor: 0.352
IJFAS 2015; 3(1): 09-14

www.fisheriesjournal.com
Received: 05-06-2015
Accepted: 08-07-2015

Kefas M
Department of Fisheries, Modibbo Adama University of Technology, Yola, P.M.B 2076, Adamawa State, Nigeria.

Abubakar KA
Department of Zoology, Modibbo Adama University of Technology, Yola, P.M.B 2076, Adamawa State, Nigeria.

Ja'afaru A
Department of Zoology, Modibbo Adama University of Technology, Yola, P.M.B 2076, Adamawa State, Nigeria.

Correspondence
Kefas M
Department of Fisheries, Modibbo Adama University of Technology, Yola, P.M.B 2076, Adamawa State, Nigeria.

Haematological indices of tilapia (*Oreochromis niloticus*) from Lake Geriyo, Yola, Adamawa State, Nigeria

Kefas M, Abubakar KA, Ja'afaru A

Abstract
The increases in agricultural activities and rapid urban development which occur along the lake have resulted in serious pollution problems. A toxicological study was carried out to ascertain the effects of aquatic pollutants in the blood of O. niloticus at the Lake Geriyo for a period of eighteen months. The Haematological indices such as Haemoglobin (Hb), Packed cell volume (PCV), Red blood cell (RBC), White blood cell (WBC),Mean corpuscular volume (MCV), Mean corpuscular haemoglobin (MCH), Mean corpuscular haemoglobin content (MCHC) were investigated. The mean value recorded for Haemoglobin were 4.05±0.808 to 10.43±0.817g/dL, PCV ranged from 10.12±0.008 to 24.90±3.266%, RBC ranged from 0.81±0.008 $x10^6$/mL to 1.82±0.016 $x10^6$/mL,WBC ranged from 108.67±8.206 $x10^3$/mL to 238.00±39.171$x10^3$/mL, while the MCV were 120.33±7.348 to 167.67±8.525 (fL), MCH ranged from 45.03±8.165 to 63.21±8.165 (pg/cell), MCHC were 28.47±0.327 to50.32±4.090(g/dL). All the haematological parameters shows significant different in variation between months (P<0.05).This study indicates that there was an alteration in haematological profile in O. niloticus blood which may cause biochemical dysfunction in this specie. These results provide a useful tool in monitoring the condition and state of health of fish by knowing the normal value with respect to their responses to stress which affects body metabolism.

Keywords: *Haematolocal parameters, Lake Geriyo, Oreochromis niloticus.*

1. Introduction
Water pollution has become a menace in recent times, causing a great damage to the aquatic ecosystems. Waste from farms, industries and domestic uses gradually find their way into the aquatic environment. Most of the water bodies have become polluted due to haphazard and extravagant pouring of wastes into them and making it unfavourable for aquatic organism (Chindah *et al.*, 2008) [12]. Pollution of the aquatic environment by organic and inorganic chemicals has been considered a major threat to the aquatic organisms including fishes. The agricultural drainage water containing pesticides and fertilizers and effluents of industrial activities and runoffs in addition to sewage effluents supply the water bodies (ECDG, 2002). Aquatic contamination by industrial and domestic sewage outlets are a constant sources of public health concern. The series of contamination may vary between organic pollutant such as polycyclic aromatic hydrocarbons from oil explorative activities, resin acids, and heavy metals from industries and also alkylphenols deriving from domestic activities (Chindah 2004) [11]. The release of chemicals into the aquatic environment results in some changes, which may threaten functional attributes, the integrity and existence of aquatic organisms, especially fish (*Ayoola et al, 2014*) [8]. Recently, haematological parameters have become promising biomarkers in measuring the effects of chemical pollutant in fish. Blood samples can regularly be obtained from test organisms, thus allowing the use of non- destructive approach in effecting assessment (Akinrotimi *et al.*, 2010) [4]. Typically, haematological parameters are non-specific in their responses towards chemical stressors. Nevertheless, they may provide important information in assessment studies, by providing an indication as to the general physiology and health status of the organism under investigation (Ayoola *et al.*, 2014) [8].
Several researcher have investigated the toxicity, uptake and tissues distribution and haematological changes of pollutants in fish (Chindah *et al.*, 2008, Akinrotimi *et al.*, 2007) [12, 5], and the use of hematological techniques in fisheries research is growing rapidly, as it is very important in toxicological research which result in monitoring and predicting health conditions of the fish (Ayoola *et al.*, 2014, Akinrotimi *et al.*, 2009) [8, 6]. Since fish are so intimately associated with the aqueous environment, the blood will reveal measurable physiological

