Aquatic Ecology
Research Article

ACTA ZOOLOGICA BULGARICA
Acta zool. bulg., 66 (4), 2014: 539-546

Some Life-History Traits of the Adriatic Brown Trout, *Salmo farioides* (Karaman, 1938) (Salmonidae) from the Morača River (Montenegro)

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Abstract: The length–weight relationship, condition factor, sex ratio and fecundity of the Adriatic brown trout, *Salmo farioides,* were studied in specimens from the upper part of the Morača River during the pre-spawning period. A total of 253 fish were examined; the ratio of matured males to females was estimated as 1:0.5. The length–weight relationship showed a positive allometric growth only in juveniles (age 0^+) and isometric growth pattern in the other groups. The absolute fecundity ranged from 137 to 1455 eggs, while the relative fecundity from 210 to 596 (mean 310.89) eggs per 100 grams of body weight. The minimum size at first sexual maturity was 13.7 cm total length in females, and 11.4 cm total length in males. Estimates of the average K ranged from 1.03 in juveniles 0^+, to 1.10 in mature males.

Key words: *Salmo farioides,* weight–length relationships, condition factor, fecundity, sex ratio, Montenegro

Introduction

The western part of Adriatic–Mediterranean area possesses a high degree of endemism of the salmonid species (Benhke 1973). This area is also distinguished by high phenotypic diversity among trout populations (Bernatchez 2001). This great phenotypic diversity of the local trout populations accounts for the recent scientific reports of more than 20 salmonid fish species (Kottelat, Freyhof 2007). Thus, numerous *Salmo* taxa have been described within this important region, and it is still debatable whether these taxa should be considered as phenotypic variants or distinct species (Apostolidis *et al*. 2011). In the older literature (Karaman 1933, Drecun 1962, Ivanović 1973, Šorić 1990, Marić 1995), three brown trout species were reported for the Morača River: *S. farioides*, *S. montenegrinus* and *S. dentex*. According to Marić, Milošević (2011), only one single species of brown trout (*Salmo farioides*) occurs in the Morača River system. Skadar Lake basin is inhabited by native Adriatic lineage of brown trout (Marić *et al*. 2006, Sušnik *et al*. 2007, Snoj *et al.* 2009, Mrdak *et al*. 2012). According to Kottelat, Freyhof (2007), the common name is Balkan brook trout and it is widespread in the streams of the eastern Adriatic slope, from Zrmanja (Croatia) to Mornos drainages (Greece). In addition to the brown trout, two other widespread species of salmonids, *S. zetensis* and *S. marmoratus*, are reported (Marić, Milošević 2011, Mrdak *et al*. 2012).

The Adriatic brown trout is the most frequent and most important fish for the anglers in the Morača River. However, the data about the biology of the species are scarce and our knowledge is mostly based on the old literature (Drecun 1952, 1984, Ivanović 1973). For example, Drecun (1984) reviewed the reproductive potential of the species populations from the Zeta River and middle part of the Morača River, particularly the size at first maturity and variations in fecundity related to body length and body weight.

The aim of this study was to evaluate the life-history traits, such as sex ratio, size at first maturity, length-weight relationship and fecundity of the native Adriatic brown trout population from the Morača River system (the upper part). The study of life-history styles of fish species is essential for analysing the population performance and thus, for the theoretical ecology and fisheries management (Winemiller, Rose 1992). Among the several applications of weight–length relationships (WLR) in fish biology, knowledge of these relationships is useful to the prediction of the weight from the length values as an indication of the fish condition or for fish stock assessments (Petrakis, Stergiou 1995, Froese, Pauly 2006, Vaslet *et al*. 2008). The importance of WLR has been extensively documented elsewhere (Froese 2006). The condition of fish is a widely used parameter that helps to evaluate the general well-being of fish, their growth, survival, maturity and reproduction (Le Cren 1951). Sex ratio studies are important and provide basic yet vital information for assessing the reproductive condition of the fish stock (Wang *et al*. 2003, Vicentini, Araújo 2003). Furthermore, studies related to fish sex determination are important to calculating the spawning stock biomass per size class (Chiang *et al*. 2006). Fecundity is important parameter for calculating the reproductive potential of native species population (including farming potential), or invasive potential in non-native (alien) species (Novomeská, Kováč 2009). Consideration of the life-history strategies of the Adriatic brown trout should be of great significance to fisheries management, restoration and monitoring.

