

## HEMATOLOGICAL CHARACTERISTICS OF *TELESTES METOHIENSIS* (STEINDACHNER, 1901) FROM DIFFERENT HABITATS

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### HEMATOLOŠKE KARAKTERISTIKE *TELESTES METOHIENSIS* (STEINDACHNER, 1901) SA RAZLIČITIH STANIŠTA

#### *Apstrakt*

*Telestes metohiensis* (Steindachner, 1901) predstavlja endemičnu vrstu riba područja Istočne Hercegovine, sa specifičnim životnim ciklusom i prilagođenostima da određeni dio godine provodi pod zemljom, a sa pojavom velikih voda izlazi na površinu. Istraživanja fizioloških i ekofizioloških karakteristika ove vrste imaju poseban značaj jer se radi o nedovoljno istraženoj vrsti, pri čemu su podaci ovoga karaktera oskudni i fragmentirani, a radi se o vrsti koja se obuhvaćena Crvenom listom Republike Srpske, dok je dvije najveće baze ovih podataka WCMC i IUCN svrstavaju je u prvu kategoriju ranjivih vrsta. Fiziološka istraživanja endemičnih vrsta riba su od posebnog značaja, jer daju uvid u niz procesa u organizmu, a posredno i u životnoj sredini, što je neophodno pri prilikom planiranja i provođenja mjera zaštite. Hematološki parametri predstavljaju pouzdane indikatore stanja organizma, a posredno i stanja životne sredine. Za utvrđivanje hematološkog statusa koriste se kvantitativni karakteri ćelija crvene i bijele loze, koji pružaju uvid u čitav niz procesa u organizmu, a na osnovu analize različitih komponenti krvi može se suditi o promjenama koje nastaju u određenim sistemima pod uticajem faktora spoljašnje i unutrašnje sredine. Hematološki parametri su obuhvatali: broj eritrocita, koncentraciju hemoglobina, hematokrit, MCV, MCH i MCHC. Praćeni parametri su analizirani kod jedinki gatačke gaovice koje su lovljene u vodotocima: Vrijeka, Opačica i Zalotka. Prve dvije rijeke se nalaze na području Dabarskog polja, dok rijeka Zalotka teče kroz Nevesinjsko polje. Rezultati pokazuju postojanje značajnih razlika u vrijednostima praćenih parametara prilikom komparacije po lokalitetima, pri čemu su jedinke iz rijeke Vrijeka imale veće vrijednosti koncentracije hemoglobina, vrijednost hematokrita, broja eritrocita i srednje vrijednosti količine hemoglobina u eritrocitima (MCH) u odnosu na druga dva vodotoka.

*Ključne riječi: Telestes metohiensis, gaovica, hematološke karakteristike, endemi, ranjive vrste*

*Key words: Telestes metohiensis, striped pijaon, hematological characteristics, endemic species, vulnerable species*

## INTRODUCTION

Freshwater fish fauna of Bosnia and Herzegovina have a high diversity. Vuković (1977) cites 108 fish species in Bosnia and Herzegovina, while according to recent data, there are 118 fish species (Sofradžija, 2009). Ichthyofauna of B&H is characterized by a large number of endemic species many of which live just in particular localities. Most of the endemic species have restricted distribution areas which is the main reason for their vulnerability (Glamuzina et al., 2010; Dekić et al., 2011). Physiological and morphometric studies of endemic fish species are of particular importance because adequate knowledge is necessary for their protection and preservation (Ivanc, 2012). Endemic fish species are particularly vulnerable and they often have lower ability to adapt to changes in environmental conditions. They are mainly distributed in the former glacial remains and they are closely related to the environment which they inhabit (Glamuzina, 2013). The aim of research was to determine the hematological characteristics of striped pijaon from three rivers in the Eastern Herzegovina.

## MATERIAL AND METHODS

*Telestes metohiensis* (Steindachner, 1901) is an endemic fish species characteristic for Eastern Herzegovina. They have a specific life cycle and adjustments that allow them to spend one part of the year in groundwater. With increasing of water level they come out to the surface water. Striped pijaon lives in karst habitats such as lakes and watercourses with low flow in lowland areas, but also at higher altitudes. However, during the summer months, it generally remains in the groundwater flows because surface waters dry out. During winter, pijaon also enters the groundwater flows where it rests in mud. Before going into the groundwater, specimens form large flocks. Electrofishing was carried out from 6 to 8 September 2013, at localities Opačica, Vrijeka and Zalotka. Electrical aggregates IG 600, power 1.2 kW and ELT62II GI HONDA GCV160, power 3 kW, were used for this purpose. River Opačica is an occasional watercourse of the field Dabarsko polje. At the time of high water Opačica is torrent, and during summer it dries up. It appears in the northwestern part of the field by merging the watercourses Bijeli Potok and Trusina, and it goes underground in the part of the field which is called Lužine Bare. Water from Opačica occurs later at the springs of the watercourses Bregava and Tebišnjica after underground bifurcation (Dekić, 2013). Spring of the river Vrijeka is located at the edge of the field Dabarsko polje. Most of the river dries up during warm summer months, and water is present only in spring (Gnjato, 2004). The length of river is about 2.5 km, and it flows through the field Dabarsko polje along the entire watercourse. Zalotka is a river that flows through the field Nevesinjsko polje, at southeast part of Bosnia and Herzegovina. Spring of this river is near the field Gatačko polje. River flows through the Nevesinjsko polje and plunges into numerous chasms. During the dry season it dries up, while during the wet, rainy period, it is rich in water.

