HEAMATOLOGICAL AND SERUM BIOCHEMICAL PROFILE OF NILE TILAPIA, *Oreochromis niloticus* FROM ERO DAM IN IKUN EKITI, EKITI STATE NIGERIA

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ABSTRACT

Nile tilapia (*O. niloticus)* is one of the leading farmed fish species around the world because of its high commercial value in lakes and rivers where they are found. However, there are limited data on haematology and blood chemistry of this fish species found in their natural environment. Hence, the research is designed to investigate the haematological and serum biochemical profile of *O. niloticus* collected from Ero dam and check if these parameters will vary significantly among sex. Fish collected from mongers in the early hours of the day had their blood collected and analyzed using standard clinical methods. The test for significant difference (*P*= 0.05) among the sex was done using student t- test. The result showed that higher values were obtained in female than male in packed cell volume (PCV), red blood cell (RBC), white blood cell (WBC), mean corpuscular haemoglobin (MCH) and eosinophil from the haematological analysis done. On the other hand, neutrophil, lymphocytes, monocytes, platelets, mean corpuscular volume (MCV) and mean corpuscular haemoglobin concentration (MCHC) were higher in male than female. Despite the difference in values obtained, there was no marked significant difference between sexes (male and female) for all the haematological parameters. Also, the result of the blood chemistry showed that higher means were recorded in male than female in cholesterol, glucose, total protein, albumin, uric acid, creatinine, urea and sodium; though there was no marked significant difference shown between the different sexes except in glucose and albumin. Likewise, higher mean was recorded in female than in male in triglycerides, calcium, Aspartate Amino Transferase (AST), Alanine Amino Transferase (ALT), Alkaline Phosphate (ALP) and potassium. Using the same analytical method, no eosinophils and monocytes was found in some of the female Nile tilapia used in this study.

Keywords: Haematology, serum, Biochemistry, Tilapia, *Oreochromis niloticus*, Ero dam, Nigeria.

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INTRODUCTION

Nile tilapia (*O. niloticus)* is among the leading farmed fish species and one of the fish species of great economic importance around the world. It is usually a native fish species of high commercial value in lakes and rivers where they are found. The good health status of fish is the main element for their welfare, thus it is of great significant (Atanasova *et al*., 2008). Blood parameters analyses have proven to be valuable tools to analyze the health status of farmed and uncultured fish as these indices provide reliable information on possible exposure to mutagens, metabolic disorders, deficiencies and chronic stress status before clinical symptoms appears (Bahmani *et al*., 2001). Esonu *et al*., (2001) opined that haematological profile reflects the physiological responsiveness of the animals to its internal and external environment. Therefore, any change in the external environment can cause a dysfunction of blood and as such have severe effects on the physiological activities such as resistance to disease, metabolism, breeding performance and health condition of the entire body. These are reflected as alterations in blood constituents which can be used as diagnostic indicators. Thus, the challenge of lack of proper management practices of fisheries can be cushioned by the availability and accessibility of the haematological and biochemical profile of such species as these will form the data base for most diagnostic investigations.

Many works has been done on the haematological and serum biochemical profile of some fish species around the globe. Surprisingly, there are few reports of normal blood and serum values for tilapias (Terao and Ogaw, 1984; Palti *et al.,* 1999; Bittencourt *et al*., 2003; Chen *et al.,* 2003 and Mauel *et al*., 2007). Among these are works done on the haematological and biochemical values of *O. niloticus* cultured in a semi- intensive system (Nilza *et al*., 2003; Bittencourt *et al*., 2003). Cnaani *et al*., (2004) did a comparative study on the biochemical parameters of *O. aureus*,

O. mossambicus and two strains of *O. niloticus* in response to stress. The physiological and haematological study of the response of *O. niloticus* exposed to single and consecutive stress of captured was done by Martins *et al*., (2004). The effect of varying levels of fish oil on growth performance, body composition and haematological characteristics of *O. niloticus* was also studied. Kpundeh *et al*., (2013) worked on densities and chronic zero culture water exchange stress on biological performances, haematological and serum biochemical indices of *O. niloticus* juvenile. Also, Catalina *et al*., (2013) worked on the haematological and biochemical characterization of *O. niloticus* reared intensively in a recirculating Aquaculture system in relation to water temperature. A lot of studies have been carried out on the physiology, life activities of *O. niloticus* from different water bodies but there is paucity of information on the haematological and serum biochemical profile of *O. niloticus* from Ero Dam in Ekiti State Nigeria. Hence, this research is designed to investigate the haematological and serum biochemical profile of this species from Ero dam. The objective is to check if there will be variation in the haematological and serum biochemical profile among different sex of the species.

