

## Morphometric and Physiological Characteristics of Brown Trout (*Salmo trutta*) from the Ponor River

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### Abstract

This paper presents the data related to morphometric and physiological (hematological) characteristics of brown trout (*Salmo trutta*) from the Ponor River. This river rises near the village Podrašnica (municipality of Mrkonjić Grad), sinks above the ground and after a while, near the settlement of Krupa na Vrbasu, appears as a source of the Krupa River (tributary of the Vrbas River). Fish sampling was performed during the summer of 2015 and during this period 22 *Salmo trutta* units were caught (11 females and 11 males). On that occasion, several morphometric (total and standard body length) and hematological traits (Hct-hematocrit, Hb-hemoglobin and MCHC - mean corpuscular hemoglobin concentration), body mass and Fulton's condition factor were analyzed. The average value for total body length was 18.85 cm and for standard body length it was 16.85 cm; the mean value for body mass was 80.38 g, and for Fulton's condition factor 1.41. In case of hematological parameters, the average value for Hct was 0.39 l/l, Hb 70.62 g/l and MCHC 180.64 g/l erythrocytes. The estimated parameters can serve as indicators for the condition of these aquatic organisms, and indirectly, the state of their environment.

*Key words:* brown trout, morphometric and hematological characteristics

## Introduction

The organic species in the natural habitat conditions are constantly under a direct or indirect influence of environmental factors, which are reflected in the physiological processes of individuals. It is known that fish are vertebrates with large variations in the values of hematology parameters (Homątowska et al., 2002), and with large variations of physiological responses to the effects of stressors (Barton, 2002). These parameters are specific to the species (Fazio et al., 2012) and variable under the changed conditions of external and internal environment. The analysis of blood parameters in terms of understanding the functioning of organisms and adaptation of organisms to physical and chemical factors of the environment can serve as a reliable indicator of the state of the environment of different water bodies (Dekić et al., 2009). In addition, erythrocyte status of fish is a very reliable indicator of different sources of stress, even during extended periods (Řehulka & Adamec, 2004) and it is very sensitive to environmental pollution (Witeska, 2013). The importance of analysis of the state of fish in aquatic ecosystems is reflected in the fact that they are an important food source for the human population, and being the final link in the food chain, they accumulate all the negative changes in their environment (Serpunin & Likhatchyova, 1998). Because these are the fastest ways to detect stress in fish analysis of hematological parameters, such tests have a great importance, especially for aquaculture (Radu et al., 2009). To be certain that physiological parameters of fish could be used for such purposes, and the biomonitoring of aquatic ecosystems, it is necessary to obtain reference values of hematological parameters, and compare and match different analysis (Hlavova, 1993c). Based on morphometric and hematological parameters of brown trout from the Ponor River, the aim of this study was to obtain preliminary results of the status of this species, as well as the assessment of the quality of its habitat.

## Materials and Methods

The Ponor River (Fig. 1.) rises near the village of Podrašnica (the municipality of Mrkonjić Grad), and after a few kilometers it sinks above the ground beneath the southwestern slopes of the Mount Manjača in Podrašničko field in the form of a typical karst abyss. It appears near the settlement of Krupa na Vrbasu (the municipality of Banjaluka) as a source of the Krupa River (tributary of the Vrbas River). The total length of the river is 12.4 km and the watercourse over plains has a total drop of 60 m (Crnogorac et al., 2013).

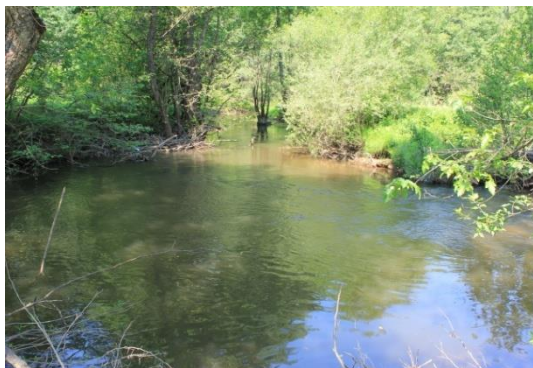


Fig. 1. The Ponor River  
*Ријека Понор*

### Fish sampling

Fish sampling was carried out by using electrofishing device DC (ELT 62 II GI, 3 kW) and fishes were collected by a hand net, after galvanotaxis and galvanonarcosis. After that, individuals were transferred into fyke net on sampling sites for resuscitation and adaptation after stress. Fish sampling was performed in the morning period because of the impact on the daily fluctuations of hematological parameters (Dekić et al., 2013).

