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Research Article

GONADOSOMATIC INDEX OF THE ENDEMIC AND ENDANGERED *PHOXINELLUS PSEUDALEPIDOTUS* (LEUCISCIDAE, CYPRINIFORMES) FROM MOSTARSKO BLATO (NERETVA RIVER BASIN, BOSNIA AND HERZEGOVINA)

***[1]Markotić, I., [2]Ćaleta, M and [3]Glamuzina, B.**

[1]Department of Biology, Faculty of Science and Education, University of Mostar, Mostar, Bosnia and Herzegovina
[2]Faculty of Teacher Education, University of Zagreb, Zagreb, Croatia
[3]Department of Aquaculture, University of Dubrovnik, Dubrovnik, Croatia

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ABSTRACT

Mostar minnow, *Phoxinellus pseudalepidotus*, is a freshwater fish endemic to the Neretva River basin. Present data suggest that the species is restricted only to wetland of the Mostarsko Blato but it is considered to be distributed more widely in the Neretva River basin. According to IUCN Red List, *P. pseudalepidotus* is listed as Endangered (EN) under criteria B (B1ab(iii)+2ab(iii)) ver. 3.1. The objective of the present study is to analyze gonadosomatic index of *P. pseudalepidotus* in the area of Mostarsko Blato (Neretva River basin, Bosnia and Herzegovina). The research was conducted at the area of Mostarsko Blato. The samples of the *P. pseudalepidotus* species were collected once a month using stationary fishing nets, 7 meters long, 0.7 meters high, and with a mesh diameter of 7 mm and using a "krtol" - a traditional fishing tool. Gonadosomatic index values were calculated for 360 individuals of the species *P. pseudalepidotus*. The mass of the individuals and the mass of the gonads were weighed with an accuracy of 0.001 g. Based on the mass of the gonads and the body mass, the values of the gonadosomatic index (GSI%) were calculated according to the formula: Gonadosomatic index (GSI%) = Weight of gonads (g) / Weight of fish (g) x 100. In females of the species *P. pseudalepidotus*, the gonadosomatic index (GSI) was lowest in May (3) and highest in January (21.9). The average annual value of the gonadosomatic index (GSI) for females was 10.73. In males, the gonadosomatic index was lowest in July (1.8) and August (1.8) and highest in February (15.6). The average annual value of the gonadosomatic index for males was 7.42. Based on the movement of average values of the gonadosomatic index by month, separately for females and for males, it was determined that the species *P. pseudalepidotus* spawns multiple times during the reproductive season, which lasts from the end of January to the end of May.

Keywords: *Phoxinellus pseudalepidotus*, Gonadosomatic index, Spawning season, Mostarsko Blato.

INTRODUCTION

Mostar minnow, *P. pseudalepidotus* (Bogutskaya & Zupančič, 2003), is a freshwater fish endemic to the Neretva River basin. Present data suggest that species is restricted only to wetland of the Mostarsko Blato but it is considered to be distributed more widely in the Neretva River basin (Bogutskaya & Zupančič, 2003). *P. pseudalepidotus* inhabits streams or shallow canals with little current and clean water (Crivelli, 2006; Bogutskaya & Zupančič, 2003). During unfavorable periods, it lives in subterranean waters (Markotić, 2013). According to IUCN Red List, *P. pseudalepidotus* is listed as Endangered (EN) under criteria B (B1ab(iii)+2ab(iii)) ver. 3.1 (Ford, 2024; IUCN). It is endangered because of the extremely limited range of distribution, river regulation, and influence of non-indigenous species (Mihinjač *et al*., 2014). Studies on reproductive biology of fish are crucial, needed, and a basic

*Corresponding Author: Ivana Markotić, Ph.D., Associate Professor, Department of Biology, Faculty of Science and Education, University of Mostar, Matice hrvatske bb, 88000 Mostar, Bosnia and Herzegovina, Email: ivana.markotic@fpmoz.sum.ba.