METHODOLOGY

Study Area

Ero dam in Ikun- Ekiti, Nigeria is located in Moba Local Government area of Ekiti State. The dam is located on intersect of latitude $7^{0}35^{1}$N of the equator and on the longitude $5^{0}31^{1}$E of the Greenwich meridian. The reservoir is an earthly embankment with a length of 662m with an impounded reservoir area of 4.5km^{2} and normal water level of 504.5m. The dam supplies drinkable water to three Local Government areas which are Oye, Ido-Osi and Moba itself.

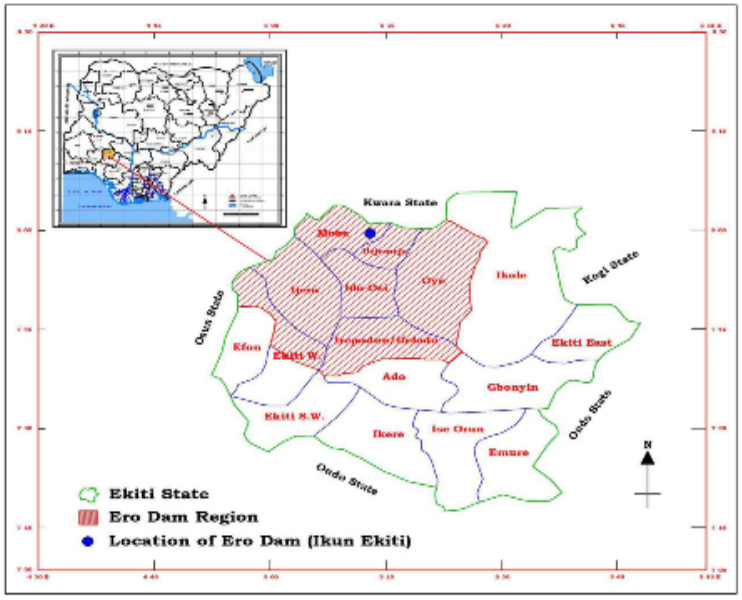

Fig 1: The Study Region

Source: Ministry of Lands and Housing Ado, Ekiti State, Nigeria, 2013.

Sample Collection

Fish samples used for the study were obtained from Ero dam. The fish were collected early in the morning from the fish mongers at the bank of the dam and were transported immediately in ice cooler to a medical laboratory (City Medical Laboratory Diagnostic Center Nigeria limited) opposite Ekiti State University Teaching Hospital (EKSUTH), Ado- Ekiti for blood collection and analysis.

Sample Preparation

Fish were weighed prior to blood collection. Blood were taken from each of the fish from behind the anal fin using a 5ml plastic syringe and 22gauge needle. Blood samples of about 2.5ml collected were placed into dry plastic tubes containing anti-coagulants EDTA and lithium heparin to prevent clotting.

Determination of Haematological Parameters

In the laboratory, standard haematological test was performed in order to identify the various types of cell found in the blood.

Biochemical Analyses

Separated serum was analyzed for aspartate amino transferase (AST), alanine amino transferase (ALT) and other parameters using modified method of different researchers.

Statistical Analysis

The haematological and serum biochemical values obtained was statistically analyzed using Microsoft Excel 2010 parametric student T- test.

RESULTS

Table 1 shows the weight and haematological composition of male and female *O. niloticus.* The weight of male and female *O. niloticus* used for the study ranged from 78- 120g. The haematological values obtained ranged from 50- 71μl^{-1}(Neutrophils), 23-46μl^{-1} (Lymphocytes), 1- 2μl^{-1} (Monocytes), 2.9x10^{6}- 8.3x$10^{6}\mu l^{-1}$ (Red Blood Cells), 4.8- 12.6μl^{-1} (White Blood Cells) and 1- 5 μl^{-1} (Eosinophil). No values was recorded for Eosinophil in female 1 and 2 as shown in the table 1 and also, no value was recorded for Monocytes in female 1. The following range of values 49- 65%, 190-600, 7.1- 16.9gd^{-1}, 16.7- 21.6gd^{-1}, 33.1- 33.4gd^{-1} and 3.0- 5.6$Pgcell^{-1}$ was obtained for Packed Cell Volume, Platelet, Mean Corpuscular Volume, Haemoglobin, Mean Corpuscular Haemoglobin Concentration and Mean Corpuscular Haemoglobin respectively.