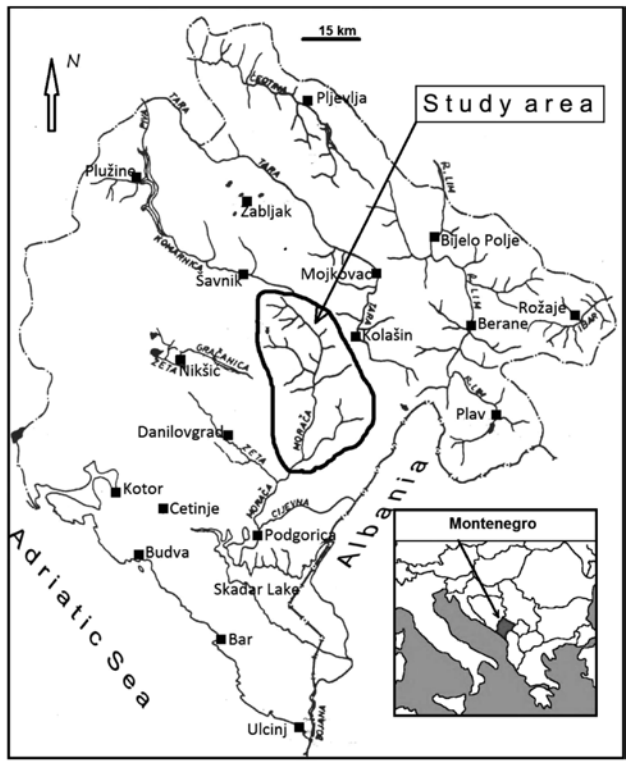

Fig. 1. Map of location of River Morača and study area

Material and Methods

The study was carried out at the Morača River. It originates in the northern part of Montenegro, under Rzača Mountain**.** Its source is at an altitude of 975 m, while its mouth is at altitude of 6 m. The Morača River is 99 km long and its catchment area has an area of 390 ha. The Morača is a relatively small river, rarely more than 100 m wide and mostly shallow. It is a typical karstic Mediterranean river, which flows through limestone and dolomite bedrock. The average slope of the whole Morača River is near 10 m per km of flow (near 10% slope). In its upper part (from the spring to Medjuriječje), the Morača is a fast mountain river and has cut a canyon north of the town of Podgorica. In this part, the river has an average slope of 26.6% and the average annual water temperature is about 8ºC (ranging 4-17ºC). Downstream of the confluence with its largest tributary (Zeta River) just north of Podgorica, the Morača River enters the Zeta plain. It flows through this flat area of Montenegro until it empties into Skadar Lake. The Morača River has several tributaries, of which the rivers Zeta, Mrtvica and Cijevna are the most important (Fig. 1). These three rivers are permanent, whereas other inflows often dry out during summer (Martinović–Vitanović, Kalafatić 1995).

The life-history traits of Adriatic brown trout from 4 sites located in the upper part of the Morača River were investigated during October 2010 (the end of the pre-spawning period). Specimens were caught by electro-shocking with 200–300 V, 40 Hertz frequencies and one anode. The river area where specimens were captured is about 150 m long and 5-15 m wide, with a depth of 30-100 cm.