requirement to plan a better conservation and management strategies of fishery resources (Ali & Kadir, 1996; Ezenwaji, 1998; Brewer *et al*., 2008; Grandcourt *et al*., 2009; Muchlisin *et al*., 2010), for the examination of basic life-history information and for evaluating the impacts of environmental variability on the dynamics of fish populations (Schlosser, 1990). Knowledge about the reproductive biology of *P. pseudalepidotus* is scarce and insufficient. It has extended its period of spawning which lasts from January to May, when water temperature is between 9.6 and 23.9 ºC. During spawning season, females lay around 2500 eggs in more than one portion. The period of spawning coincides with flooding of the Mostarsko Blato (Markotić, 2013). The absolute fecundity ranged from 965 to 4740 oocytes, with the mean value and standard deviation of 2524.7±820.63. The diameter of the oocytes inside the gonads varies. The diameter of oocytes of *P. pseudalepidotus* was between 10.02 µm and 1367.10 µm. The average diameter of the oocyte of *P. pseudalepidotus* reaches its maximum in January with a value of 446.59±351.74 µm, while the minimum average size was measured in November with a value of 188.05±138.94 µm. The average diameter of oocytes of *P. pseudalepidotus* during the spawning period was 315.01±284.16 µm. The type of *P. pseudalepidotus* spawning was categorized as 'partial spawning' (Markotić, 2013; Markotić *et al*., 2025). Gonadosomatic index (GSI) is one of the important parameters of the fish biology, which gives a detailed idea regarding the fish reproduction, reproductive status of the fish species and help in ascertaining breeding period of fish (Sindhe & Kulkarni, 2004). Hence, the objective of the present study is to analyze gonadosomatic index of *P. pseudalepidotus* in the area of Mostarsko Blato (Neretva River basin, Bosnia and Herzegovina).

MATERIAL AND METHODS

Study area

Mostarsko Blato (43°19'55"N 17°41'53"E) is an enclosed karst field in the Neretva River basin, in the southwest of Bosnia and Herzegovina. The area of Mostarsko Blato characterizes sub-Mediterranean climate, i.e. temperately warm humid climate with hot summers (Cfa), according to Köppen's climatic classification. In general, winters are mild and rainy; while summers are hot and relatively dry, (there is no specific minimum like in Csa climatic zones, such as the neighboring littoral and insular part of Southern Croatia). Pluviometric regime is maritime, with most of precipitation concentrated in fall and winter (primary maximum), and in April and May (secondary maximum). The influence of the Adriatic Sea reaches from three sides; southeast, east and south respectively. The strongest inflow of air arrives through the Neretva River valley, and over low Varda ridge, which separates lower alluvial plain of Mostarsko Blato from higher Mostarsko Blato Field. This inflow relieves the influence of colder air coming from the mountains in the north. During fall, winter and spring, the most part of Polje is flooded, but during summer, almost all water springs dry out (Mostarsko Blato Hydropower Plant, Feasibility Study, EP HZHB, Salzburg, 2000).

Limestone hills of Orlovac, Mikuljača, Virača, Trtla, and Varda surround the area of Mostarsko Blato, through which the Lištica River flows. On average, the field is flooded for 5 to 6 months during the year. In addition to permanent water sources in Mostarsko Blato, significant amounts of periodic waters of rivers Ugrovača, Orovnik, Mokašnica, and a number of small torrents are active during major rainfall seasons. In order to reduce the floods, the tunnel Varda was built in 1947. Additionally, water from Mostarsko Blato is managed by the following sinkholes: Krenica, Košina, Renkovača, Kruševo, and the Velika Jama (Great Hole). These sinkholes are important for *P. pseudalepidotus* life cycle (Bogut *et al*., 2007).