Zalomka flows through the area which represents a tectonic depression which is limited by the mountain ranges (Vuković, 2014).

### *Hematological analysis*

Blood for hematological analysis was collected by heart puncture using sharp and wide sterile needle (1.0 to 1.2 mm), by applying the rules of the sterile work. Native blood, with any anticoagulant, is used for further analyses. Erythrocyte count was performed in the hemocytometer using diluent by Kekić and Ivanc (1982). Hemoglobin concentration (Hb) was determined by hemoglobin cyanide method using Drabkin reagent (Blaxhall and Daisly, 1973). Hematocrit (Hct) was determined by microhematocrit centrifuge. Haematological indices were calculated using values of hematocrit, erythrocyte number and hemoglobin concentration.

$$\text{Mean corpuscular volume (MCV)} \quad MCV = \frac{Hct}{Br. \text{ eritrocita/l}}$$

$$\text{Mean corpuscular hemoglobin (MCH)} \quad MCH = \frac{Hb/l}{Br. \text{ eritrocita/l}}$$

$$\text{Mean corpuscular hemoglobin concentration (MCHC)} \quad MCHC = \frac{Hb/l}{Hct}$$

## RESULTS AND DISCUSSION

Results of the research of haematological parameters are presented in Table 1. The sample from the river Opačica contained 30 individuals, from the river Vrijeka 20 individuals and from the watercourse Zalomka 11 individuals. Comparing the average values of the parameters of erythrocyte lineage among individuals from three different watercourses, it is evident that these groups differ in most of the monitored parameters. Mean corpuscular hemoglobin concentration was significantly higher in specimens from the river Vrijeka in relation to individuals from the river Opačica ( $p = 0.002$ ) and to individuals from the river Zalomka ( $p = 0.000$ ). Significantly higher values of hemoglobin had also individuals from the river Opačica in relation to specimens from the river Zalomka ( $p = 0.009$ ).

**Table 1.** Parameters of erythrocytes lineage of striped pijaon from rivers Opačica, Vrijeka and Zalomka

River	Statistical parameters	Hb g/l	Hct l/l	Number of erythrocytes $\times 10^{12}/l$	MCV fl	MCH pg	MCHC g/l erit.	
Opačica	Mean value	82,35 <sup>bc</sup>	0,410 <sup>b</sup>	1,533 <sup>b</sup>	270,92	54,69 <sup>c</sup>	203,90	
	Standard deviation	11,577	0,058	0,182	48,900	11,357	37,059	
	Minimim	66,67	0,333	1,250	182,149	36,430	155,555	
	Maximim	114,81	0,500	1,960	384,615	90,405	344,444	
	95 % Confidence Interval for mean	Lower bound	78,02	0,386	1,463	252,657	50,449	190,064
		Upper bound	86,66	0,430	1,599	289,176	58,926	217,740
	Coefficient of variation %	14,060	14,045	11,934	18,050	20,755	18,175	
Vrijeka	Mean value	92,77 <sup>ac</sup>	0,449 <sup>a</sup>	1,651 <sup>a</sup>	276,48	57,14 <sup>c</sup>	211,36	
	Standard deviation	11,741	0,055	0,165	33,236	7,885	34,227	
	Minimim	66,67	0,330	1,350	214,286	43,150	148,140	
	Maximim	103,70	0,560	2,060	340,136	68,680	259,250	
	95 % Confidence Interval for mean	Lower bound	87,28	0,420	1,573	259,390	53,465	193,767
		Upper bound	98,27	0,477	1,728	293,567	60,818	228,963
	Coefficient of variation %	12,654	12,327	10,050	12,021	13,799	16,194	
Zalomka	Mean value	71,72 <sup>ab</sup>	0,419	1,531	274,21	46,83 <sup>ab</sup>	181,08	
	Standard deviation	8,490	0,084	0,078	56,199	5,106	60,540	
	Minimim	59,30	0,227	1,390	141,163	37,745	118,518	
	Maximim	85,19	0,500	1,610	335,570	53,576	342,222	
	95 % Confidence Interval for mean	Lower bound	66,015	0,363	1,479	236,452	43,403	140,411
		Upper bound	77,423	0,430	1,584	311,962	50,263	221,754
	Coefficient of variation %	11,838	20,061	5,110	20,495	10,902	33,432	

*a,b,c* Different superscript letters indicate statistically significant difference ( $p < 0.05$ ) compared to specimens from different localities: a-Opačica, b-Vrijeka and c-Zalomka.

