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An Investigation on Some Haematological and Biochemical Parameters in *Capoeta trutta* (Heckel 1843) from Munzur River (Tunceli, Turkey)

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Abstract: Fishes are largely used for the assessment of aquatic environment quality and are accepted as bioindicators of environmental pollution. This study evaluated haematological and biochemical responses of *Capoeta trutta* captured in Munzur River, Tunceli, Turkey. Blood and biochemical parameters (Sodium (Na), Potassium (K), Calcium (Ca), Phosphorus (P), Chlorine (Cl) and Iron (Fe)) were determined. The values of Red Blood Cell (RBC) Haemoglobin (Hb), Haemotocrit (Ht), Mean Corpuscular Volume (MCV), Mean Corpuscular Haemoglobin (MCH), Mean Corpuscular Haemoglobin Concentration (MCHC), Platelet Count (PLT), Mean Platelet Volume (MPV), Plateletcrit (PCT), Platelet Distribution Width (PDW) from examined haematological parameters were found as 0.67±0.09 10^{12} L^{-1}, 13.5±0.90 g dL^{-1}, 14.78±0.19%, 218.40±4.29 fL, 194.75±15.36 pg, 108.52±1.09 g dL^{-1}, 34.12±5.89 10^{9} L^{-1}, 11.92±1.65 fL, 0.061±0.01% and 19.77±1.71 fL, respectively. The values of Fe, Cl, P, Ca, K and Na from examined biochemical parameters were determined as 91.75±28.27 g dL^{-1}, 92.00±4.02 mmol L^{-1}, 13.20±1.26 mg dL^{-1}, 11.65±0.74 mmol L^{-1}, 1.30±0.21 mmol L^{-1} and 132.5±3.90 mg dL^{-1} respectively. Observation of blood parameters allows the most rapid detection of changes in fish after the exposure to xenobiotics. It can conclude that hematological and biochemical parameters could be ranked as possible biomarkers of pollution.

Key words: Haematology, biochemistry, *Capoeta trutta*, Munzur river, biochemical parameter, bioindicator

INTRODUCTION

Biomarkers are defined as a change in a biological response, ranging from molecular to cellular and from physiological responses to behavioral changes which can be related to the toxic exposure or to the toxic effects of environmental chemicals (Depledge *et al*., 1995). On the other hand, biomarkers are responses to environmental effects that occur at higher levels of the biological organization than suborganism and can be measured in the individual, population, community (primary production, disruption of the nutrient cycle) and ecosystem levels (Walker *et al*., 2001). Fish and crustaceans are largely used for the assessment of aquatic environment quality and are accepted as bioindicators of environmental pollution (Borkovic *et al*., 2008). The physiological stress is clearly reflected by blood patterns of the experimented fish. Studies have shown that when the water quality is affected by toxicants, any physiological changes will be reflected in the values of one or more of the hematological parameters (Van Vuren, 1986). Thus, water quality is one of the major factors, responsible for individual variations in fish hematology, since they live in close association with their environment and are sensitive to slight fluctuation that may occur within their internal milieu (Casillas and Smith, 1977). The use of hematological parameters as fish health indicators has been proposed by Hesser (1960). Hematology is used as an index of fish health status in a number of fish species to detect physiological changes following different stress conditions like exposure to pollutants, diseases, metals, hypoxia, etc. (Blaxhall, 1972; Duthie and Tort, 1985). Therefore, hematological techniques are the most common method to determine the sub-lethal effects of the pollutants (Larsson *et al*., 1985). Evaluation of the haemogram involves the determination of the RBC, total white blood cell count (WBC), Ht, Hb, erythrocyte indices (MCV, MCH, MCHC), white blood cell differential count and the evaluation of stained peripheral blood films (Campbell, 2004). Mean cell hemoglobin

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concentration measure was used to assess the amount of red cell swelling (decreased MCHC) or shrinkage (increased MCHC) present (Milligan and Wood, 1982). The macro elements Ca, Magnesium (Mg), Na, K and P are essential to human health (Przybyl and Koligot, 1997a).

Microelements such as Zinc (Zn), Fe, Copper (Cu) and Manganese (Mn) which occur in physiological concentrations, play key roles in living processes and either an excess or deficit can disturb biochemical functions in both humans and animals (Przybyl and Koligot, 1997b).