Sampling

The samples of the *P. pseudalepidotus* species were collected once a month using stationary fishing nets 7 meters long, 0.7 meters high and with a mesh diameter of 7 mm and using a "krtol" - a traditional fishing tool. In the Mostarsko Blato area, during spring, summer and autumn, samples of the species *P. pseudalepidotus* were collected at three stations: Pisak (channel 1), Međurić (channel 2) and Pološki gaz (lower course of the Lištica river) (Figure 1). During the flooding period, fish were also caught in the open waters of the Mostarsko Blato in the village of Biograci (Figure 2) in order to collect a more representative sample.

Gonadosomatic index (GSI)

The gonadosomatic index values were calculated on 360 individuals of the species *P. pseudalepidotus* collected every month during 2009. The mass of the individuals and the mass of the gonads were weighed with an accuracy of 0.001 g. Based on the mass of the gonads and the mass of the body, the values of the gonadosomatic index (GSI%) were calculated according to the formula: Gonadosomatic index (GSI%) = Weight of gonads (g) / Weight of fish (g) x 100.

RESULTS AND DISCUSSION

Figure 3 shows the movements of the average values of the gonadosomatic index (GSI) by month, separately for females and for males, which shows that there are large oscillations of the values within a single month. In females of the species *P. pseudalepidotus*, the maximum value of the gonadosomatic index was recorded in January and it was 21.9. In February, the average value of the gonadosomatic index of females was 21.3, in March it was 18.2, while at the end of April it drops to 6.7 and continues to fall until the end of May. The lowest average value of the gonadosomatic index for females was recorded in May and was 3. In June, the average value of the gonadosomatic index for females was 3.04, in July it was 3.2, and in

August, it was 4.6. The average value of the gonadosomatic index for females in September was 7, in October it was 10.2, and it continued to increase until January, when it reaches its maximum. The average annual value of the gonadosomatic index (GSI) for females was 10.73. In November, December, January, February and March, the average value of the gonadosomatic index for females was higher than the average annual value of the gonadosomatic index recorded for females. The maximum value of the gonadosomatic index for males was 15.6 and it was recorded in February. In March, the average value of the gonadosomatic index was 13.6, in April it was 4.7 and it continued to fall until the end of May when it was 2.1. In June, the average value of the gonadosomatic index for males was also 2.1. The lowest average values of the gonadosomatic index for males of the species *P. pseudalepidotus* were recorded in July and August when it was 1.8. The average value of the gonadosomatic index for males in September was 3.9, in October it was 7, and in November, it was 11.3. In December, it was 11.7 and it continued to increase until February, when it reaches its maximum value. The average annual value of the gonadosomatic index for males was 7.42. In addition, in November, December, January, February and March the average value of the gonadosomatic index for males was higher than the average annual value of the gonadosomatic index recorded for males.

