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SHORT PAPER

Effects on Mortality of Biochemical and Limnological Properties on Some Fish Species in Sultansuyu Dam Lake (Malatya), Turkey

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Abstract

In this study, hematological and biochemical investigations were carried out on *Cyprinus carpio*, *Leuciscus cephalus*, *Capoeta trutta* ve *Capoeta capoeta umbla* fish species living in Sultansuyu Dam Lake (Malatya). The physical, chemical and microbiological features of the reservoir were specified and heavy metal analyses were also conducted. As a result of hematological investigation, a statistical increase of $P<0.05$ in total leukocyte count, and a decrease of $P<0.05$ in erytrocyte, hemoglobin and hematocrit values were observed. With respect to microbiological analysis, the presence of *Enterococcus* shows the drainage of domestic wastes into the reservoir. With water analyses, it was found out that the water quality was second rate and the sulphur content in the water was above standard levels. The existence of large amount of sulphur was presumed to be the result of apricot processing. Significant heavy metal pollution was not observed.

Keywords: Hematological and Biochemical parameters, Heavy metal**,** Sultansuyu Dam Lake, *Cyprinus carpio*, *Leuciscus cephalus*, *Capoeta trutta*, *Capoeta capoeta umbla.*

Sultansuyu (Malatya) Baraj Gölünde Yaşayan Bazı Ekonomik Balık Türleri Üzerinde Biyokimyasal ve Limnolojik İncelemeler

Özet

Bu çalışmada Sultansuyu (Malatya) Baraj Gölü'nde yaşayan *Cyprinus carpio*, *Leuciscus cephalus*, *Capoeta trutta* ve *Capoeta capoeta umbla* balık türlerinde hematolojik ve biyokimyasal inceleme yapılmıştır. Ayrıca gölün fiziksel, kimyasal, mikrobiyolojik ve ağır metal analizleri yürütülmüştür. Hematolojik incelemede total lökosit sayılarında istatistiksel olarak bir artma $P<0,05$, eritrosit hemoglobin ve hematokrit değerlerinde bir azalma $P<0,05$ olduğu saptanmıştır. Mikrobiyal analizde *Enterococcus* varlığı evsel atıkların drenaj edildiği göstermektedir. Su analizlerinden suyun ikince kalite su olduğu ve suyun kükürt miktarının standartların üstünde bulunduğu belirlenmiş olup, kaynağının yörede yapılan kayısıcılıktan kaynaklandığı düşünülmüştür. Ağır metal yönünden önemli bir kirliliğe rastlanılmamıştır.

Anahtar Kelimeler: Hematolojik, Biyokimyasal parameter, Ağır metal Sultansuyu Baraj Gölü, *Cyprinus carpio, Leuciscus cephalus, Capoeta trutta, Capoeta capoeta umbla.*

Introduction

Every year, the increase of human population and the accompanying with the growth of industrialization results in the increase of pollution in aquatic ecosystems (Caussy *et al*., 2003). Water pollution is caused by industrial waste materials (petroleum, dyes, detergents, heavy metals, sewage), agricultural insecticides and surplus fertilizers, natural and domestic wastes, and lead, mercury and sewage that are released into water. As it influences aquatic life forms directly, the pollution also reaches humans through the food chain.

Hematological and biochemical parameters are being used as indicators in the measurement of health conditions and toxicological symptoms of organisms (Thrall, 2004; Pimpao, 2007; Venkateswara Rao, 2006). While providing information about the health status of organisms, these parameters may also indicate abnormal environmental conditions (Elahee and Bhagwant, 2007).

Information about the existence, status and degree of possible sickness in organisms can be rapidly obtained by hematological and biochemical

clinical tests (Blaxhall and Daisley, 1973). Moreover, the degree of adverse effects on organisms can be found out by various clinical tests, such as enzyme, hormone, histopathology, microbiology etc. tests (Cazenave *et al*., 2005; Van der Oost *et al*., 2003).