changes in the fish more rapidly than any physiological assessment parameters (Ezeri *et al.*, 2004) [17]. Pollutants such as herbicides, pesticide and industrial effluent are known to alter the haematological indices of fish (Ayoola *et al*., 2014; Akinrotimi *et al.*, 2007) [8, 5]. Long term exposures of fish to effluents and sewages have been reported to alter haematological parameters by disrupting haematopoiesis, consequently resulting in anemia condition (Nikinmaa and Oikari, 1992; Ellis *et al.*, 2003) [24, 16].

Fish is in close contact with their environment, and are very susceptible to physical and chemical changes which may be seen in their blood components (Ayoola *et al*., 2014) [8]. Haematology is used as an index of fish health status in many fish species to detect physiological changes following different stress condition such as exposure to pollutants, diseases, heavy metals, hypoxia etc. According to Svobodova *et al*., (1996) [26] study of haematological parameter are carried out on the fish to ascertain the normal range of blood parameter, find out the variation with age, sex, season, and determine the effects of disease condition on the fish. This study is aimed at assessing haematological indices of Tilapia specie (*Oreochromis niloticus*) of the Lake Geriyo.

2. Materials and Methods

2.1. Description of Study Site

Lake Geriyo is located at the outskirts of Jimeta- Yola metropolis on the north-west region (Longitude $12^{o} 25^{I}$ E and between latitude $9^{o} 81^{I}$ N and $9^{o} 17^{I}$). It has a high level of 750ha and low level of 200ha. Storage at level about 7,500,000$cm^{2.}$ The area available to fisheries development is about 250ha; consequently, most of the settlers around the lake are fishermen, (upper Benue River Basin Development Authority, 1985) [28].

Lake Geriyo is a natural lake that started as a small gulley, but later filled with water from rains and some influx from River Benue. The lake came into recognizable existence in 1950. Initially, the lake was not used for fishing. It has now become a major fishery, with fishing activities taking place all year round. It is also a major source of water for irrigation, during the dry season farming that takes place around the lake. The level of the lake is reasonably constant with regards to the movement of water in and out. This has given rise to a stable growth of water plants that give the basin the appearance of a typical lake.

The lake experiences two seasonal period: the rainy and dry seasons. The rainy season starts in the months of May and last till October and is characterized by heavy down pour which may sometimes result in extensive floods. The dry season on the other hand is from late October to April and is characterized by the cold dusty dry winds of December and January (Harmattan) and intense heat of February, March through April. Atmospheric temperature can be as low as 20^{0} c in December and January and as high as 40^{o} c in March and April (upper Benue River Basin Development Authority, 1985) [28].

