

Life history traits, growth and feeding ecology of a native species (*Barbus strumicae* Karaman, 1955) in Nestos River, a flow regulated river in northern Greece

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Abstract. *Barbus strumicae* Karaman 1955 is an endemic species from the Balkan Peninsula. It was distinguished from other species like *B. cyclolepis* recently. Unfortunately, little information exists on the biology of *B. strumicae*, especially for a population found in a flow regulated river (Nestos River, North-East Greece). Males were dominating in both locations. The length-at-first maturity for the males was 95.23 ± 4.35 mm and 112.54 ± 4.97 mm for the females. The Fulton's condition factor exhibited monthly variation with two peaks in April (1.517) and November (0.903). The GSI revealed a two months spawning period, from April to June. The Absolute fecundity (F) ranged between 1,981 and 14,280 oocytes (mean=9,468.09) and relative fecundity between 35.85 and 304.33 oocytes/g of net weight (mean=85.03).

Key words: age-growth, *Barbus* spp., diet, growth parameters, fecundity, maturity, spawning, northern Greece

Introduction

During the last decades, a growing demand for exploitation of both surface and underground water resources for hydroelectricity, recreation, agriculture, and municipal water supplies has been observed (Vogl & Lopes 2009). In freshwater ecosystems, human interference in terms of water flow regulation, river impoundment, construction of bridges, weirs and dams have contributed greatly to the development of human society, however with significant environmental impacts (Dynesius & Nilsson 1994, Daniels et al. 2005). Such habitat deterioration and the loss of suitable spawning and feeding areas are responsible for the decreasing of the fish fauna diversity (Schulz & Schoonbee 1999). According to Mims et al. (2010) life history strategies of a species could be favoured or not, when it is subjected to specific environmental conditions. There are cases where human intervention has resulted in the conversion of the once rich ecosystems into oligotrophic, which thus affects the planktonic and fish populations due to the decline of the food resources (Kawara et al. 1998, Zheng et al. 2006).

The Strumica Barbel (*Barbus strumicae* Karaman, 1955) was distinguished from other *Barbus* species, such as *B. cyclolepis*, *B. plebejus* and *B. balcanicus*, by a combination of various characters enumerated in Kottelat & Freyhof (2007). Its distribution area, located in the Aegean Sea basin, ex-

tends from Lake Volvi to River Nestos drainages (Greece, Bulgaria, FYROM) and Halkidiki peninsula catchment area (Greece) (Kottelat & Freyhof 2007). Koutrakis et al. (2013) considered *B. strumicae* as a native species of the broader area of the south-eastern Balkan Peninsula. It is a benthic fish that lives in rivers and creeks with rocky or sandy habitats (Apostolou & Koutrakis 2009). Recent validations characterized the species as Least Concern (LC) (IUCN 2013). Furthermore, the species is listed in the Annex II (*Animal and plant species of community interest whose conservation requires the designation of special areas of conservation*) of the Directive 92/43/EEC, under the name *Barbus* spp. The Directive considers that species populations are threatened by habitat degradation due to human activities. Moreover, this consideration seems to be valid for some isolated populations. Regarding the biology of *B. strumicae*, little information is available, concerning the age structure and reproduction. The available information concerns *B. cyclolepis* in Doirani stream (Vasiliou & Economidis 2005) and in the Dospat Dam and the Dzerman River (Bulgaria) (Dikov & Zhivkov 1985) and for the species *B. meridionalis* in Rentina stream (Neophitou 1987). Nevertheless, the non-studied population of *B. strumicae* from Nestos River is subjected to major environmental variations due to the occurrence of two hydroelectric power plants by the Public Electricity Company (PEC), which might have affected some important

aspects of the species' biology.

The aim of this study is to describe some biological aspects, such as age structure, maturity, growth and feeding strategy of the *B. strumicae* population from a flow regulated river in the Balkan, Nestos River, which were divided into two sections after the construction of the hydroelectric power plants by the PEC. This information presented in the current study are important for establishing management measures and policies for the conservation of this important fish species.

Methods

Study Area and Sampling

Nestos River is a transboundary river originating from the Rila Mountains, between the Aimos and Rodopi mountains located in southern Bulgaria, flowing after a total distance of 234 km (104 km in Bulgaria and 130 km in Greece) into the North Aegean Sea (Fig. 1).