In order to interpret the hematological and biochemical analyses on fish which rapidly respond to environmental conditions; blood sampling and laboratory techniques (Hardig and Höglung, 1984), seasonal changes (Andersen *et al*., 1985; Cech *et al*., 1982; Hardig and Höglung, 1984), size and ontogenesis, genetic features (Murray, 1984), sex and population density (Atkinson and Juddy, 1978; Murray, 1984), geographical distribution (Blaxhal and Daisley, 1973; Bridges, 1984), habitat, starvation, stress, pH, water and transportation (Wilhem Filho *et al*., 1992) should also be taken into consideration.

The aim of this study is to investigate the reasons of the mass mortality of economically valuable fish in Sultansuyu Reservoir (Malatya) in terms of hematology, biochemistry, microbiology and limnology.

Material and Methods

The economically valuable fish in Sultansuyu Dam Lake, namely, *Cyprinus carpio*, *Leuciscus cephalus*, *Capoeta trutta* and *Capoeta capoeta umbla* were studied. Hematological and biochemical analyses of the fish samples obtained from Sultansuyu Dam Lake between June 2007 and September 2007 were conducted; and water quality criteria, inorganic pollution parameters and microbiological analyses of the water were determined for the same period. Sultansuyu Dam Lake, which has been dammed up for irrigation purposes, is located 28 km west of Malatya at an elevation of 930 m. It has a wall elevation of 60 m and an average capacity of 53.3 hm^3. It has been actively in service since 1994 as the irrigation unit for Kuzova (Figure 1).

Determination of the Hematological and Biochemical Parameters

Blood samples (approximately 2 ml) were drawn from the caudal vein of each fish. Blood samples were stored in glass tubes containing anticoagulant (EDTA) stored in cooled bags and blood analyses were carried out immediately after sampling. The blood was centrifuged at 3,000 g, at 4°C for 5 minutes. Serum biochemical parameters metabolites; glucose, creatinin, urea nitrogen, Triglyceride, total cholesterol as mg dl^{-1} and total protein as g dl^{-1}; Serum biochemical parameters enzymes; Alanine transaminase/serum glutamate pyruvate transaminase (ALT/GPT), Aspartat Amino-transferase (AST/GOT), Laktat Dehydrogenase (LDH), alkaline phosphatase (ALP), Amilase (AML), gamma glutamil transferase (GGT) as IU L^{-1}. Serum biochemical parameters electrolytes; Chloride, sodium, potassium, total calcium and phosphatase mmol L^{-1} in blood plasma were assayed by Olympus AU 600 autoanalyser (Olympus Optical Corp, Shizuoka-ken, Japan), using commercially available kits (Roche).

Blood samples were stored in glass tubes containing anticoagulant (EDTA) and analyses were carried out immediately after sampling. The determination of eryhtrocytes and thrombocytes (per mm^3) in a blood sample was carried out by pipetting and diluting (1/200) the samples in Hayem solution. One drop of hemolysed blood was transferred onto Thoma lamella and examined under the light