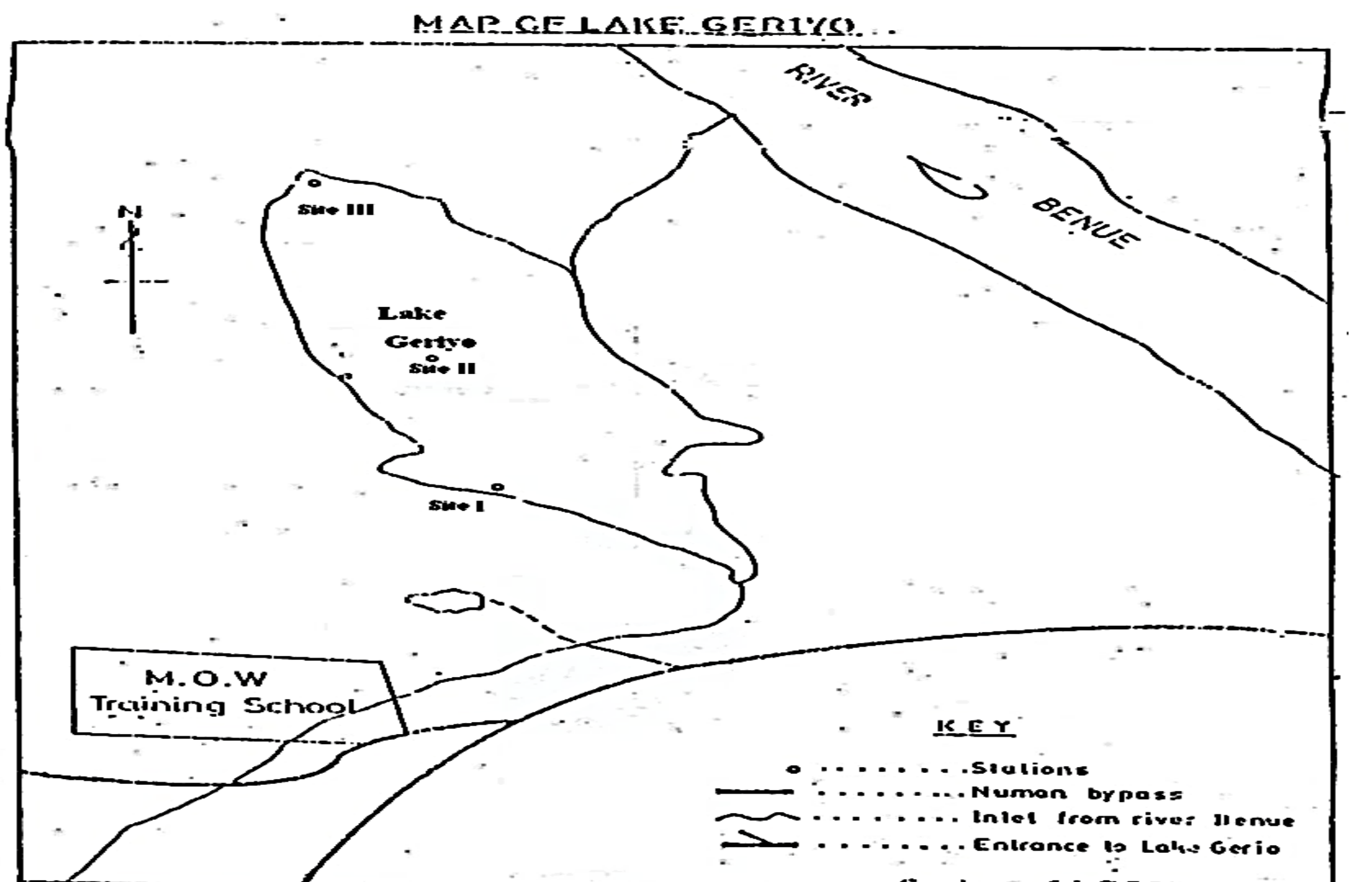

Fig. 1: Source: Ministry of Land and Survey, Yola

2.2. Fish samples collection

Samples of Tilapia (*Oreochromis niloticus*) were collected fortnightly from January, 2013 to June, 2014. The fishes were transported to the laboratory for subsequent analysis.

2.3. Blood Collection and Haematological Examination

Blood samples were collected from the caudal penduncle with the use of 5ml syringe and needle that has been treated with anti-coagulant such as heparin to prevent clotting into small sampling bottles containing Ethylene diamine tetra-acetic acid (EDTA). After the collection, the blood samples were taken to the laboratory of the Department of Zoology, Modibbo Adama University of Technology, Yola where the haematological analysis was carried out.