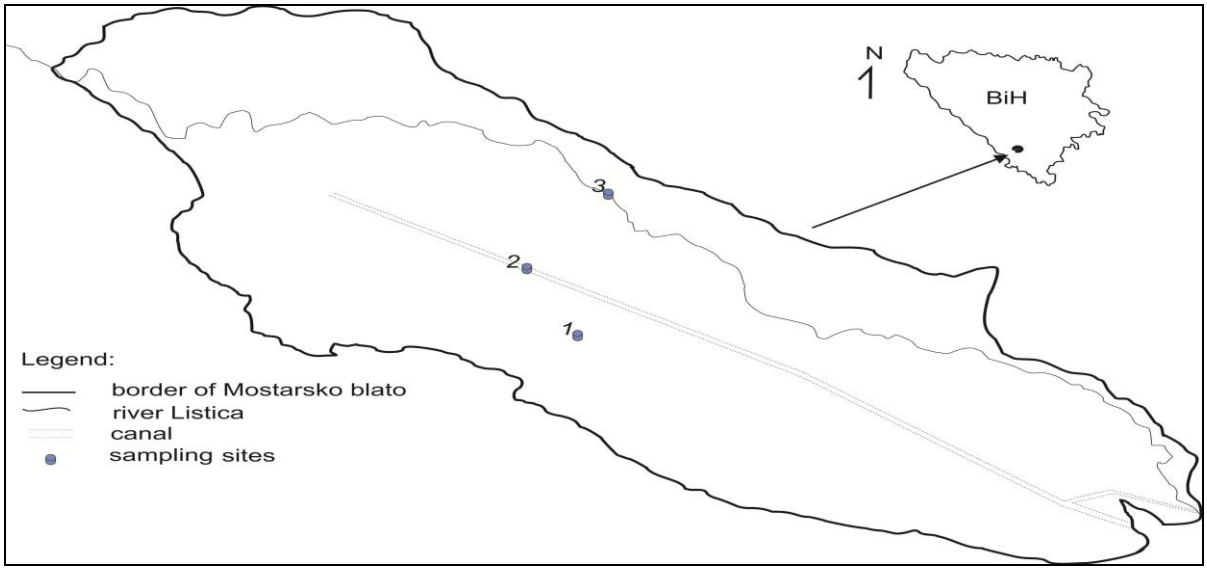

Figure 1. Map of sampling sites: Station 1 (Pisak), Station 2 (Međurić) and Station 3 (Pološki gaz).

Figure 2. Mostarsko Blato in February 2009.

A comparative overview of the movement of the average values of the gonadosomatic index (GSI) for females and males is shown in Figure 3. It was found that there was no statistically significant difference in the gonadosomatic index between the total sample of females and males (ANOVA, p = 0.215). Knowledge of the reproductive ecology of fish is important because the long-term persistence of a population is determined by its ability to successfully produce viable offspring in the environment in which that population lives (Wootton, 1990; Moyle & Cech, 2004). Research into the basic reproductive ecology of fish involves a variety of approaches, ranging from observation of spawning behavior to histological examination of reproductive tissues. However, as with other aspects of ecology, each approach is inherently limited in scope and scale (Levin, 1992). For example, some of the most commonly used approaches in reproductive ecology research are frequency length distributions and gonadosomatic index, which provide good information at the population level but provide little detail about individual fish. In contrast, studies based on laboratory observation and histology provide significant information about the reproductive activities of individual fish, but at the same time, expensive and time-consuming research limits sample size, which in turn limits the drawing of conclusions at the population level. For these reasons, most researchers use only one or two approaches, although several methods would be necessary to fully understand the overall reproductive ecology of a species or group of species (Durham & Wilde, 2008). The gonadosomatic index (GSI) values coincide with the gonadogenesis and spawning cycle. In females of the species *P. pseudalepidotus*, the maximum value of the gonadosomatic index was recorded in January and it was 21.9. In February, the average value of the gonadosomatic index was 21.3, in March it was 18.2, while at the end of April it dropped to 6.7 and continued to fall until the end of May. The average annual value of the gonadosomatic index for females was 10.73. The maximum value of the gonadosomatic index in males was 15.6 and it was recorded in February. The average annual value of the gonadosomatic index for males was 7.42. The average value of the gonadosomatic index in females was consistently higher than in males. The gonadosomatic index was correlated with the proportion of mature fish, i.e. the gonadosomatic index increased with the proportion of mature fish.