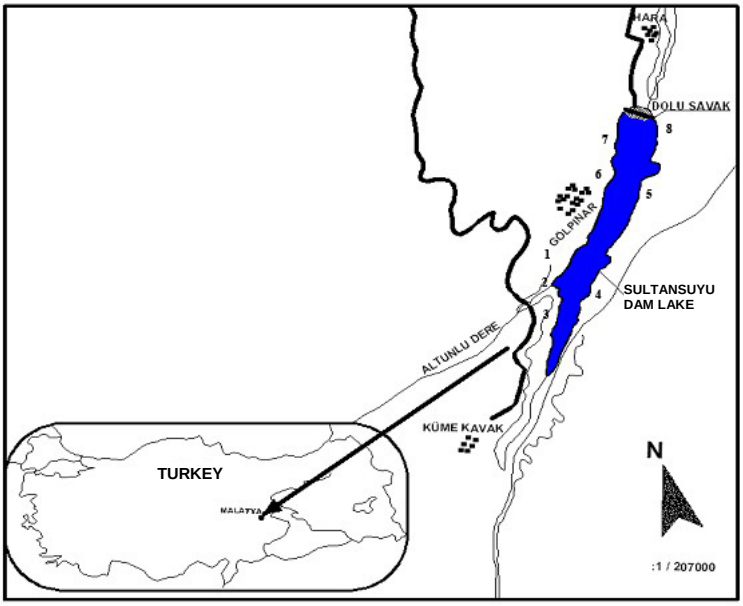

Figure 1. Map of Sultansuyu Reservoir and sampling points.

microscope (Soif, XZS-107B model) with a magnification of 400X. Leucocyte count was performed by transferring blood samples (diluted in Turck solution) with a leucocyte pipette onto counting lamella and examined as for erythrocytes (Blaxhall and Daisley, 1973). The amount of hemoglobin was determined according to the cyan-methemoglobin procedure (Kit 525-A, Sigma Chemical Co.). Non-clotted blood (20 µ) was diluted with 1 ml Drabkin solition and left to stand for 10 min at room temperature. The absorbance of the mixture was read at 540 nm and the amount of hemoglobin was calculated according to hemoglobin Standard (Kampen and Zijlstra, 1961). The microhematocrit method was utilized in hematocrit determination (Blaxhall and Daisley, 1973). Non-clotted blood was pipetted with a 85 microhematocrit pipette, centrifuged at 3000 g for 5 minutes and the ratio of blood components in plasma was determined.

Physical, Chemical and Microbiological Analyses of Water

General Physical and Inorganic Parameters: Temperature and pH (WTW pH330i/SET–Germany); dissolved oxygen (WTW Oxi340i/SET–Germany); chloride (4500 B–Argentometric Method); sulphate (14791 Merc Spectroquant–Germany); ammonium–nitrogen (14752 Merc Spectroquant–Germany); nitrite–nitrogen (14776 Merc Spectroquant–Germany); nitrate–nitrogen (09713 Merc Spectroquant–Germany); orto–phosphate, (14848 Merc Spectroquant-Germany); Commerical kits ThermoSpectronic–Aquamate Spectrophotometer-England; total dissolved material (WTW LF330/SET-Germany); conductivity (WTW LF330/SET-Germany); sodium, calcium and magnesium (Perkin Elmer Optima 2100 DV ICP-OES-USA); total hardness (2340-C. EDTA Titrimetric Method); alkalinity (2320-B Titrimetric Method); turbidity (WTW-TURB 355 IR–Germany) were measured.

Organic Parameters : Chemical oxygen demand (COD; 5220–D Closed Reflux, Colorimetric Method) measurements were done by using Spectro UV–VIS RS (USA) spectrophotometer. Biochemical oxygen demand for five days (BOD_5; WTW OxiTop–Germany); total organic carbon (TOC) and total nitrogen (TN; Shimadzu TOC-V_{CPN} total organic carbon analyzer) were also measured.

Inorganic Industrial Pollution Parameters : Mercury, cadmium, lead, arsenic, copper, chrome (total), chrome (6+), cobalt, nickel, zinc, sulfide, iron, manganese, boron, selenium, barium and aluminum element analyses were done by using Perkin Elmer Optima 2100 DV ICP OES–USA.

Microbiological Analysis: Fecal coliform (EMS/100 ml) (9222-D Fecal Coliform Membrane Filter Procedure) and Total Coliform (EMS/100 ml) (9222–B Standart Total Coliform Membrane Filtration Procedure) were determined according to Standard Methods for the Examination of the Water and Wastewater (APHA–AWWA–WEF, 2005).