The weight and biochemical composition of male and female *O. niloticus* are shown in table 2. The range of values obtained are: 8.6- 12.8mg/dl (cholesterol), 3.2-6.9mg/dl (triglycerides), 0.4-1.0mg/dl (glucose), 7- 15mg/dl (total protein), 3- 8mg/dl (albumin), 1.5- 3.0mg/dl (uric acid), 0.3- 0.9mg/dl (urea), 0- 23mg/dl (creatinine), 1.0- 4.0mg/dl (calcium), 2.4- 3.6mol/l (potassium) and 121- 125mol/l (sodium). Also, the range of values recorded for Aspartate Amino Transferase, Alanine Amino Transferase and Alkaline Phosphate are 12-23, 6-13 and 11-34 respectively.

Table 1: The weight and haematological composition of Male and Female *O. niloticus* from Ero dam

	Weight (g)	NEUT(μl^{-1})	LYMP(μl^{-1})	MONO(μl^{-1})	PCV(%)	RBC(μl^{-1})	PLATELET	MCV(gd^{-1})	MCH(Pgcell^{-1})	WBC(μl^{-1})	HGB(gd^{-1})	MCHC(gd^{-1})	EOSIN(μl^{-1})
M1	120	61	46	01	60	3.9	600	15.4	5.1	5.6	20.0	33.3	2.0
M2	102	55	43	01	49	2.9	250	16.9	5.6	12.6	16.3	33.2	1.0
M3	111	61	36	02	60	3.9	300	15.4	5.1	4.8	20	33.3	1.0
M4	108	70	23	02	56	6.8	301	8.2	2.8	4.9	18.7	33.4	5.0
F1	100	60	40	-	65	7.2	340	9.0	3.0	11.0	21.6	33.2	-
F2	78	71	28	01	58	4.8	310	12.1	4.0	7.4	19.2	33.1	-
F3	105	65	33	01	61	3.9	280	15.6	5.2	12.5	20.3	33.3	2.0
F4	98	50	45	02	59	8.3	190	7.1	2.4	6.7	19.7	33.3	3.0

Key: F= Female, M= Male, NEUT= Neutrophils, LYMP= Lymphoctes, MONO= Monocytes, PCV= Packed Cell Volume, RBC= Red Blood Cell, MCV= Mean Corpuscular Volume, MCH= Mean Corpuscular Haemoglobin, WBC= White Blood Cell, HGB= Haemoglobin, MCHC= Mean Corpuscular Haemoglobin Concentration, EOSIN= Eosinophils.

Table 2: The weight and biochemical composition of male and female *O. niloticus* from Ero dam

	Weight (g)	Cholesterol (mg/dl)	Triglyceride (mg/dl)	Glucose (mg/dl)	Total protein(mg/dl)	Albumin (mg/dl)	Uric acid (mg/dl)	Calcium (mg/dl)	Aspartate Amino Transferase	Alanine Amino Transferase	Alkaline Phosphate	Creatinine (mg/dl)	Urea (mg/dl)	Potassium (mpl/l)	Sodium (mol/l)
M1	120	8.6	6.9	0.9	11	6	1.7	2.3	16	07	11	14	0.7	3.0	126
M2	102	14.9	5.0	0.8	15	8	2.0	1.0	22	13	23	23	0.6	4.0	137
M3	111	11.2	4.1	1.0	12	7	3.0	4.0	12	07	12	12	0.9	2.4	121
M4	108	8.9	3.2	0.9	09	4	1.5	2.2	14	05	12	09	0.4	2.9	145
F1	100	8.8	4.6	0.4	07	3	2.1	2.5	22	09	34	07	0.5	2.9	123
F2	78	8.9	4.6	0.5	10	4	1.9	1.6	23	11	12	12	0.6	2.9	132
F3	105	10.6	5.8	0.7	11	3	1.1	3.8	11	06	33	08	0.3	3.1	121
F4	98	12.8	5.7	0.5	15	3	1.6	1.9	14	12	23	0	0.4	3.6	143

Table 3 and 4 shows the mean and standard deviation of the haematological and biochemical composition of male and female *O. niloticus.* Also, both male and female fish were compared and the test showed no significant differences (P= 0.05) in all the parameters except in glucose and albumin.