### Morphometric analysis

To determine the body mass, a digital scale (accuracy 0.1 g) was used and to determine standard and total body length, we used the ichthyometer. Sex is determined by examining gonads after specimen dissection. Morphometric measurements were done after blood sampling and sex was determined several hours after fish sampling. Fulton's condition factor (K) was calculated by using the following formula:

$$K = W * 100/L^3, \text{ where:}$$

W - weight of the fish (g),

L - standard length of the fish (cm).

### Hematological analysis

Blood sampling was carried out in the same period and with the rules of a sterile work. The heart puncture was carried out at a disinfected injection site with sharp and wide sterile needle (from 0.8 to 1.2 mm) and for the further analysis we used the blood with anticoagulant.

To determine the concentration of hemoglobin, we used Drabkin hemoglobin cyanide method (Blaxhall & Daisly, 1973) based on the absorbance of the standard blood. Hematocrit was determined by centrifugation with microhematocrit centrifuge. We also determined a MCHC (mean value of hemoglobin in one liter of red blood cells) according to the following formula:

$$MCHC = \frac{Hb/l}{Hct}$$

### Statistical analysis

Statistical analyses were obtained by using Microsoft Office Excell 2010- Statistical Analysis Tools and t-test for data comparison ( $p < 0.05$ ).

## Results and Discussion

### Morphometric characteristics

During the field sampling of fish, we caught and analyzed 22 specimens of brown trout (11 males and 11 females). The results of morphometric measurements and Fulton's condition factor are given in Table 1.

Tab. 1. Statistical index of morphometric characteristics and Fulton's coefficient of brown trout from the Ponor River

*Статистички индекси морфометријских особина и Фултонов коефицијент за поточну пастрмку из ријеке Понор*

Statistical parameters* <i>Статистички параметри</i>	Standard length (cm) <i>Стандардна дужина (cm)</i>	Total length (cm) <i>Укупна дужина (cm)</i>	Mass (g) <i>Маса (g)</i>	Fulton's condition factor <i>Фултонов коефицијент</i>
Mean	16.85	18.85	80.38	1.41
SD	4.76	5.24	64.51	0.11
MIN	11.10	12.50	20.94	1.13
MAX	26.30	29.80	249.38	1.56
CV	28.25	27.79	80.25	7.80

\*Mean (mean value), SD (standard deviation), MIN (minimal value), MAX (maximal value), CV (coefficient of variation)

Mean value of Fulton's condition factor for brown trout from the Ponor River was 1.41. The mean values of this parametar for brown trouts from rivers of Krušnica, Svetinja and Una varied from 0.94 to 1.31 (Šarić, 2015).

Trožić-Borovac (2002) states that this index for brown trout on fifteen sites in the Una River Basin (area of Bihać) ranged from 1.00 to 1.59. In the Vrbas River, mean Fulton's condition factor of brown trout was 1.20 (Bek, 2013). By comparing the value of this parameter with these analyses, it can be concluded that the brown trout from the Ponor River were in good condition.

### Hematological parameters

Mean values of erythrocyte status parameters for brown trout from the Ponor River, and other statistical indicators are given in Table 2. It should be noted that the sample of brown trout, on which hematological characters were analyzed, comprised 15 individuals (9 males and 6 females).

Tab. 2. Statistical index of erythrocyte status of brown trout from the Ponor River  
*Статистички параметри особина еритроцита за поточну пастрмку из ријеке Понор*

Statistical parameters <i>Статистички параметри</i>	Hb (g/l)	Hct (l/l)	MCHC (g/l erit.)
Mean	70.62	0.394	180.84
SD	8.34	0.056	19.04
MIN	59.26	0.333	155.56
MAX	81.48	0.500	232.80
CV	11.81	14.21	10.52

Mean value of concentration of hemoglobin was 70.62 g/l. Comparing these results with the surveys conducted in the catchment of the Una River (Šarić, 2015), where the higher value for mean concentration of hemoglobin was 74.81 g/l, and the lowest 48.51 g/l it could be said that this parameter, for the fish from river Ponor, was at a high, satisfactory level. Pavlović (1962) established seasonal variations in the amount of hemoglobin of brown trout in the area of the Vrelo Bosne springs with the following average values: for the period of winter 67.0 g/l, spring 71.7 g/l, summer 64.0 g/l and autumn 69.4 g/l. Hlavova stated several mean values of brown trout hemoglobin: 65.1 g/l (1988), 72.1 (1993a) and 67.4 (1993b). Ivanc and Dekić (2012) reported lower limit of the amount of hemoglobin 65.28 g/l and upper of 99.24 g/l for brown trout at 3 locations in the Republic of Srpska. We may infer that the results of this research are closer to the lower limit values. The amount of hemoglobin of brown trout farmed (average of 64 g/l) coincides with the above bibliographical data (Romestand et al., 1983).