The *Capoeta trutta* used in the present study came from Munzur River. *Capoeta trutta* is grown commercially in this reservoir. The present study was designed to determine whether some hematological and biochemical parameters could be useful indicators of the aquatic pollution in *Capoeta trutta*.

MATERIALS AND METHODS

The experiment was organized on Aquaculture Department, Fisheries Faculty and Faculty of Engineering, Department of Environmental Engineering on Tunceli University (Tunceli, Turkey).

Locality: The localities were shown in Fig. 1. The fish was caught from river mouth of Munzur River with gill net (20 m length and 10 cm diameter) on 10 April 2010.

Fish: In this study, wild fishes of *Capoeta trutta* (Heckel, 1843) were used for biomonitoring purpose. The fish (n = 5) was caught from their natural areas in Munzur River (Tunceli, Turkey). These fishes had been anaesthetized immediately 0.7 g L^{-1} benzocaine dissolved in ethyl alcohol (Sardella *et al*., 2004) and observed anesthesia of fish being deep sedation, losing of swimming actions and partial losing of equilibrium (Altun and Danabas, 2006).

Blood sampling: Blood samples were collected by piercing in the ventro-lateral side of the caudal peduncle with 5 mL disposable syringe and transferred to K3 EDTA tubes (2.5 mL) for hematological analyses and to gelled and vacuumed tubes (5 mL) for biochemical analyses. The tubes were closed and rinsed to prevent hemolysis and stored in cold until analysis (Das *et al*., 2004) (Table 1).

Analyses

Temperature and pH measurement: Temperature and pH of water was measured by pH meter. Water analyses were conducted at Tunceli Public Health Laboratories (Turkey).

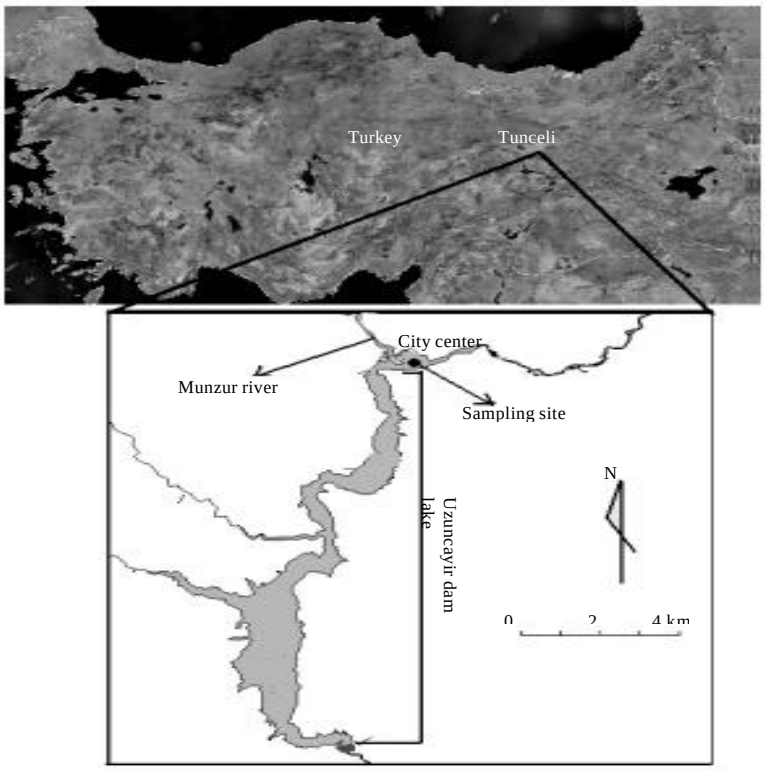

Fig. 1: Map of sampling site on Munzur river, Tunceli, Turkey

Table 1: Some biochemical blood parameters of *C. trutta* caught from sampling site on Munzur river

Parameters	Means±SE	Parameters	Means±SE
RBC (10^{12} L^{-1})	0.67±0.090	PCT (%)	0.061±0.01
HGB (g dL^{-1})	13.50±0.900	PDW (fL)	19.77±1.710
HCT (%)	14.78±0.190	Fe (g dL^{-1})	91.75±28.27
MCV (fL)	218.40±4.290	Cl (mmol L^{-1})	92.00±4.020
MCH (pg)	194.75±15.36	P (mg dL^{-1})	13.20±1.260
MCHC (g dL^{-1})	108.52±1.090	Ca (mg dL^{-1})	11.65±0.740
PLT (10^{9} L^{-1})	34.12±5.890	K (mmol L^{-1})	1.30±0.210
MPV (fL)	11.92±1.650	Na (mmol L^{-1})	132.5±3.9000

SE: Standart Error, n = 5

Hematological analyses: Blood parameters (RBC, HGB, HCT, MCV, MCH, MCHC, PLT, MPV, PCT and PDW) were determined by auto haematology analyzer (BC-5500).

