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Seasonal Changes in Some Hematological Parameters in Association with Acted in Oxygen Transport in Nile Tilapia (*Oreochromis niloticus L*)

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Abstract

In this study, seasonal changes in some blood parameters related to oxygen transport in adult tilapia (*Oreochromis niloticus* L.) were detected. Blood samples were collected from caudal vein of each fish. Red blood cell (RBC), mean cell volume (MCV), mean cell hemoglobin (MCH), mean cell hemoglobin concentration(MCHC), hematocrit (Hct), methemoglobin (Hi), serum iron concentration (SI), total iron binding capacity (TIBC), unsaturated iron binding capacity (UIBC), serum transferrin (TRF), transferrin saturation (TRF %) levels were determined. RBC, length and weight of fish were found to be linear relation. It was found that there were linear relations between SI, RBC, Hct, Hb, TIBC, UIBC, TRF and %TRF. Negative relation was found between oxygen level in the water RBC, Hb, MCH, SI, TIBC, UIBC, TRF and %TRF parameters. There is a linear relation among MCH, MCHC and Hi levels, which is statistically insignificant (P>0.05). Negative correlation was found between Hi and other parameters, which are detected. But Hi was found to be differ significantly when compared with SI, TIBC, UIBC, TRF and %TRF by statistical analysis (P>0.05). The data obtained indicated that all parameters were found to be effected significantly by the seasonal changes (P<0.05). In addition, these parameters were determined to be lower in autumn and winter than in spring and summer. However, blood parameters of all the fish tested were detected within the normal values.

Keywords: Tilapia, *Oreochromis niloticus* L., Hematological parameters, Oxygen transport

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Introduction

All living organisms need oxygen for metabolic activity and energy consumption. Although continental livings supply oxygen from the air easily, the rate of oxygen uptake of fish is limited by diffusion through the gills dissolved oxygen in the water.

In addition, the oxygen concentration in the water can be less 30 times than the air. This shows that the uptake and transport of oxygen is very important for the oxygen requirements of fish [1]. The body temperature of aquatic poikilothermic as fish is maintained close to that of water, their metabolic activities as well as enzymatic reactions depend on exchange of heat between the body temperature and the surrounding water [2]. The differences among hematological values are affected by factors as photoperiod, temperature, reproduction, and feed or by impacts of changes in the combination of these factors, in addition by age, sex, season, capture, collection methods, water quality, starvation [3, 4, 5].

It has been reported to have an effect on blood parameters of stress caused by intensive stocking and environmental factors under culture conditions [6, 7]. Hematology that provided the information about habitat of livings is also important to determine the rate of feed intake and the condition of fish stocks in ichthyologic researches. Economic losses can be eliminated when hematological methods are used as aids (early diagnosis etc.) in fish disease diagnostics [8]. Therefore, hematology contains important information to determine the conditions of taxonomical, ecological, physiological and pathological of fish.

Nile Tilapia (*Oreochromis niloticus* Linnaeus, 1758) which belongs to the class Pisces is from the family Cichlidae and is native to Africa. Tilapia is important aquaculture species due to its ability to tolerate a wide range of environmental conditions, fast growth, successful reproductive strategies, and ability to feed at different trophic levels [9].

Hematological parameters have been recognized as valuable tools for the monitoring of fish health. Before using hematology as a standard for assessing the health of fish, primarily differences for each species under the influence of many factors should be identified and the normal ranges of hematological separately determined. OMOREGIE and OYEBANJI [10] reported that the information on the blood parameters of *O. niloticus* is limited although tilapia is a good experimental fish for studying the effect of environmental conditions on blood, especially only a few studies have investigated on the relationship with the seasons of hematologic parameters.

The aim of this study was to investigate some blood parameters (hematocrit, red blood cell count, hemoglobin, total iron binding capacity, serum transferrin, transferrin saturation % etc.) acted in oxygen transport in fish blood and to determine seasonal variations of these parameters.