When it comes to the volume of hematocrit, the average value of this parameter for trout from the Ponor River was 0.393 l/l. Average hematocrit values of brown trout from rivers of Krušnica, Svetinja and Una varied from 0.399 l/l to 0.377 l/l (Šarić, 2015). In Hlavova research (1988) hematocrit values of brown trout from the river of Svratka were average 0.350, while in the later research Hlavova (1993a) stated that this parameter had mean value of 0.340 l/l on two localities, Bily stream and the Svratka River. In a review of hematological parameters of indigenous fish species of the Republic of Serbia, Ivanc and Dekić (2012) reported the mean hematocrit values for brown trout from three sites: 0.378 l/l, 0.399 l/l and 0.418 l/l.

Romestand et al. (1983), mentioned the average hematocrit value of farmed trout, which was 0.400. Clauss et al. (2008) stated that values of hematocrit greater than 45% included polycythemia phenomenon that may be caused by dehydration, an increased amount of protein, chlorides, etc., while values under 20% indicated anaemia. It could be noted that the sample of fish from our research had no health problems in terms of polycythemia or anaemia.

The mean value of hemoglobin quantity in red blood cells of liter of brown trout from the Ponor River was 180.84 g/l; the mean value of the same parameter in the three studies performed at the Una catchment varied from 136.56 g/l to 177.26 g/l (Šarić, 2015). In Hlavova research on the locality of Bily potok this parameter had an average value of 210 g/l, and at the Svratki River, this parameter was 190 (1993b). Also, Hlavova (1993a) stated that the maximum value of MCHC appeared in the winter, and that it was hematologic characteristics of brown trout. Ivanc and Dekić (2012) reported the mean MCHC values of brown trout from three locations in the RS, which amounted as follows: 200.23 g/l, 209.22 g/l i 216.46 g/l. For farmed brown trout, MCHC value amounted to 182 g/l (Romestand et al., 1983). The value of this parameter for brown trout from the Ponor River was closer to the upper values in relation to the previous states.

When comparing the analyzed hematological parameters between the sexes, statistically significant differences were established for hematocrit (0.0006), while the hemoglobin concentration was at the limit of statistical significance (0.052). Hlavova (1993a) mentioned some statistically significant differences between the sexes for brown trout hematocrit values ( $p = 0.0004$ ), and Luskova (1998) highlighted that hematocrit and hemoglobin values were always higher in males compared to females, which coincided with our results. This fact can be explained by the metabolic needs of females to develop ovaries and eggs, which can be associated with a voracious diet before and after the spawning period, which causes a temporary anaemia. Unlike them, the males were fed with the same intensity throughout the whole period (Joshi, 1980).

## Conclusion

While researching some morphometric and hematological characteristics of brown trout (*Salmo trutta*) from the Ponor River, we established that fish were in good condition ( $K = 1.41$ ). In addition, the examined hematological parameters (Hb, Htc and MCHC) were within the reference values and were not indicative of significantly altered state of water quality.

A statistically significant difference between the sexes was recorded only for hematocrit (Htc) ( $p = 0.0006$ ) that was higher in males than in females, which coincided with the results of similar studies. Hematologic indicators have great application significance in assessing the health status of fish as well as the quality of their environment. At the same time, the value of the concentration of hemoglobin and hematocrit indicate changes such as anemia and infection, often caused by water pollutants, such as nitrates, nitrites, heavy metals and pesticides.

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# Морфометријске и физиолошке особине поточне пастрмке (*Salmo trutta*) из ријеке Понор

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## Сажетак

Овај рад представља анализу података о морфометријским и физиолошким (хематолошким) особинама поточне пастрмке (*Salmo trutta*) из ријеке Понор. Ова ријека извире близу села Подрашница (општина Мркоњић Град), затим понире у земљу и убрзо, близу насеља Крупа на Врбасу, извире као притока ријеке Крупа (слив ријеке Врбас). Узорковање риба је извршено током љета 2015, при чему су уловљене 22 јединке *Salmo trutta* (11 женки и 11 мужјака). Том приликом, неколико морфометријских (укупна и стандарда дужина јединке) и хематолошких (Hct – хематокрит, Hb – хемоглобин и МСНС – просјечна тјелесна концентрација хемоглобина) особина, као и маса јединке и Фултонов фактор су анализирани. Просјечна вриједност за укупну дужину јединке је била 18,85 cm а за стандардну дужину јединке 16,85 cm; просјечна маса јединке је била 80,38 g, а просјечан Фултонов фактор 1,41. Када су у питању хематолошки параметри, просјечна вриједност за Hct је била 0,39 l/l, за Hb 70,62 g/l а за МСНС 180,64 g/l еритроцита. Процијењени параметри могу послужити као индикатори тренутног стања ових акватичних организама и индиректно као индикатори стања окружења у којем живе.

*Кључне ријечи:* поточна пастрмка, морфометријске и хематолошке особине

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