Results

Water quality criteria obtained from Sultansuyu Dam Lake (Malatya) from June 2007 to September 2007 and the comparison of these values with water pollution control regulation are given in Table 1. When the water quality criteria obtained during the periods between June 2007 and September 2007 are compared, a significant statistical decrease can be observed in biological oxygen level, the soluted

Table 1. Water quality criteria of Sultansuyu Reservoir

Parameters	September 2007	June 2007	Water Quality Class			
	Normal	Mortality	I	II	III	IV
Soluted Oxygen(mg/L)	7.70±0.52	4.46±0.25				
BOD_5 (mg/L)	7.20±0.6[1]	5.20±0.2	4	8	20	>20
COD (mg/L)	17.60±1.4	35.80±1.5	25	50	70	>70
Temperature (°C)	18.0±0.2	19.6±0.2	25	25	30	>30
pH	8.043±0.1	7.14±0.1	6.5–8.5	6.5–8.5	6.0–9.0	out of 6.0–9.0
Electrical Conductivity (µS/cm)	352.0±5.6	412.0±7.6			–[2]	
Total Dissolved Solids (mg/L)	221.0±4.5	276.0±6.7	500	1500	5000	>5000
Turbidity (NTU)	43.69±2.5	51.10±4.6			–[2]	
Total Hardness (mg $CaCO_3$/L)	176.0±2.1	154.0±3.6			–[2]	
Nitrate-N (mg NO^-_3-N/L)	0.3547±0.001	6.78±0.4	5	10	20	>20
Total Organic Carbon (mg C/L)	10.86±1.2	22.70±1.7	5	8	12	>12
Total Kjeldahl Nitrogen (mg N/L)	1.557±0.03	6.74±0.4	0.5	1.5	5	>5
Chloride (mg Cl^-/L)	16.0±1.5	18.0±1.4	25	200	400	>400
Sulphate (mg SO_4^-/L)	60.40±0.1	230.60±0.3	200	200	400	>400
Ammonium-N (mg NH^-_4-N/L)	0.70±0.1	5.20±0.1	0.2[3]	1[3]	2[3]	>2

[1] The mean values in each row (MV±SD) are statistically different from one another $P<0.05$.
[2] This parameter is absent in the Water Pollution Control Regulation.
[3] The concentration of free ammonia-nitrogen should not be exceed the value of 0.02 mg NH_3–N/L.

oxygen level increased to 7.70 mg/L in the normal period whereas it was 4.46 mg/L in the massive mortality period, pH and total hardness values P<0.05, whereas a statistically significant increase can be seen in chemical oxygen level, electrical conductivity, total dissolved substance, turbidity, nitrate, total carbon, total nitrogen, sulfide and ammonium values P<0.05. The average values of copper and nickel in June are statistically different from the values in September P<0.05 (Table 2).

At the microbiological analyses conducted in June and September 2007, more than 20 enterocci colonies were detected in June. No significant growth of *E. coli* and *P. auroginesa* were observed (Table 3).

Serum biochemical parameters (Metabolite, enzyme and electrolyte) of four fish species (*Cyprinus carpio*, *Leuciscus cephalus*, *Capoeta trutta* and *Capoeta capoeta umbla*) from the period when mortality was observed (June 2007) and from the period when mortality was not observed (September 2007) in Sultansuyu Dam Lake are given in Table 4. In the period when high mortality rate was observed for all of the species (June 2007), a statistically significant increase was recorded for serum metabolites glucose, total protein, creatine and urea; and for the activities of the serum enzymes AST, ALT, LDH and AML; whereas a statistically significant decrease was observed for the amount of serum electrolyte serum potassium P<0.05.

In our study, the erythrocyte, hemoglobin and hematocrit values of the samples obtained in June 2007 were significantly less than the values in September (P<0.05). Total leukocyte count has increased in all of the species during the periods when high mortality rates were observed P<0.05. It was found out that the significant increase of the MCV value in *Leuciscus cephalus*, has turned into a micrositer anemia (Table 5).

Discussion

The study of the adverse effects of environmental pollution on fish species in terms of hematology and biochemistry contributes new and important information both to the literature and to efforts in studying animal health. Clinical tests such as these may provide useful data both for the determination of the reasons of possible mass mortality of animals and in the development of educational programs for environmental preservation.