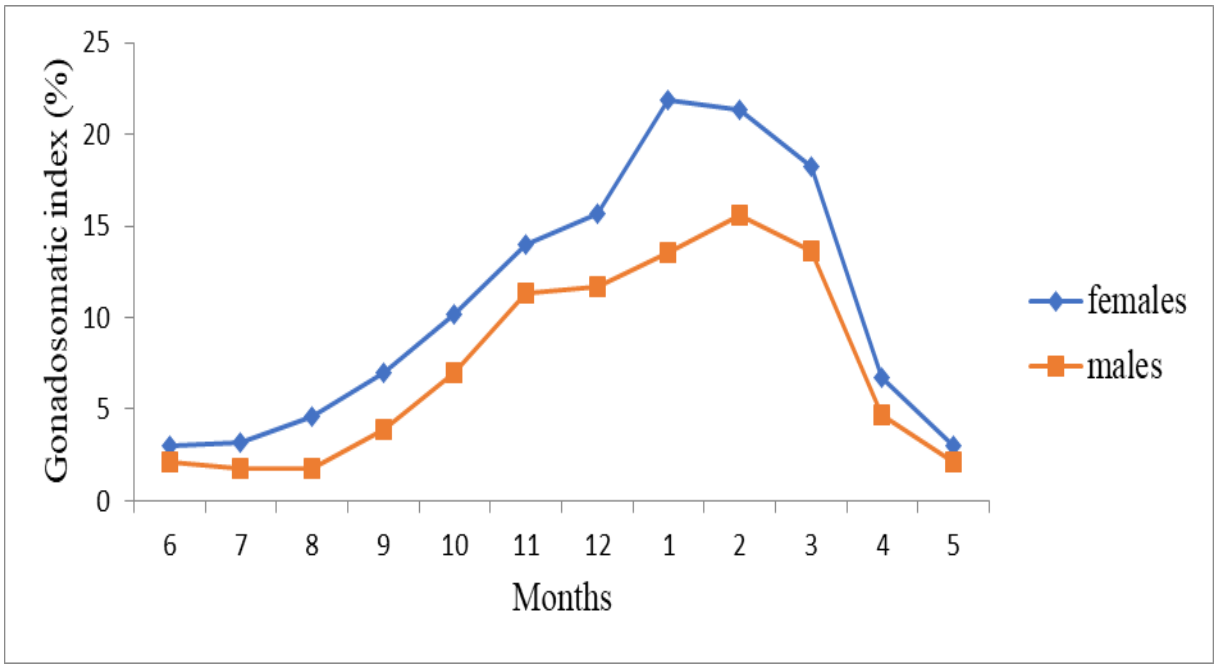

Figure 3. Gonadosomatic index (%) of the species *Phoxinellus pseudalepidotus* in the area of Mostarsko Blato during 2009.

There are no literature data on the GSI values for females and males of species of the genus *Phoxinellus*. In females of the *Telestes ukliva*, the value of the gonadosomatic index ranges from 2.8 to 23.2 for females at various stages of maturity. The highest gonadosomatic index was in April immediately before spawning, and the lowest in May after spawning. For this species, it has been established that the gonadosomatic index does not increase with the body length of the females, but depends only on the stage of sexual maturity (Zanella, 2003). In females of the species *Telestes souffia* from the Drina River, the value of the gonadosomatic index of females ranged from 6.33 to 22.56,

and in males from 2.25 to 6.75 (Vuković, 1985). The range of values of the gonadosomatic index of females of the species *P. pseudalepidotus* was from 3 to 21.9, which is almost the same result as that obtained for the species *Telestes ukliva* and a similar to the result for the species *Telestes souffia*. The gonadosomatic index (GSI) is one of the main parameters used to assess gonadal development in fish. The high correlation of the gonadosomatic index with the number of mature females and males can be used to determine the peak of the spawning season (Durham & Wilde, 2008). The gonadosomatic index also indicates the amount of energy invested in the production of offspring (Moyle & Cech, 2004).

CONCLUSION

In females of the species *P. pseudalepidotus*, the gonadosomatic index (GSI) was lowest in May (3) and highest in January (21.9). The average annual value of the gonadosomatic index (GSI) for females was 10.73. In males, the gonadosomatic index was lowest in July (1.8) and August (1.8) and highest in February (15.6). The average annual value of the gonadosomatic index for males was 7.42. Based on the trend of the average values of the gonadosomatic index by months, separately for females and males, it was determined that the species *P. pseudalepidotus* spawns multiple times during the reproductive season, which lasts from the end of January to the end of May.

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CONFLICT OF INTERESTS

The authors declare no conflict of interest

ETHICS APPROVAL

Not applicable

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AI TOOL DECLARATION

The authors declares that no AI and related tools are used to write the scientific content of this manuscript.

DATA AVAILABILITY

Data will be available on request

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