Biochemical analyses: The blood sample was centrifuged at 4000 rpm for 20 min (centrifuge; Universal 320 R (Hettich Zentrifugen)) to separate the plasma and analyzed. Na, K, Ca, P, Cl and Fe were measured by kits (Lot numbers of kits; E485, E485, F133, F108, E485 and E389, respectively (purchased Thermo SCIENTIFIC)) using Konelab Prime 60I.

Statistical analysis: SPSS v13.0 statistical software was used for statistical analysis (SPSS Inc., Chicago, IL, USA). Data was statistically analyzed for means±standard error.

RESULTS AND DISCUSSION

In this study, haematological and biochemical parameters of *Capoeta trutta* in Munzur River were evaluated. The values of RBC, Hb, Ht, MCV, MCH, MCHC, PLT, MPV, PCT and PDW from examined haematological parameters were found as 0.67±0.09 10^{12} L^{-1}, 13.5±0.90 g dL^{-1}, 14.78±0.19%, 218.40±4.29 fL, 194.75±15.36 pg, 108.52±1.09 g dL^{-1}, 34.12±5.89 10^{9} L^{-1}, 11.92±1.65 fL, 0.061±0.01% and 19.77±1.71 fL, respectively. The values of Fe, Cl, P, Ca, K and Na from examined biochemical parameters were determined as 91.75±28.27g dL^{-1}, 92.00±4.02 mmol L^{-1}, 13.20±1.26 mg dL^{-1}, 11.65±0.74 mmol L^{-1}, 1.30±0.21 mmol L^{-1}, 132.5±3.90 mg dL^{-1}, respectively.

Since haematological parameters reflect the poor condition of fish more quickly than other commonly measured parameters and they respond quickly to changes in environmental conditions (Atkinson and Judd, 1978), they have been widely used for the description of healthy fish (Blaxhall, 1972) for monitoring stress responses (Soivio and Oikari, 1976) and for predicting systematic relationships and the physiological adaptations of animals. The increased RBCs count may be due to stimulation of erythropoietin by elevated demands for O_2 or CO_2 transport as a result of increased metabolic activity or destruction of gill membranes causing faulty gaseous exchange. The increase Hb content could be explained as a process where the body tries to replace the oxidized denatured Hb (Cyriac *et al*., 1989). The increase of HCT value and MCHC may be attributed to swelling of RBCs due to increased CO_2 in blood, hypoxia or stressful procedures (Ellis, 1981; Nemesok and Boross, 1999).

Kori-Siakpere and Ubogu (2008) investigated sublethal haematological effects of zinc on the freshwater fish, *Heteroclarias* sp. (Osteichthyes: Clariidae). Haemotocrite, haemoglobine, RBC, MCHC, MCH, MCV values were found in the range of 24.8-38.4%, 8.34-15.38 g dL^{-1}, 1.24-1.63 10^{6} mm^{-3}, 34.63-35.47%, 68.30-97.28 pg, 204.85-240.18 μg in blood of fishes, respectively. Fantin *et al*. (1988) investigated effects of acute experimental pollution by lead on some haematological parameters in *Carassius carassius* (L.) Var. *auratus*. The following differences were observed between control and treated samples; the number of RBC was decreased after 48 h of treatment; Ht, Hb percentage and MCHC were also decreased. Maximum and minimum RBC×10^{3} mrr^{-3}, Thrombocytes×10^{3} mn^{-3},HCT (%), HGB (%), MCV (μm^{3}) ,MCHC (pg) values were found to be 1634.00-1488.00, 70.18- 63.21, 26.7-24.3, 11.9-9.9, 168.71-161.08, 43.65-40.5, 70.9-69.6. in blood of fishes, respectively. Kandemir *et al*. (2010) determinated heavy metal levels, oxidative status, biochemical and hematological parameters in *Cyprinus carpio* (L., 1978) from Bafra (Samsun) fish lakes. They show that heavy metals levels may increase in fishes living in Bafra Lakes because of both domestic waste water and agricultural activities. Atamanalp and Yanik (2003) investigated alterations in hematological parameters of rainbow trout (*Oncorhynchus mykiss*) exposed to mancozeb. It was determined that the use of the pesticide caused a slight increase in RBC numbers and a decrease in Hb, MCH, MCHC, PCV, MCV and WBC levels. Significant decreases in Hb content and in MCH were observed during exposure to the pesticide.