The collected material was analysed on site or immediately frozen (84 specimens were returned to the river, 253 were sacrificed). Length and weight were measured using a digital balance and calliper. Each specimen was weighted (body weight) to the nearest milligram using an electronic balance and measured (total length) to the nearest millimetre using a measuring board. The age of the fish-juveniles ranged from 0^+ to 3^+ years, while the age of the matured specimens ranged from 1^+ to 5^+ years. For age determination the scales were removed from the left side of the fish, above the lateral line, near the dorsal fin and stored in ethanol (33%). The fish age was

Table 1. Some basic data for S. farioides from the Morača River during the pre-spawning periods (L = Total length, W = total weight)

	Juvenile (n=39) (age 0^+)	**Juvenile (n=65) (age 1^+)**	**Non mature ♀♂(n=112) (age 1^+,2^+,3^+)**	**Mature ♂ (n= 68)**	**Mature ♀ (n= 34)**
L (max)	9.9	14.7	17.7	36.0	36.1
L (min)	5.1	10.1	10.1	11.4	13.7
L (average)	7.0	12.7	14.34	18.22	20.37
W (max)	12.0	37.0	60.0	458.0	458.5
W (min)	2.4	11.2	11.2	15.0	25.4
W (average)	7.1	21.91	32.91	80.42	106.45

determined under a binocular microscope.

The parameters of the weight–length relationships (WLR) of identified fish species were estimated using the equation (Le Cren 1951, Froese 2006):

$W = aL^b$,
Where:
W = Weight (total) of fish (g)
L = Length (total) of fish (cm)
a = y-intercept or the initial growth coefficient
b = Slope or the growth coefficient.

Because the WLR for the same fish species is often different between adults and juveniles (Safran 1992), the WLR was calculated separately for juveniles and adults.

The condition factor (K) was calculated by Fulton's equation: $K (CF) = W^x 100 / L^3$, where: K - Condition factor; W – total weight of body; L – total lenght of body (in cm) and the factor 100 is used to bring K close to unity (Froese 2006). The coefficient of determination (R^2) was used as an indicator of the quality of the linear regression (Scherrer 1984). All the statistical analyses were considered at a significance level of 5% or 1% ($P<0.05$ or $P<0.01$). In order to test differences between groups, the t–test was conducted ($p=0.05$).

The sex was determined macroscopically by dissecting the organisms and looking directly at the gonads. The fish were dissected and gonads (testis or ovary) were removed, with their weight being measured to the nearest 0.01 g. In the analysis, the immature fish were those with gonads at stage I, while the mature fish consisted of the grouped individuals with gonads at stages III, IV and V. A subset of 34 mature-ripe females was considered for determining the relationship between fecundity, weight of the ovary, W and L. The fecundity (number of eggs) was calculated by separating each egg. Ten ova, selected at random from twenty ovaries, were measured by means of a micrometer eyepiece, and the mean egg diameter was then computed for the entire sample. The Gonadosomatic Index (GSI) was defined as gonad weight/total body weight percentage. Linear regression was used to examine the correlation between relative fecundity and TL, as well as the correlation between relative fecundity and total body weight. The relative fecundity (RF) was expressed as the number of eggs produced per unit of total body weight.

A total of 102 adult specimens were examined for determination of the sex ratio. The sex ratio (males : females) was calculated and the significant differences from the expected ratio (1:1) were tested by means of test - χ^2 (Sokal, Rohlf 1981).

Results

A total of 253 fish specimens from 4 localities were analysed during this study. The other fish species recorded during the electro-fishing survey of the upper part of the Morača River were: *Salmo marmoratus*, *Telestes montenegrinus*, *Barbus rebeli*, *Pachychilon pictum*, *Phoxinus lumaireul* and *Anguilla anguilla*. The Adriatic brown trout was the dominant species in the four localities (100%, 100%, 50% and 30%, respectively). The longest male and female fish captured were 36.0 mm and 36.1 mm, respectively. The maximum weight of males and females in the catches were 458 g and 458.5 g, respectively (Table 1).