Over the past years, various studies were conducted on the subject of environmental health. Especially in these studies the factors resulted in environmental pollution were focused on the fish which constitutes the important ring of our food chain. In general, these studies were conducted in relation to pesticides, heavy metals and industrial pollution, which are observed more frequently in environmental pollution.

Enterococcus is usually found in human feces in lesser numbers than *E. coli* and it cannot multiply in water as well. However, *Enterococcus*, which can maintain its vitality in water for longer periods than coli forms, are used as an indicator of fecal

Table 2. Inorganic pollution parameters of Sultansuyu Reservoir

Parameter (µg/L)	September 2007	June 2007	Water Quality Class			
	Normal	Mortality	I	II	III	IV
Cadmium	0.0±0[1]	0.0±0	3	5	10	> 10
Lead	10.2±0.3	10.2±0.3	10	20	50	> 50
Arsenic	10.7±10.5	10.7±10.5	20	50	100	> 100
Copper	10.2±10.2	60.2±10.2	20	50	200	> 200
Total Chromium	0.0±0	0.0±0	20	50	200	> 200
Cobalt	0.0±0	0.0±0	10	20	200	> 200
Nickel	18.1±11.6	120.1±14.1	20	50	200	> 200
Zinc	0.0±0	0.0±0	200	500	2000	> 2000
Iron	280.3±17.6	320.4±18.2	300	1000	5000	> 5000
Manganese	0.0±0	0.0±0	100	500	3000	> 3000
Boron	245.4±22.1	275.4±12.7	1000	1000	1000	> 1000
Barium	292.8±11.4	380.8±21.9	1000	2000	2000	> 2000
Aluminum	0.0±0	0.0±0	300	300	1000	> 1000

[1] The mean values in each row (MV±SD) are statistically different from one another P<0.05.

Table 3. Microbiological analysis of Sultansuyu Reservoir

Enterococci 37°C; 44±4st CFU/100 ml		*E. coli* (37°C; 44±4st)		*P. auroginesa* (37°C; 44±4st)	
September 2007	June 2007	September 2007	June 2007	September 2007	June 2007
+	+	-	-	-	-
<20	**>20**	**<20**		**<20**	

Table 4. Comparison of the biochemical parameters of some of the fish species in Sultansuyu Reservoir at mortality and postmortem