2.4. Packed cell volume

Blood were collected into microhaematrocrit haprinised tube which was sealed with critaseal at one end. The sampled tubes were then centrifuged for 5 minutes at 12000rpm using Hawksley microhaematocrit centrifuge. The haematocrit values were read on a microhaematocrit reader. A mean of two readings was recorded as percentage for the fish haematocrit

Determination of Red blood cells counts

Standard haemocytometer was used in the counting of the red blood cells according to method of Blaxhall and Daisley (1973) [10].

$$\text{RBC } (10^6/\text{ml}) = \frac{\text{C X D x 100 x 4000}}{\text{S x 80}}$$

Where, C = Number of cells counted
D = Diluting factors
S = Number of 1mm square counted

Determination of White blood cells counts

White blood cells were determined using the method described by Blaxhall and Daisley, 1973 [10]; Ibu and Adeniyi, 1989 [19]

$$\text{WBC } (10^3/\text{ml}) = \frac{\text{C x D x 100 x 10}}{\text{S x 4}}$$

Where C = Number of cells counted
D = diluting factor
S = Number of 1mm square counted.

Determination of haemoglobin

The cyanmet-haemoglobin method was used as described by Larsen and Snieszko (1961) [23].

Determination of mean corpuscular hemoglobin (MCH)

The mean corpuscular hemoglobin (MCH) was calculated using the formula described by dacie and lewis (1977) [13].

$$\text{MCH (pg)} = \frac{\text{Packed cell volume /dL x 10}}{\text{RBC/uL (in 10}\mathbf{6}\text{)}}$$

Determination of mean corpuscular volume (MVC)

The mean corpuscular volume (MCV) was determined as described as described by Dacie and lewis (1977) [13].

$$\text{MCV (dL)} = \frac{\text{Packed cell volume /dL x 10}}{\text{RBC/uB (in 106)}}$$

Determination of mean corpuscular haemoglobin concentration (MCHC)

The mean corpuscular haemoglobin concentration (MCHC) was calculated using the formula described by Dacie and lewis (1977) [13].

$$\text{MCHC (g/dL)} = \frac{\text{Hb/dL x 100}}{\text{PCV (\%)}}$$

3. Results

The monthly mean values of blood parameters of the examined *O. niloticus* from January, 2013 to June, 2014 shown in table1. It was shown that the packed cell volume (PCV) ranged between 10.12±0.817 % and 24.90±*3*.266% in the month of July and October respectively. The values had significant difference in variation between the months ($P<0.05$).

The monthly mean of hemoglobin (Hb) of *O. niloticus* minimum value was 4.05±0.808g/dL in January and maximum value was 10.43±0.817 g/dL in October. The values had significant difference in variation between months ($P<0.05$).

The monthly mean red blood cells (RBC) of *O. niloticus* ranged from 0.81±0.008 x10^6/mL in February and December to 1.82±0.016 x10^6/mL in October. There was significant difference in variation between months ($P<0.05$).

The monthly mean white blood cells (WBC) of *O. niloticus* minimum value was 108.67±8.206 x10^3/mL in the month of January, maximum value was 238.00±39.171 x10^3/mL in October. There was significant difference in variation between months ($P<0.05$).

The hematological indices of *O. niloticus* showed that mean corpuscular volume (MCV) ranged between 120.33±7.348 and 167.67±8.525 (fL) in the months of April and December respectively. The values showed no significant difference in variation between months ($P<0.05$).

The estimation of the mean corpuscular hemoglobin (MCH) of *O. niloticus* value ranged between 45.03±8.165 and 63.21±8.165 (pg/cell) in August and January respectively. The values exhibited significant difference in variation between months ($P<0.05$).

The estimation of the monthly mean corpuscular hemoglobin concentration (MCHC) of *O. niloticus* value ranged between 28.47±0.327 and 50.32±4.090 (g/dL) in December and January respectively. The values had significant difference in variation between months ($P<0.05$).