The mature female specimens of *S. farioides* in this study were longer than the mature males. In males, we found the weight of two specimens to be less than 20 g (at about 12 cm TL), of two other specimens less than 25 g (at about 13 cm TL), of 4 specimens (5.8 %) less than 30 g, and of 14 (20%) specimens less than 40 g (at 15-16 cm). In females, two specimens (5.8%) weighted less than 30 g (13.0 cm), and 3 specimens (8.8%) less than 40 g (15.0 cm).

The specimens from the four localities were adjusted adequately to the length–weight exponential (power function) model. No significant differences were found between the immature specimens at age

1^+, 2^+, 3^+ ($P > 0.05$ in all cases); therefore, the data were pooled by using all individuals, and treating them as a single group (Fig. 2-5).

Estimates of the coefficient *b* ranged from 2.91 in the immature specimens (1^+-3^+), to 3.33 in the juveniles at age 0^+. The comparison of the weight–length relationships between the two-studied juvenile groups demonstrated that the juveniles at age 0^+ had greater exponent *b* of the length-weight relationship than the immature specimens at age 1^+, 2^+ and 3^+. There was no difference in the WLR between mature males and mature females. This shows that the juveniles at age 0^+ exhibited an allometric, while the other groups an isometric growth pattern (Fig. 2-5).

The coefficients of determination (R^2) of the WLR regressions ranged between 0.94 (juveniles 0^+) and 0.99 (mature males) and were all statistically significant ($P < 0.01$).

The estimates of the four groups of the Adriatic brown trout from the Morača River during the pre-spawning period, showed total variations of the condition factor (K) between 0.85 and 1.48. The estimates of the average K ranged from 1.03 in the juveniles 0^+ to 1.10 in the mature males (Table 2). These data were not representative for the other seasons.

The comparison of K between the four studied groups demonstrated that the mature specimens of *S. farioides* in the pre-spawning period had greater K (males = 1.10, females = 1.08) than the other groups. No significant differences were found between the mature males and females ($P > 0.05$) or between the juveniles at age 0^+ and the mature specimens ($P > 0.05$). Significant differences were found between the mature specimens (males and females) and between the immature specimens at age 1+ to 3^+ ($P < 0.01$).

The coefficients of correlation found between the weight or length and K of any group were not significant ($P > 0.05$ in all cases), and thus, the coefficients of correlation between K and GSI (3.5-8.8%) were also not significant in the mature females.

Out of the total 253 Adriatic brown trout specimens analysed during the study, 34 specimens (13.8%) were mature females, 68 specimens (26.88%) were mature males and 151 (59.68%) were sexually undetermined, immature specimens. The overall sex ratio during the investigation period was in favour of males (m/f = 2.0); it significantly deviates from the hypothetical distribution of 1:1 (χ^2= 11.34; *d.f.* = 1; $p<0.01$).

The analysis of the reproductive biology of the Adriatic brown trout population in the Morača River revealed that sexual maturity is reached after the second summer of life (1^+ age-class). All males >18 cm TL and >60 g were mature. At the same time, 20.0% of the specimens < 16 cm TL and < 40 g were mature as well. The size of the females that matured for the first time ranged from 13.8 to 18.0 cm TL. The males from the Morača River reached maturity at minimum 11.4 mm TL, *i.e.* 15.0 g. The portion of mature males smaller than 15 cm TL (age 1^+ and 2^+) was 22.06%, and only 2.9% of mature males <12 cm TL (<20 g) at age 1^+ were found. Our results showed that *S. farioides* matures for the first time at the age of 1^+ or 2^+ years.

The value of GSI in females was higher than that of males (Table 3). The absolute fecundity ranged from 137 to 1455 eggs, while the relative fecundity from 210 to 596 (mean 310.89) eggs/100 g of body weight. There was no significant correlation between the relative fecundity and body weight or the relative fecundity and total length in the females of *S. farioides* from the pre-spawning period. The same is also true for the correlation between GSI and TL or weight.