Serum Biochemical Parameter	*Cyprinus carpio*		*Leuciscus cephalus*		*Capoeta trutta*		*Capoeta capoeta umbla*	
Metabolites	Normal	Mortality	Normal	Mortality	Normal	Mortality	Normal	Mortality
Glucose (mg/dl)	75.4±1.51[1]	87.8±1.8	64.1±0.9	95.0±1.14	60.2±1.2	91.2±0.80	56.3±1.1	95.2±2.4
Total protein (g/dl)	4.5±0.09	3.1±0.06	4.2±0.08	2.9±0.04	3.7±0.06	2.5±0.08	3.4±0.05	2.3±0.02
Creatinin (mg/dl)	0.12±0.004	0.4±0.02	0.16±0.03	0.35±0.02	0.2±0.002	0.45±0.07	0.26±0.002	0.45±0.02
Urea nitrogen (mg/dl)	5.1±0.22	8.2±0.30	5.4±0.28	7.9±0.30	6.75±0.4	9.3±0.80	7.2±0.5	10.1±0.6
Triglyceride (mg/dl)	360.2±26.2	375.1±22.4	382.1±21.2	392.4±19.6	435.1±20.4	427.5±18.1	491.7±22.2	518.2±16.5
Total cholesterol (mg/dl)	179.3±10.5	182.2±9.7	184.4±12.2	181.2±10.4	231.4±14.5	228.4±12.6	264.7±19.4	290.7±22.6
Enzymes								
Alanine Transaminase (ALT / GPT)(IU/L)	32.4±6.2	56.8±5.1	27.2±3.3	45.6±2.7	20.6±2.8	41.8±3.1	18.7±2.1	32.4±3.6
Aspartat Aminotransferase (AST / GOT)(IU/L)	576.5±22.5	762.4±43.1	480.6±26.3	645.7±29.6	302.6±28.2	512.4±32.1	290.5±26.5	431.7±21.6
Laktat dehidrogenase (LDH)(IU/L)	764.2±40.2	866.5±35.2	812.1±14.1	942.4±22.5	870.2±62.6	967.6±40.3	874.7±81.5	1140.6±50.7
Alkaline Phosphatase (ALP)(IU/L)	51.3±4.9	44.20±5.6	49±2.2	43.8±2.4	38.5±1.8	33.2±2.6	34.1±2.1	32.7±3.4
Amilase (AML)(IU/L)	247.5±10.2	372.4±15.2	262.8±16.2	352.1±18.9	410.1±44.1	530.4±37.6	535.7±62.6	721.8±55.7
γ Glutamiltransferase (GGT) (IU/L)	4.9±0.91	6.4±1.6	5.60±0.28	6.3±0.7	9.4±2.2	10.5±1.1	10.2±1.7	11.3±1.8
Electrolytes (mmol/L)								
Chloride	129.2±3.6	134.1±2.4	126.5±1.7	131.2±2.5	120.6±1.1	121.7±1.4	118.7±2.01	119.1±1.7
Sodium	186.5±3.1	185.2±4.2	178.6±6.2	181.4±5.1	172.6±22.2	175.6±15.1	188.5±10.5	182.4±7.65
Potassium	4.2±0.8	2.4±0.3	2.1±0.1	1.2±0.1	3.2±0.4	1.7±0.2	2.7±0.5	1.1±0.1
T. Calcium	10.1±3.9	9.6±2.7	12.3±4.2	11.3±2.1	11.5±1.8	12.1±1.65	12.2±2.2	12.3±2.14
Phosphate	26.5±2.2	24.4±1.8	29.8±1.2	25.7±2.5	24.1±2.1	25.1±1.9	26.7±1.6	25.8±1.4

[1] The mean values in each row (MV±SD) are statistically different from one another P<0.05.

Table 5. Comparison of the hematological parameters of some of the fish species in Sultansuyu Reservoir at Mortality and Postmortem

Hematological Parameter	*Cyprinus carpio*		*Leuciscus cephalus*		*Capoeta trutta*		*Capoeta capoeta umbla*	
	Normal	Mortality	Normal	Mortality	Normal	Mortality	Normal	Mortality
T. Leukocyte ($mm^3/10^3$)	11.5±0.7[1]	18.1±2.3	12.5±2.9	25.4±1.3	14.3±1.9	23.4±1.1	17.5±2.4	26.1±1.6
Granulocyte (%)	81.2±1.1	70.2±0.8	76.2±1.7	66.5±1.1	72.1±1.33	60.1±1.7	66.4±2.6	58.1±2.1
Agranulocyte (%)	18.9±1.15	29.8±0.7	23.8±1.45	33.5±1.15	27.9±1.4	20.5±1.25	33.6±2.7	24.45±1.3
Erythrocyte ($mm^3/10^6$)	1.56±0.03	1.2±0.05	1.49±0.02	1.15±0.03	1.3±0.03	0.95±0.02	1.45±0.02	1.15±0.04
Hemoglobin (g/dL)	8.8±0.12	7.4±0.2	8.6±0.5	7.2±0.2	8.2±0.7	7.15±0.4	8.6±0.5	7.8±0.6
Hematocrit (%)	30.5±2.2	22.6±1.4	28.9±1.8	20.1±0.7	27.6±1.4	20.1±1.1	31.4±1.2	24.5±1.5
Erythrocyte Indexes								
MCV	195.5±6.21	188.3±4.1	193.9±4.75	174.8±5.2	212.3±7.1	211.5±7.2	229.5±8.1	213.1±7.4
MCH	56.4±3.7	61.6±2.92	57.7±2.9	62.6±3.1	63.0±3.75	75.2±4.1	67.8±4.2	67.8±3.9
MCHC	28.8±1.1	32.7±1.5	29.7±1.15	35.8±1.65	29.7±1.4	37.3±2.1	29.5±1.3	31.8±1.6