The average hematocrit value was significantly higher in specimens from the river Vrijeka in relation to individuals from the river Opačica ( $p = 0.039$ ). The same relation was seen between average values of numbers of red blood cells ( $p = 0.014$ ) in specimens from these two localities. The average value of mean corpuscular hemoglobin (MCH) was significantly lower in individuals from the river Zalomka in relation to individuals from the river Opačica ( $p = 0.022$ ) and in relation to individuals from the river Vrijeka ( $p = 0.005$ ). MCHC and MCV values are approximately equal in individuals from all three watercourses. Parameters of erythrocytes lineage of striped pijaon show that individuals from the river Vrijeka have highest values of all parameters. On the other hand, comparing the quality of water among these watercourses indicate that they are approximately the same. The river Vrijeka had slightly different values of conductivity,  $BOD_5$  and concentration of total suspended solids. Variations in number of blood elements in fish depend on the species of fish (Langston et al., 2009), environmental conditions (Hickey 1982; Aldrin et al., 1982), nutritional status (Casillas and Smith, 1977), sex (Sidiqie and Nasim, 1979 ; Collazos et al., 1998), size of fish (Garcia, 1992) and seasonal changes (Cech and Wohlschlag, 1981). All these issues should be considered in order to determine hematological values of specific species. When we compare our results with results obtained for striped pijaon from the river Pribitil, we can conclude that the average value of hemoglobin concentration in specimens from the river Vrijeka are higher compared to individuals from Pribitil, whose value was 83.54 g/l. While specimens from the river Opačica had an approximate value, individuals from the river Zalomka had slightly lower value of hemoglobin concentration compared to individuals from watercourse Pribitil (Dekić et al., 2012). Hematocrit values in blood of striped pijaon from watercourse Pribitil ranged from 0.294 to 0.545 l/l, and the average value of this parameter was 0.411 l/l (Dekić et al., 2012). Compared with our results, only individuals from the river Vrijeka had slightly higher values, while individuals from two other watercourses showed approximate values. The average value of the number of red blood cells in striped pijaon from watercourse Pribitil was  $1,747 \times 10^{12}$  /l (Dekić et al., 2012), which was slightly higher compared to all three samples we analyzed. However, the number of erythrocytes varies in certain physiological limits and can be changed depending on the physiological state of an organism.

Data on the number of red blood cells in fish are different (Dekić et al., 2012). The number of erythrocytes in striped pijaon is relatively high because the majority of our freshwater fish species have lower values of this parameter (Dekić et al., 2012). Mean corpuscular volumes for all three of studied populations are higher than the values of MCV determined in fish from watercourse Pribitil 236,79 fl (Dekić et al., 2012). The average value of MCH in specimens from Pribitil was 48.42 pg (Dekić et al., 2012). Specimens from rivers Vrijeka and Opačica had significantly higher values of this parameter, but individuals from the river Zalomka had slightly lower values of MCH. The average value of MCHC in specimens from Pribitil was 211.32 g/l (Dekić et al., 2012). Specimens from the river Vrijeka had significantly higher values of this parameter, but individuals from rivers Opačica and Zalomka had slightly lower values of MCHC. When we compare results of monitored parameters of erythrocyte lineage of fish in different watercourses with our study, we can see that other authors had similar results. Studies of haematological parameters of carp, grayling (Ivanc et al., 1993; 1994) and trout (Kekić, 1985) showed slightly different values for individuals which live in various environmental conditions. There are significant differences of hematological parameters between balkan barbel from rivers Suturlija and Jakotinska rijeka

(Dekić et al., 2009). Significant differences of the total number of red blood cells, hematocrit values and hematological indices: MCH and MCHC were observed between chub from rivers Krupice and Željeznice (Mitrašinović et al., 2009).

## CONCLUSIONS

Observed hematological parameters of striped puior from three watercourses show significant differences for most parameters. They all have relatively high values of the number of red blood cells and high concentration of hemoglobin.

Comparing parameters of erythrocyte lineage it can be concluded that individuals from the river Vrijeka had higher concentrations of hemoglobin, hematocrit concentration, the number of red blood cells and the mean corpuscular hemoglobin (MCH) in relation to the individuals from other two rivers.

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