Three dams are located in the Greek part of the Nestos River catchment. Both major dams have been constructed by Public Electricity Corporation (PEC) as Hydro-Electric Power Plants (HEPP). The first one is the multipurpose dam of Thisavros (operating since 1997), the farthest upstream, and the second is the Platanovrysi dam (operating since 1999), forming two artificial lakes, the Thisavros and Platanovrysi, respectively (Sylaios & Bourmanski 2009) (Fig. 1). Finally, a third smaller irrigation dam which was constructed in 1966 is located 30 km before the mouth of the river, in the area of Toxotes village. The presence of these dams has divided the river into four sections, forming four ecosystems - two lacustrine and two riverine - with different physico-chemical and hydrological conditions (Psilovikos et al. 2006, Kamidis et al. 2010). From north to south there is a lacustrine system that extends from the Greek - Bulgarian borders downstream to the Thisavros dam and a second lacustrine system downstream from the Thisavros dam to the Platanovrysi dam. The riverine systems extend from the Platanovrysi dam downstream to the Toxotes dam and the second riverine part downstream the Toxotes dam to the river mouth.

Fish samples were collected monthly during the period March 2007 - November 2007, from five stations in the main river course (M) and six stations in tributaries (T) of the Nestos River (Fig. 1). The bad weather conditions prevailed during the winter months was the reason why no fish samples were collected during winter. Samplings were conducted using a backpack portable electrofishing device (Hans Grassl Direct Pulse Current Electrofishing Device IG200/2), working at 80 Hz. Electric fishing surveys were carried out according to CEN standards on electric fishing (CEN 2003).

Samples elaboration and statistical analysis

All the specimens were anesthetized prior to sacrifice using 10% eugenol (clove oil). The specimens were pre-

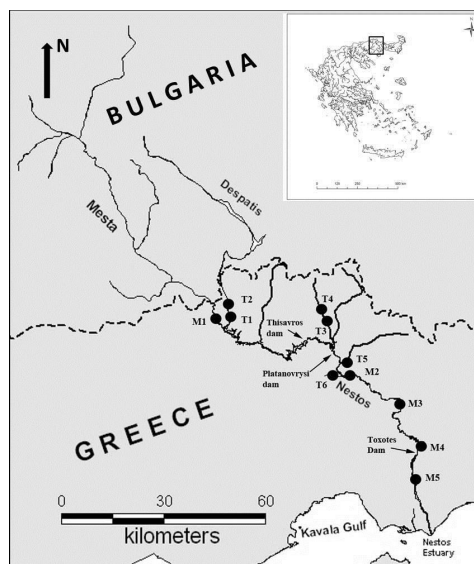


Figure 1. Map of Nestos River with sampling stations in the main course (M) and tributaries (T) of the river catchment.

served later in 7% formal solution. For all 200 specimens of *B. strumicae* captured, total length (TL) was measured and weighted to the nearest 0.1 mm and 0.01 g, respectively. Gonads (W_{gonad}) and liver weights (W_{liver}) were weighted to the nearest 0.001 g. The sex determination of each specimen was obtained by optical observation of the gonads. Scales were removed from each specimen from the left side of the body above the lateral line in order to determine the age using the scalimetric method (Bagenal & Tesch 1978). The reading of the scale was performed by two different operators at x20 magnification. The scales were mounted dry between two slides for stereomicroscopic study and the identification of the annuli was performed according to Ricker (1987). Each operator carried out three independent readings, which were used to determine the Average Percent Error (APE) according to the equation provided by Beamish & Fournier (1981):

$$APE = 1/N * \sum [1/R * \sum (|X_{ij} - X_j| / X_j)]$$

where N is the total number of samples, R is the number of times each scale was read, X_{ij} is the i^{th} age determination of the j^{th} fish and X_j is the average age calculated for the j^{th} fish.

The growth of fish was determined by back-calculation from scales. Total scale radius (R_{tot}), from the nucleus of the scale to the edge of the scale, as well as the radius of the age rings (R_i) from the centre of scale were measured along the major axis (Bagenal & Tesch 1978), to the nearest 0.01 mm, using a NIKON Digital Sight DS - L2 image analysis system with a NIKON DS-Fi1 camera fitted on a Nikon SMZ-1500 stereoscope.

The relationship between body length and scale radius was exponential so lengths at the time of annulus formation were back-calculated using Monastyrsky's

equation (Francis 1990):

$$TL_i / TL = (R_i / R_{tot})^b$$

where TL_i is the total length at the time of the formation of the i th annulus, TL the total length of the fish at the time of capture and b the constant derived from the exponential TL - R_{tot} relationship.