Table 1: Mean and standard deviation of the Haematological Indices of Tilapia *(Oreochromis niloticus)* from January, 2013 to June, 2014. Data=mean±SD, n=6

Moths	PVC%	HB(g/dl)	WBC(10^3/ml)	RBC(106/ml	MCV(fl)	MCH	MCHC
Jan	16.33±2.464	4.050±0.808	149.890±16.833	1.400s±0.082	140.32±8.045	63.21±8.165	50.32±4.090
Feb	20.63±1.633	6.030±0.817	170.770±18.257	0.810±0.008	139.31±3.268	57.35±0.245	49.22±0.163
March	23.32±0.808	7.570±0.016	167.670±24.495	1.450±0.082	122.43±4.096	51.87±7.349	44.76±3.266
April	22.73±1.632	6.700±0.816	157.020±8.327	1.380±0.081	120.33±7.348	47.39±2.872	44.02±1.633
May	19.23±1.633	7.230±1.633	153.100±5.715	1.360±0.025	140.52±4.899	50.41±4.083	45.00±0.817
June	18.19±0.816	6.740±0.817	159.440±7.348	1.180±0.082	147.38±8.168	55.40±8.165	42.21±0.817
July	10.12±0.817	4.560±0.025	163.790±16.513	1.650±0.163	149.36±7.353	62.65±1.633	44.86±4.083
Aug	22.22±1.625	6.920±0.817	136.180±5.715	1.690±0.082	133.55±24.495	45.03±8.165	35.28±0.163
Sept	16.50±0.816	4.680±0.018	131.250±4.899	1.770±0.028	153.48±16.339	52.97±4.397	33.90±2.450
Oct	24.90±3.266	10.430±0.817	238.000±39.171	1.820±0.016	142.20±1.633	50.10±3.266	40.57±8.165
Nov	14.63±0.025	8.920±0.825	178.800±6.532	1.140±0.033	166.43±8.165	50.30±4.899	30.16±1.633
Dec	13.53±0.024	5.420±0.008	156.200±2.455	0.820±0.016	167.67±8.525	47.83±1.635	28.47±0.327
Jan	10.30±0.163	7.430±0.081	108.670±8.206	1.030±0.025	137.67±8.165	58.67±8.165	49.37±3.266
Feb	14.03±0.025	7.800±0.082	121.000±13.379	1.060±0.016	147.33±8.045	56.90±4.899	50.25±0.823
March	15.95±0.033	7.380±0.163	142.500±4.089	1.000±0.005	135.00±4.082	52.15±1.633	47.30±2.015
April	16.44±0.817	7.200±0.163	153.000±8.525	1.240±0.033	131.50±4.8987	50.83±4.0833	38.99±0.817
May	20.22±1.633	6.890±0.082	150.000±4.083	1.300±0.081	143.37±2.449	49.78±4.083	35.57±4.083
June	19.20±0.816	5.030±0.816	166.000±12.659	1.100±0.082	165.60±4.082	49.98±2.062	30.40±4.899

Note P<0.05=Significant

4. Discussion

Haematological components have been developed for evaluation of fish health conditions (Aldrin *et al*., 1982). As a matter of fact, blood serves as the most convenient indicator of the general condition of the animal body. Subsequently, haematological studies are promising tools for investigating physiological changes caused by environmental pollutants (Zaghloul, 2001 [30] and Zaghloul *et al*., 2005) [31].

Packed cell volume (PCV) of *O.niloticus* values ranged from 10.12 % to 24.90 %.The observed values is higher than 16.3 ± 9.45% reported by Ahmed *et al*. (2013) [3]. The high value observed in dry seasons mighty be as result of complete decrease in water volume that lead to high concentrations of packed cell volume. Ayandiran *et al*. (2010) [7] stated that normal haematocrit values usually fall within the range of 20-35% and are rarely greater than 50% for fish. However, since transport of metals in fish occurs through the blood where the ions are usually bound to proteins and pollutants generally produce relatively rapid changes in blood characteristics of fish. Adamu and Audu (2008) [2] reported that the significant decrease in PCV may be attributed to gill damage and/or impaired osmoregulation causing anaemia and haemodilution.