Discussion

The weight–length relationship, condition factor, sex ratio and fecundity of the Adriatic brown trout, *Salmo farioides,* were studied in specimens from the upper part of the Morača River during the pre-spawning period. The results presented contribute to the knowledge about sex ratio, size at first maturity, weight-length relationship and the condition factor of *S. farioides* from the Morača River. The Adriatic brown trout from two sites located in the upper part of the Morača River represent a single species. The upper course of the Morača River is characterised by

Table 2. Some values of the condition factor (K) for S. farioides from the Morača River during the pre-spawning periods (max = maximum K, min = minimum K, SD = standard deviation of the average K)

	Juvenile (n=39) (age 0^+)	**Juvenile (n=65) (age 1^+)**	**Non mature ♀♂ (n=112) (age 1^+, 2^+,3^+)**	**Mature ♂ (n=68)**	**Mature ♀ (n=34)**
Max	1.37	1.21	1.21	1.48	1.25
Min	0.88	0.92	0.85	0.95	0.96
Average	1.061	1.036	1.033	1.10	1.076
SD	0.131	0.069	0.072	0.078	0.074

extreme and unpredictable changes of the environmental conditions, which are typical for the karstic Mediterranean rivers, and represent a severe limiting factor for fish. The species inhabiting these streams possess several life-history and behavioural attributes to cope with seasonal flow variability (Pires *et al.* 1999). The results of the present study confirmed ability of the species to adjust some of its biological traits in response to the ecological conditions, for example to achieve sexual maturity earlier.

The mature female specimens of *S. farioides* in this study were longer than the mature males, which may be the result of the earlier sexual maturation of males. We determined that the smallest mature female had a length of 13.7 cm and weight of 25.4 g, while the smallest mature male had a length of 11.4 cm, *i.e.* 15.0 g. All males and females >18 cm TL were mature, this body length corresponding to the legal minimum capture size in Montenegro (25 cm). The largest specimen observed in this study was 36.1 cm long, but the maximum size of this fish, as reported by Drecun (1984), was 48.7 cm. In the past, according to Ivanović (1973), specimens heavier than 10 kg (known as *S. dentex*), caught in Skadar Lake during winter, would enter the large tributaries (the rivers Morača and Zeta) during summer. The disappearance of those large specimens is probably a result of overfishing.

The weight-length relations of fish are useful to estimation of biomass from the length observations, *e.g.*, in fisheries or conservation research. This study provides for the first time information on the length-weight relationships in *S. farioides* from the Morača River. Among the several applications of the weight–length relationships (WLR) in fish biology, the knowledge of these relationships is helpful in predicting the weight from the length values as an indication of fish condition or for a fish stock assessment (Petrakis, Stergiou 1995, Froese, Pauly 2006). Most fishery studies were conducted for commercial-sized and/or adult populations. The weight-length relationship in fish juveniles was also investigated. The estimates of the coefficient *b* ranged from 2.91 to 3.33. Significant differences were found between the juveniles at age 0^+ and the immature specimens at age (1^+-3^+). Similarly, significant differences were found between the juveniles at age 0^+ and the mature specimens. We concluded that no single regression could adequately describe the weight–length relationship in *S. farioides* (for all age classes) from the pre-spawning period. The range of values of *b* (Table 2), obtained in this study, is close to the values mentioned by Prpa *et al.* (2007). We think that our results are an adequate estimation of the weight–length relationship, since the parameter *b* falls within the expected range of 2.5–3.5 (Froese 2006). It is well known that the weight-length parameters depend on biological and environmental factors, as well as geographical and temporal factors, such as the age and condition of fish or the season of the year when samples are collected (Froese 2006, Ferreira *et al*. 2008, Vaslet *et al*. 2008). According to Tesch (1971) and Wootton (1998), the weight-length relationships in fish can be affected by several factors, including habitat, gonad maturity, sex, health, and stomach fullness. Turkmen *et al.* (2001) argued that the exponent *b* in fish differ depending on the species, sex, age, season, and fish feeding.