In this study, the values of RBC, Hb, Ht, MCV, MCH, MCHC, PLT, MPV, PCT, PDW from examined haemetological parameters were found as 0.67±0.09 10^{12} L^{-1}, 13.5±0.90 g dL^{-1}, 1478±0.19%, 218.40±4.29 fL, 194.75±15.36 pg, 108.52±1.09 g dL^{-1}, 34.12±5.89 10^{9} L^{-1}, 11.92±1.65 fL, 0.061±0.01% and 19.77±1.71 fL, respectively.

Atamanalp *et al*. (2002) investigated that the effects of cypermethrin (a synthetic pyrethroid) on some biochemical parameters (Ca, P, Na and TP) of rainbow trout. After 15 days exposure, Ca and P decreased while TP and Na took various values depending on the doses of synthetic pyrethroid. Zeynali *et al*. (2009) determinated the Cu, Zn and Fe levels in edible muscle of three

commercial fish species from Iranian coastal waters of the Caspian Sea. Mean concentrations of Fe in muscles of mullet, sefid and common carp were 81.11, 73.59 and 94.78 mg kg^{-1}, respectively. Three species fish samples collected from Bangladesh and Ca, Na, Mg and Fe concentrations were found in the range of 3650-6570, 2950-4580, 2060-2560 and 71-186 μg g^{-1} in the muscle tissue of fishes, respectively (Begum *et al*., 2005). Mendil *et al.* (2010) investigated the trace metals in different fish species and sediments from the River Yesilirmak in Tokat, Turkey.

Luczynska *et al.* (2009) analyzed muscle of four fish speices such as, roach, bream, perch and pike and they found Fe concentrations 1.877, 1.881, 1.083, 0.987 mg kg^{-1} wet weight, Na concentrations 51.3, 55.1, 61.4, 62.0 mg kg^{-1} wet weight, K concentrations 437.1, 421.2, 415.7, 425 mg kg^{-1} wet weight, Mg concentrations 22.8, 19.4, 19.8, 22.8 mg kg^{-1} wet weight, Ca concentrations 17.2, 14.0, 13.1, 11.0 mg kg^{-1} wet weight, P concentrations 234.1, 231.8, 233.4, 238.0 mg kg^{-1} wet weight, respectively. The concentrations of macro elements were noted in the following order: K>P>Na>Mg> Ca in different fish muscle tissues samples. Roach and bream had more Fe than perch and pike (Luczynska *et al*., 2009).

Fantin *et al*. (1988) assayed Na^{+}, K^{+}, Cl^{-}, Ca^{++} in specimens of *Carassiu carassius* (L.) var. *auratus* submitted to sublethal acute exposure to lead (5 ppm., 24 and 48 h). K, Na, Cl and Ca concentration (meg mL^{-1}) in reference site were determined as 7.04, 131.5, 106.2, 3.57, respectively but in polluted site K, Na, Cl and Ca concentration (meg mL^{-1}) were found as 8.98, 127.8, 100.9, 3.61, after 24 h, 7.81, 132.5, 109.1, 2.9, after 48 h (Fantin *et al*., 1988).

As a result of the study, the values of Fe, Cl, P, Ca, K, Na from examined biochemical parameters were determined as 91.75±28.27g dL^{-1}, 92.00±4.02 nmol L^{-1}, 13.20±1.26 mg dL^{-1}, 11.65±0.74 mmol L^{-1}, 1.30±0.21 mmol L^{-1}, 132.5±3.90mg dL^{-1}, respectively.

CONCLUSION

To the knowledge this is the first report show in that haemetological and biochemical parameters of *Capoeta trutta* from Munzur River. Results of these investigations suggest that the intensity of pollution is responsible for altering the haematology and biochemistry of *Capoeta trutta* in Munzur River. Haemotological and biochemical parameters of fish should be checked regularly for food safety and environmental pollution.

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