Material and Method

Experimental Design

Totally 300 of *O. niloticus* comprising 25 sampling (each month randomly) were caught from in Cukurova University, Faculty of Fisheries, Dr. Nazmi Tekelioğlu Freshwater Fish Production Centre, Adana, Turkey. The size of the tank was 1 x 5 x 0.75 m and the health status of fish stocked in the system providing flow through fresh water was observed macroscopically examined for ectoparasites. Fish were fed once in a day with pelleted food. Water temperature and oxygen content in tanks were measured monthly (YSI pro 20). Then, blood samples were immediately taken by cutting the caudal peduncle without anesthesia from 25 fish every month (BRAUNER et al., 2000a; b). To determine the amount of hemoglobin in a blood sample, we collected in tubes containing disodium salt of ethylene diamine tetra-acetic acid (EDTA) and analyzed according to Cyanomethemoglobin method [11, 12]. Erythrocyte was counted on Thomas Chamber by using Natt-Herrick solution. Microhematocrit method was used for hematocrit. The values of mean corpuscular volume (MCV), mean corpuscular hemoglobin (MCH) and mean corpuscular hemoglobin concentration (MCHC) were calculated according to MIALE [13]. Standard techniques and procedures described by VIRANI and REES [14] and SADLER et al. [15] were employed in the assessment of serum iron (SI), serum iron binding capacity (TIBC), serum iron binding amount (UIBC), serum transferrin (TRF) and percent saturation of transferrin (TRF%).

Statistical analysis

The hematological parameters were evaluated in both monthly and seasonally. The data obtained were analyzed with DUNCAN multiple comparison test and was the significance level ($p < 0.05$).

Results

Temperature and oxygen levels obtained from the pools monthly and mean length and weight were given in Table 1.

Table 1. Temperature, dissolved oxygen values and mean length and weight values of fish

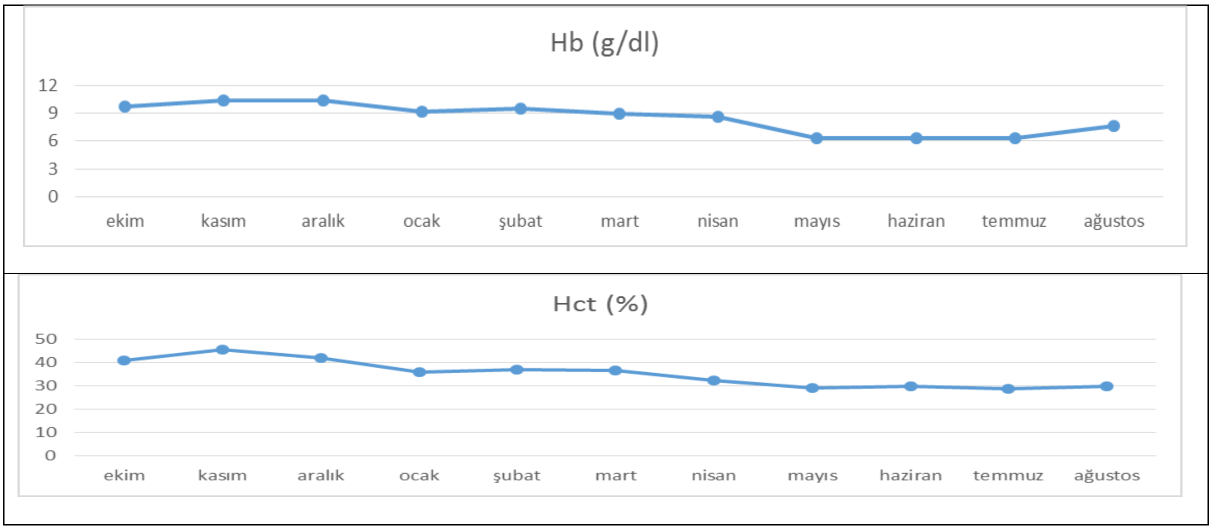
Months	**Temperature(^{o}C)**	**Dissolved Oxygen (mg/lt)**	**Lenght (cm)**	**Weight (g)**
October	21.5	9.28	18.63	121.95
November	17.5	6.40	20.75	172.27
December	19.0	5.50	19.30	130.63
January	19.7	9.84	19.38	122.63
February	16.0	7.20	18.92	109.22
March	24.5	8.10	18.00	100.10
April	27.5	6.00	17.98	93.94
May	28.5	8.84	18.95	106.51
June	27.0	10.57	18.05	91.34
July	28.0	9.50	18.70	103.27
August	29.5	8.30	18.27	99.26
Mean (SD)	**23.5**	**8.14**	**18.80±0.18**	**113.65±4.04**
Min-Max Values	**16.0-29.5**	**5.5-10.57**	**17.93±0.37-20.75±1.03**	**91.34±7.44-172.27±11.81**