Our data showed that the gonad maturity probably affected the WLR. During their development, fish pass through stages in their life history, which are defined by different weight-length relationships. Kharat *et al*. (2008) clearly stated that there is a discrete pattern in changes in the exponent *b*, correlated with the reproductive phase of fish.

In fishery science, the condition factor is used to compare the "condition", "fatness" or well-being of fish. This is based on the hypothesis that heavier fish of a particular length are in a better physiological condition (Bagenal 1978). The estimates of the average K ranged from 1.03 to 1.10. The average of value of K obtained in this study is similar to the values reported by Prpa *et al.* (2007) for the Adriatic brown trout in Croatian rivers. The presented results are the first contribution to the knowledge about the condition factor of *S. farioides* from Montenegro. On using the results presented in this study, it should be noted that the samples were taken in autumn and the number of fish examined was limited. Significant differences were found only between the mature specimens and between the immature specimens at age 1+ to 3^+. In the juveniles/ immature specimens, the coefficient of condition indicated that the small fish specimens were in better condition than the bigger ones. The coefficients of correlation found between the weight or length, GSI (in mature females) and K were not significant ($P > 0.05$ in all cases). This means that in the matured specimens during the spawning period, the length and weight of individu-

Table 3. GSI and relative number of eggs in S. farioides during the pre-spawning period

	Males (n=68)			**Females (n=34)**		
	average	max	min	average	max	min
GSI	3.19	6.00	1.20	5.92	8.80	3.50
eggs/100 g				310.89	593	210
Diameter of eggs				3.1	3.4	2.7

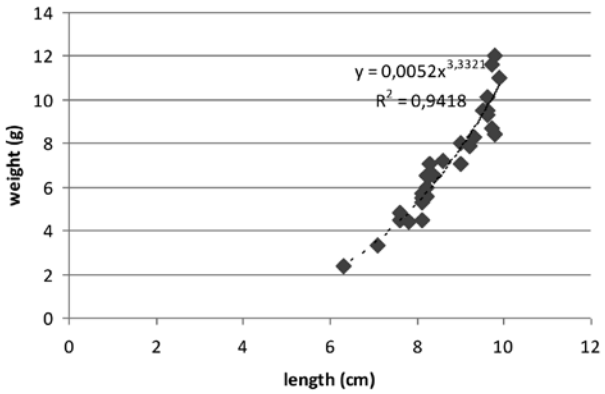

Fig. 2. Weight–length relationship in the juveniles of S. farioides at age 0+ (R2 - coefficient of determination, b – exponent, a - parameter)

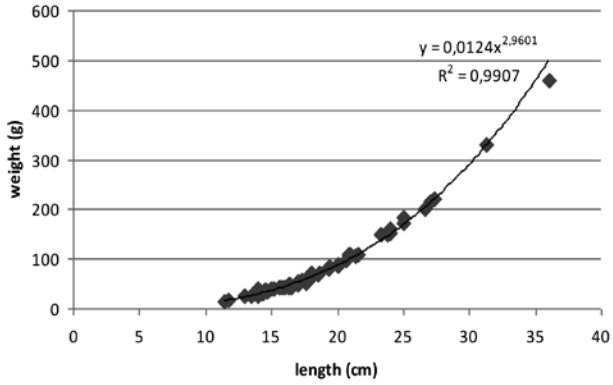

Fig. 4. Weight–length relationship in the mature males of S. farioides (R2 - coefficient of determination, b – exponent, a - parameter)