Table 3: The mean and standard deviation of haematological composition of male and female *O. niloticus* from Ero dam

Parameters	Male	Female
Weight (g)	110.25± 7.50[a]	92.25± 11.87[a]
Neutrophils (μl^{-1})	61.75± 6.18[a]	61.50± 8.89[a]
Lymphocytes (μl^{-1})	37.00± 10.23[a]	36.50± 7.51[a]
Monocytes (μl^{-1})	1.50± 0.58[a]	1.33± 0.58[a]
Packed cell volume (%)	56.25± 5.19[a]	60.75±3.10[a]
Red blood cells (10^6 μl^{-1})	4.38± 1.68[a]	6.05± 2.05[a]
Platelet	362.75± 159.95[a]	280.00± 64.81[a]
Mean corpuscular volume (gd^{-1})	13.98± 3.91[a]	10.95± 3.72[a]
Mean corpuscular haemoglobin (pg $cell^{-1}$)	4.65± 1.26[a]	3.65± 1.23[a]
White blood cell (μl^{-1})	6.98± 3.77[a]	9.40± 2.80[a]
Haemoglobin (gdl^{-1})	18.75± 1.74[a]	20.20± 1.04[a]
Mean corpuscular heamoglobin conc (gd^{-1})	33.30± 0.08[a]	33.23± 0.10[a]
Eosinophils (μl^{-1})	2.25± 1.89[a]	2.50± 0.71[a]

Means with the same alphabet across the column are not significantly different at P<0.05.

Table 4: The mean and standard deviation of biochemical composition of male and female *O. niloticus*

Parameters	Male	Female
Weight (g)	110.25± 7.50[a]	95.25± 11.87[a]
Cholesterol (mg/dl)	10.9± 2.91[a]	10.27± 1.88[a]
Triglyceride (mg/dl)	4.80± 1.58[a]	5.18±0.67[a]
Glucose (mg/dl)	0.90± 0.08[b]	0.53± 0.13[a]
Total protein (mg/dl)	11.75± 2.50[a]	10.75± 3.30[a]
Albumin (mg/dl)	6.25± 1.77[b]	3.25± 0.50[a]
Uric acid (mg/dl)	2.05± 0.67[a]	1.68± 0.43[a]
Calcium (mg/dl)	2.38± 1.23[a]	2.45± 0.97[a]
Aspartate Amino Transferase (AST)	16.00± 4.32[a]	17.50± 5.92[a]
Alanine Amino Transferase (ALT)	8.00± 3.46[a]	9.50± 2.65[a]
Alkaline Phosphate (ALKP)	14.50± 5.69[a]	25.50± 10.28[a]
Creatinine (mg/dl)	14.50± 6.03[a]	6.75± 4.99[a]
Urea (mg/dl)	0.65± 0.21[a]	0.45±0.13[a]
Potassium (mol/l)	3.08± 0.67[a]	3.13± 0.33[a]
Sodium (mol/l)	132.25± 10.81[a]	129.75± 10.05[a]

Means with the same alphabet across the column are not significantly different at P=0.05, while means with different alphabet are significantly different.

DISCUSSION

This study showed that higher values were obtained in female than male in packed cell volume, red blood cell, white blood cell, mean corpuscular haemoglobin and eosinophil. On the other hand, in neutrophil, lymphocytes, monocytes, platelets, mean corpuscular volume and mean corpuscular haemoglobin concentration were higher in male than female according to the result of haematological analysis done. Despite the differences in values obtained, there was no marked significant difference between sexes (male and female) for all the haematological parameters. Red blood cells (RBCs) count was lower in male than the female but showed no significant

difference at P=0.05. This result contradict the report made by Karimi *et al.,* (2013) in their work on the effect of sexuality on some haematological parameters of the yellow fin sea bream *Acanthopaguslatus* in Persian Gulf, that Red Blood Cells (RBCs) count was higher in male than in female. Elevation in white blood cell reported in this study may be attributed to stress (of reproduction, sourcing for food etc), infectious disease and tissue damage. Baker *et al.*, (2000); Buckham-Sporer *et al*., (2008) and Adenkola *et al*., (2009) attributed elevated leucocytes count in animals to infectious diseases, tissue damage and stress and suggested that it could be due to the mobilization of leucocyte from their pool to the peripheral circulation. No eosinophils or its precursor was found in blood smears of Nile tilapia according to Bittencourt *et al.,* (2003) but our study showed that eosinophils was present in both sexes of Nile tilapia collected from Ero-dam, though it was absent in some of the female fish. This support the report of Pitombeira and Martins (1970), Ezzat *et al.* (1974) that though, eosinophils are generally lacking in fish blood but are present in some fish species. The result of this study support the report of Yu *et al.,* (2013) that there were no significant differences in haematological values between male and female in their work on haematological and plasma biochemical reference values of the yellow pond turtle *Mauremys mutica* and the effects of sex and season. Also, it supports the report of Aladi *et al.,* (2008) that age and sex has no association with haematological values. However, this does not agree with Egbunike and Akusu (1983), who reported that sex and age significantly affect blood values in pigs.