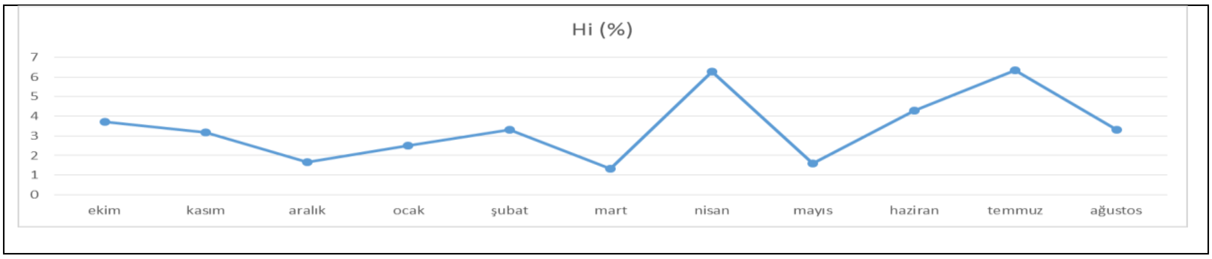
While a gradually reduction was recorded ($p< 0.05$) in the erythrocyte amount (RBC) of *O. niloticus*, the opposite situation observed in MCV. Declines in MCH and MCHC values were determined in summer (Figure 1).

Figure 1. Seasonal variation of RBC amount (x10^6), mean corpuscular volume, MCV (µ3), mean corpuscular hemoglobin, MCH (pg) and mean corpuscular hemoglobin concentration, MCHC (%) in *O. niloticus*

Methemoglobin (Hi) known as brown hemoglobin pigment is a derivative of oxidized hemoglobin and it [16]. Methemoglobin that is responsible for fish kills in extreme cases cannot perform the function of carrying oxygen [16, 17]. The data of hemoglobin and Hematocrit and Methemoglobin were given in Figure 2.

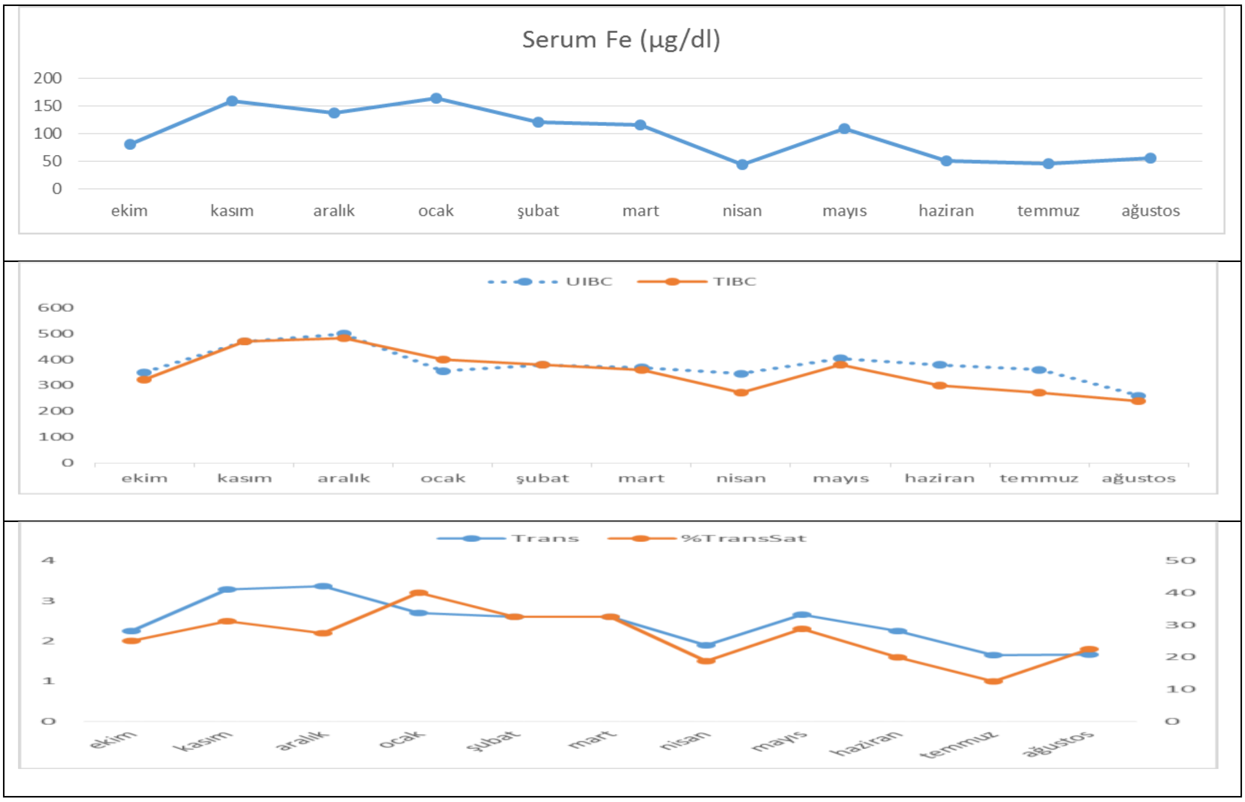
Figure 2. Seasonal variation of Hb (g/dL), Hct (%) and Hi (%)