Physiologically, haemoglobin is crucial to the survival of fish, being directly related to the oxygen binding capacity of blood. The monthly mean of hemoglobin (Hb) value of *O. niloticus* value ranged between 4.05 g/dL in January to 10.43 g/dL in October. The observation made is lower than values evaluated by Adakole (2012) [1]. The low Hb recorded mighty be as result influx of water from the farms, industrial, sewage that contained heavy metals such as cadmium, Nickel and lead, which alter the properties of hemoglobin by decreasing their affinity towards oxygen binding capacity rendering the erythrocytes more fragile and permeable (Adakole, 2012) [1]. Gafaar *et al*. (2010) [18] reported that prolonged reduction in haemoglobin content is deleterious to oxygen transport and degeneration of the erythrocytes could be due to pathological condition in fish exposed to toxicants. The decrease in Hb corresponds with the decrease in dissolved oxygen; an indication that the decrease in haemoglobin resulted in haemodilution. The Hb values fall lower than the range reported for catfish (Iheukwumere *et al*., 2002; Ayotunde *et al*., 2011) [20, 9]. The reduction may be due to increased rate of breakdown of red blood cells and/or reduction in the rate of formation of red blood cells (Ayotunde *et al*., 2011) [9].

The monthly mean red blood cells (RBC) of *O.niloticus* values ranged from 0.81x10^6/mL in February and December respectively to 1.82 x10^6/mL in October. The observed results are high than 135.08 ± 10.55 to 367.42 ± 94.88 x 10^3 cells/mm3 reported by Adakole (2012) [1]. Vinodhini and Narayanan (2009) [29] reported that there is a significant decrease in RBC's of fresh water fish exposed to heavy metals. According to Jimoh *et al*. (2012) [21] erythrocyte count greater than 1x10^6mm^3 is considered high and is an indicative of high oxygen carrying capacity of the blood which is characteristic of fishes capable of aerial respiration and with high activity. The reduced erythrocyte parameters are indications of macrocytic anaemia emanating from increase destruction and subsequent enhanced erythropoiesis in the liver.

The monthly mean white blood cells (WBC) of *O. niloticus* ranged between 108.67 x10^3/mL in the month of January, and 238.00 x10^3/mL in October. The recorded values are higher than 80.2 ± 79.7 x10^3/mL reported by Ahmed *et al*. (2013) [3]. The high value observed in dry season mighty be as result of low volume of water which lead to increase in WBC counts. This is as a result of high concentration of pollutant as results of wide chemical of agriculture used around the Lake. Das (1998) [14] stated that increase in WBC counts in the wild *C. gariepinus* might be a protective response to stress. The increase in WBC of fish was suggested to indicate alteration in defense mechanism against the action of the highly toxic and the bioaccumulated heavy metals in fish tissues as previously reported by Zaghloul (2001) [30] and Zaghloul *et al*. (2005) [31]. Kefas *et al* 2015 [22] also reported that the levels of heavy metals in Lake Geriyo exceeded the WHO limits indicating some levels of pollution.

The MVC, MCH, and MCHC of the *O. niloticus* were found to fluctuate in both rainy and dry seasons. This could be as result of agricultural activities taking place around the Lake in the rainy and dry seasons. Similar observations was made by Okomoda *et al*. (2010) [25] in C. gariepinus exposed to formalin. The mean corpuscular haemoglobin concentration which is the ratio of the mean haemoglobin concentration is not influenced by blood volume neither by the number of cells in the blood, but can be interpreted incorrectly only when new cells, with a different haemoglobin concentration are released (Tawari-Fufeyin *et al.,* 2008) [27].

5. Conclusion

The study indicated that there was an alteration in *O. niloticus* blood which may cause biochemical dysfunction in this species. In addition, results provided evidence that haematological parameters can be sensitive indicator of aquatic pollution.

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