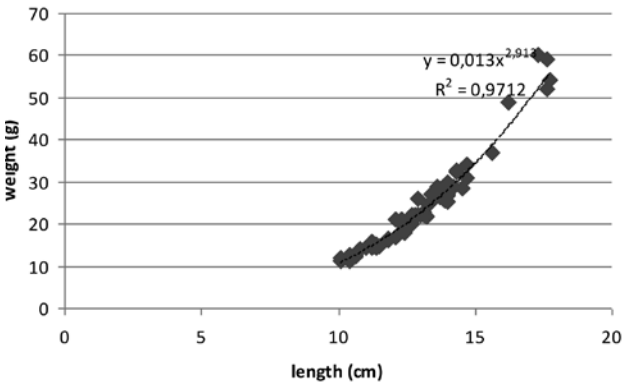

Fig. 3. Weight–length relationship in the juveniles of S. farioides at age 1+ to 3+ (R2 - coefficient of determination, b – exponent, a - parameter)

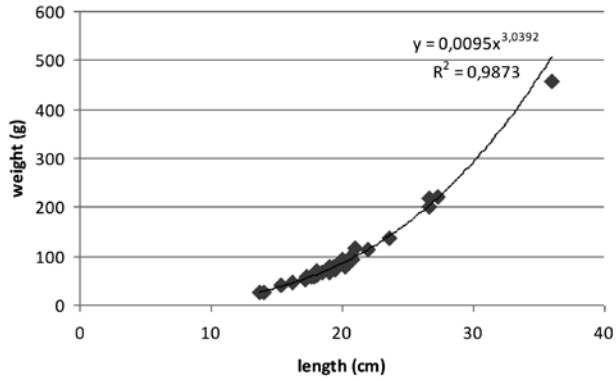

Fig. 5. Weight–length relationship in the mature females of S. farioides (R2 - coefficient of determination, b – exponent, a - parameter)

als, as well as the level of sexual maturity, did not affect the K value.

The overall sex ratio is close to 1:1 in many species, but may vary from species to species and may change from year to year in the same population (Nikolsky 1963). The sex ratio found in the trout population from the Morača River was close to 1:2, which suggests a significantly unbalanced population. These parameters are not similar to those reported by Drecun (1984). According to Drecun (1984), the sex ratio is close to 1:1. In our study, there was preponderance of males over females, which could be a result of the earlier sexual maturation in males. The uneven sex ratio could also be a consequence of the many other factors, such as the period of the study, sampling methods, and environmental conditions. (Nikolsky 1963)

The estimates of GSI and adult specimens with ripe ovaries indicated that the spawning season of *S. farioides* in the Morača River starts in October. According to Drecun *et al.* (1985), the Adriatic brown trout in the Morača River spawn from October through January. Our results showed that *S. farioides* matures for the first time at the age of 1+ or 2^+ years. This may be a successful life-history strategy for *S. farioides* in unstable environments.

Spawning begins when the water temperature is above 10°C and therefore *S. farioides* is considered to be a late autumn spawner, although the main spawning season may be extended from November to February, depending on the local water conditions. In this period (the end of the pre-spawning period), the ovaries in the developing stage contained yolked oocytes, which reached a maximum diameter of 3.5-4.0 mm. The long incubation period typical of salmonids is associated with unusually large eggs (Hutchings, Morris 1985). Spawning usually takes place on the shallow sand or gravel bottoms. The fecundity ranged from 137 to 1455 eggs per ripe ovary, while Drecun (1984) reported from 822 to 3186 eggs per female in specimens of 30-46 cm TL. Fecundity of salmonids is comparatively low for their body size; most teleosts have much smaller, but more numerous eggs (Elgar

1990, Chambers, Leggett 1996). Our data showed that there was no significant correlation found between the relative fecundity and body weight and the relative fecundity and total length in females from the pre-spawning period. The fecundity is important parameter for calculating the reproductive potential for farming.

Our study represents an additional contribution to understanding better biology of *S. farioides*. Moreover, our research provides the first references on GSI, LWR and K in *S. farioides* from Montenegro. The results here suggest that conservation strategies should be directed towards preserving the biology, integrity and uniqueness of each population.

Acknowledgements: We are grateful to the anonymous reviewers for their constructive comments on the manuscript.

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Received: 08.05.2014
Accepted: 06.07.2014