In blood chemistry, higher means were recorded in male than female in cholesterol, glucose, total protein, albumin, uric acid, creatinine, urea and sodium; though there was no marked significant difference showed between the different sexes except in glucose and albumin. Likewise, higher mean was recorded in female than in male in triglycerides, calcium, AST, ALT, ALP and potassium. A higher means was recorded by Palti *et al.*, (1999) for cholesterol (267g/dl) and ALP (35U/l) while a lower means was recorded for total protein (4.5g/dl) and albumin (2.2g/dl) in *O. aureus* than those observed in current study. El Hawarry (2012) reported that variations obtained in blood chemistry could reflect that some parameters could have been affected significantly by culture conditions. According to Bittencourt *et al*., (2003) some variations influenced by environmental conditions, gender, age, origin, breeding system, feeding and lineage, may interfere with the results obtained in their tests. Maita *et al.,* (1998) reported that total cholesterol differed significantly among genotype which was found to be associated with disease resistance in fish. The values obtained in our study showed no significant among sexes.

Blood glucose is an important source of energy for many cells. Blood glucose is normally maintained by the breakdown of dietary carbohydrates and a rather complex system of endogenous production. Chavin and Young (1970) pointed out that temperature affects the blood sugar levels. The values obtained for blood sugar in this study was quite low (0.90± 0.08, 0.53± 0.13mg/dl for male and female) when compared to the values (60.32mg/dl reported for Nile tilapia; 39.7mg/dl for channel catfish *Ictalurus melas* and 76.4mg/dl for "pacu") as reported by Bittencourt *et al.,* (2003) and Tavares-Dias *et al*. (1999) respectively. According to Bittencourt *et al.,* (2003), some fishes become hyperglycemic at low temperature while some become hypoglycemic under same condition. It was further said that other environmental conditions

(such as seasonal temperature changes) and physiological factors may contribute to the variations observed. Seasonal temperature changes may affect blood sugar levels.

Albumin is the major component which is characterized by a high negative charge and relatively low molecular mass that function in the regulation of colloidal osmotic pressure of the blood and transport of some exogenous chemicals and endogenous metabolites (De Smet *et al*., 1998; Baker, 2002). Metcalf and Gemmell (2005) reported that some fish species lack albumin and concluded that it has a phylogenetic significance. Our study showed that both male and female *O. niloticus* from Ero- dam possess albumin, only that it was higher in male than in female. Also, the higher means obtained in male *O. niloticus* from some of the biochemical parameters may be attributed to the bigger size of the male fish used. The total protein (11.75, 10.75mg/dl) and albumin (6.25, 3.25mg/dl) recorded from this study for the male and female with the average weight of 110. 25± 7.50g and 92.25± 11.87g respectively were higher than those reported by Yavuzcan Yildiz *et al*. (1997) for total protein (4.60g/dl) and albumin (2.96 g/dl) in small (52g) *O. niloticus.* Also, Hussein *et a*l. (1996) reported a lower total protein (3.40g/dl) and albumin (0.67 g/dl) in their study on *O. niloticus* with average weight of 38.46 g. Chen *et al*. (2003), identified the blood chemistry in 120 healthy *O. niloticus* (393.2±117g) through a year and showed higher means for Cholesterol (251.9g/dl) and lower albumin (1.32 g/dl). A lower value than what was recorded in our study was reported by Chen *et al.* (2003) for albumin (1.32 g/dl). According to Wiegertjes *et al*. (1996), high levels of serum protein and albumin are thought to be associated with strong innate response in fish.

No significant difference was observed between sexes in this study for biochemical parameters such as Aspartate aminotransferase (AST)**,** useful in evaluating muscle and liver damage; Alanine aminotransferase (ALT), a cytoplasmic enzyme and it is considered to be liver specific (the elevation of both enzymes can occur with states of altered hepatocellular membrane permeability); Alkaline phosphatases (ALP) are a group of enzymes which catalyze the hydrolysis of a phosphate group from an organic molecule at an alkaline pH. Elevation in ALP concentration could be as a result of cholestasis and normal bone growth in adult and immature animals respectively. Also, creatinine and uric acid levels are indicator of kidney function. In the present study, creatinine and uric acid showed no significant difference between male and female of *O. niloticus* from Ero- dam.

CONCLUSION

From our study, there is possibility that fish species collected from the same site will show no significant difference in their haematological and biochemical values regardless of their age, size and sex. Khalid *et al.,* (2007) worked on *O. niloticus* along different sites of lake reported that highly significant difference was shown in the red blood cells count of fish collected from different sites. This research gives room for further study on the comparative studies of the haematological and biochemical analysis of *O. niloticus* collected from different dam in Ekiti

State. Also, further work should be done to investigate while some female *O. niloticus* collected from Ero dam lacked eosinophils and monocytes.

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