Total serum iron binding capacity (TIBC), serum unsaturated iron binding capacity (UIBC) and serum iron amount (SU) play a key role in oxygen transport in the blood to determine abnormalities of iron metabolism was evaluated as seasonal (Figure 3).

Figure 3. Seasonal variation of total serum iron binding capacity (TIBC), serum unsaturated iron binding capacity (UIBC) and serum iron amount (SU)



Discussion

Many factors such as sex, age, and size, environmental and physiological conditions are known to effect hematological responses in fish. Erythrocytes are one of the most important in the determining of the erythrocyte characteristics significantly the efficiency of oxygen transport from the respiratory system to tissues [18, 19], especially changes in their number and volume could influence metabolic performance [20,21]. The mean erythrocyte value was reported as 2.731x10^6/mm^3 on blood parameters of Nile tilapia [22]. BADAWI and SAID [23] reported that erythrocyte was 1.05-1.28 x 10^6/mm^3. QIANG et al. [5] demonstrated the effect on hematological parameters of the water temperature and diet in juvenile individuals of *Streptococcus iniae*-infected *O. niloticus* and reported the value of erythrocyte in between 1.790 and 2.651x10^6/mm^3. In our study, we monitored the amount of erythrocyte monthly and the data showed that while erythrocyte amount had the highest values (2.45x10^6/mm^3) in November, the lowest value (1.43x10^6/mm^3) in May. Our data was similar to that reported by QIANG et al [5].

SAHAN and DUMAN [24] found increasing for erythrocytes after application of the glucan on *O. niloticus*. AZIZOGLU and CENGIZLER [22] reported higher the amount of erythrocytes in the blood in May and August in comparison with November and February. This shows that the spawning period of *O. niloticus* (in May) have revealed an abrupt change on the amount of erythrocytes. Mean corpuscular hemoglobin concentration (MCV) of erythrocyte hemoglobin is the most faithful erythrocyte constant. CENGIZLER and SAHAN [25] examined the total WBC production as seasonal in two carp species living in the Seyhan River and they reported an inverse relationship between the amount of erythrocytes and MCV. In our study is in agreement with the findings of CENGIZLER and SAHAN [25] when was a decline in erythrocyte number in April-July, MCV values increased as well as the opposite situation was observed in August. The similar findings reported by KOCABATMAZ and EKINGEN [26] and SCHÜTT et al. [27]. Results from this study showed that despite the decrease of MCH values were found to be statistically significant in summer. SAHAN and CENGIZLER [28] reported that MCH values (0.004pg) of *Barbus rajanorum* and *Capoeta barroisi* increased in winter. This may be the reduction in the number of erythrocytes.

KOCABATMAZ and EKINGEN [26] and SCHÜTT et al. [27] showed that there was negative relationship between the number of erythrocytes and MCH. In other study report by BENFEY and SUTTERLIN [29], the determination of mean corpuscular hemoglobin concentration (MCHC) is more useful than the measurement of RBC and HCT in the evaluation of the oxygen transport capacity. The highest MCHC was recorded on rainbow trout in April, had the lowest in July [30]. ADEYEMO et al. [31] examined the relationship between MCHC values and temperature rise on *Clarias gariepinus* found negative relation of them. In the previous studies, MHCH and Hb values have been found to be parallel to each other [27, 28]. In this study, significant relationship between the amount of Hb and MCHC ($p < 0.005$) were found.

Total iron-binding capacity and serum iron are widely used to determine abnormalities in iron metabolism. Our results of statistical analysis of serum iron and its related parameters indicated that there was significant linear relationship these parameters (serum iron, total iron binding capacity, transferrin saturation percent, and unsaturated iron-binding capacity). There were statistically significant an inverse relationship in between serum iron and methemoglobin ($p<0.05$). Previous studies showed that basal levels of SI, UIBC and TS were reported in salmonids, sea bass and tilapia [32, 33, 34, 35] and our findings confirm these conclusions.

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