[1] The mean values in each row (MV±SD) are statistically different from one another P<0.05.

contamination in water. The amount of *Enterococcus* in Sultansuyu Reservoir has been found to be at a level which threatens health. The source of this contamination is thought to be the domestic wastes from the villages in the vicinity.

It was reported that pesticides such as Copper Sulfide ($CuSO_4$), paraquat (PQ) and methidathion (MO), cause tissue damage and stress in ordinary carp, thereby resulting in increases of serum LDH, glutamate oxaloacetic transaminase (GOT), Glutamat dehydrogenase and blood sugar (Asztalos *et al*., 1991).

When the hematological and biochemical effects of the heavy metal pollution in Blue Bay and bain des Dames regions of Iceland on fish species were investigated, the erythrocyte, hemoglobin and hematocrite values of the fish living in regions with heavy metal pollution were reported to have a statistically significant decrease (Elahee and Bhagwat, 2007). It was also reported that xenobiotics, which are among the environmental pollutants, change the hematological tables of fish at an abnormal level (Jee *et al*., 2005; Chandrasekara and Pathiratne, 2007); that a deformation is observed in the erythron series of organisms living in regions with heavy metal pollution (Houston, 1997); and that water quality criteria affect the multiplication levels of fish, depending on seasonal change (Rios *et al*., 2002).

Especially in the aquatic media with anoxia and hypoxia, mass fish mortality is observed. In order to

find out the reason in a timely manner, hematological and biochemical tests need to be used (Nikinmaa, 1992; Perry *et al*., 1996). Several researchers have already figured out how pesticides and heavy metals change the hematological and biochemical profiles of fish. It has been pointed out that pesticides which are frequently used in agriculture, such as Cypermetrin, Permetrin, Deltametrin, and Fenvalerat, change the serum biochemical parameters (metabolite, enzyme and electrolyte) of fish (Velisek *et al*., 2007; El-Sayed *et al*., 2007; Guz, 1998; Borges *et al*., 2007; Dobsikova *et al*., 2006). Pesticides cause hyperglisemi and hypoproteinemi in blood. Moreover, pesticides have been reported to cause irregular surface swimming patterns, paralysis, submersion; as well as affecting vitality percentage and increasing mucus ejection in gills (Bradbury and Coast, 1989).

The determination of serum AST and ALT, LDH enzyme activities is being used as a stress factor in fish (El-Sayed *et al*., 2007). It has been reported that with the pollution of aquatic media with metals, pesticides and domestic waste, lead to an increase in the serum AST, ALT and LDH enzyme activities of fish (Borges *et al*., 2007; Velisek *et al*., 2007; Houston, 1997).

In conclusion, the reasons of the sudden mass fish mortality in Sultansuyu Dam Lake were investigated in this study; and the hematological and biochemical changes in the fish as an indicator of the possible stress were determined. The physical, organic, inorganic and microbiological changes in the water were also found out. It can be said that the reasons for mortality of fish are; high Cooper concentration, organic pollution (Amonium, Suphate) and also low ogygen concentration in Sultansuyu Dam Lake.

When the water quality criteria was examinad, it was seen that the ratio of N group composite, water temperature,copper and nickel were high.Besides these, it can be considered that the massive mortality of the fish was caused by the decrease of soluted oxygen concentration.It can also be thought that the increase in nitrat group was caused by domestic waste and the increase in copper by pesticides in the water.Furthermore,nickel increased the electric conductivitiy in the water.So, all these factors can be supposed to increase the massive fish mortality between September and June.

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