The growth rate of the species was described and calculated by von Bertalanffy model:

$$TL_t = TL_{\infty} * (1 - e^{-K*(t-t_0)})$$

where TL_t is the total length (in mm) of a fish at age t and TL_{∞} is the asymptotic length. The parameter K (in years⁻¹) is the growth coefficient and t_0 (in years) is the predicted age at zero length.

Using the values for K and TL_{∞} the growth index φ' was calculated (Pauly & Munro 1984), using the formula:

$$\varphi' = \log_{10}K + 2\log_{10} TL_{\infty}$$

Sex ratio was determined and Chi square (χ^2) test was used to identify statistical differences from the theoretical ratio 1 : 1 of the sex ratio (Zar 1984, Vasiliou & Economidis 2005). The absolute (F) and the relative fecundity (RF) were determined by using gonads of 23 females at stage IV and V of gonad development (Nikolsky 1963). All gonads were placed in Gilson fluid to facilitate egg separation (Bagenal & Braum 1978). The oocyte diameter was measured using the NIKON Digital Sight DS - L2 image analysis system. The gonad development was examined by calculating the Gonadosomatic Index (GSI), according to the formula:

$$GSI = (W_{gonad} / W_{total}) * 100$$

where W_{gonad} is the total weight of the gonads.

Fulton's condition factor was calculated according to the formula (Bagenal & Braun 1978):

$$K = (W_{total} / TL^3) * 100$$

Length-maturity relationship was determined according to the length-at-first maturity (TL_{50}), i.e. the length at which 50% of the individuals attain sexual maturity. The length-maturity relationship was estimated by fitting a logistic curve to the relation between the percentages of mature fish (P) per total length class (TL):

$$P = e^{(a+bTL)} / (1 + e^{(a+bTL)}) \text{ (Echeverria 1987)}$$

where P is the percentage of mature individuals, a and b constants, which are used to calculate the L_{50} according to the formula:

$$TL_{50} = -a/b$$

All specimens assigned to the III or higher stage of maturity, according to Kesteven (1960), were considered mature.

Diet and feeding strategy of the species were determined by analysing the stomach content using the Frequency of Occurrence (FO) and the percentage composition of each food category (N) (Hyslop 1980). Individual guts were removed, carefully unfolded and they were cut open in a petri dish using a pair of ocular scissors. The entire gut contents were examined under stereoscope and each dietary item was identified to the lowest taxonomic level as possible, while those that were difficult to be identified were categorized as unidentified items (Hyslop 1980). The frequency of occurrence is defined as the ratio of the number (n_i) of stomachs in which the prey i is located to the total number of non-empty stomachs (S_i).

$$FO = 100 * n_i / S_i$$

The percentage composition is defined as the total number of individuals of each prey species (Σn_i) expressed as a percentage of total food items identified (ΣN), and calculated according to the formula:

$$N = 100 * \Sigma n_i / \Sigma N$$

The vacuity index was also estimated as the number of empty stomachs (S_e) expressed as a percentage to the total number of non-empty stomachs (S_i):

$$VI\% = S_e / S_i * 100$$

Feeding strategy of the species was determined by using the Costello graphical method (Costello 1990). Analysis of covariance (ANCOVA) compared the volume of gut content among sampling sites and seasons.

All statistical analyses used in the present study, i.e. ANOVA, ANCOVA and ANOSIM were performed using the StatPoint STATGRAPHICS Centurion XVI.

Results

Sex ratio

Sex ratio was determined for each group and for all 200 *B. strumicae* specimens. From both populations, upstream and downstream, the ratio were 1.66 males : 1 female ($\chi^2=6.20$, $p<0.05$), and 2.33 : 1 ($\chi^2=13.61$, $p<0.001$), respectively. For the total sample the ratio was 1.93 : 1 ($\chi^2=19.32$, $p<0.001$).

Age & Growth

Of the 200 total scale preparations, 15 were rejected according to the disagreement of the determination of age between both readings. Ultimately, the age of 185 individuals (61 females and 124 males) was determined. The APE for the first reader was of 2.2% and 1.9% for the second reader, which indicates that second reader was more precise than the first one, but the APE was very low for both the readers.

The application of the ANOVA test indicated the absence of any significant difference between sexes. Moreover, the use of the Breakdown and one-way ANOVA also indicated the absence of any significant difference in the age structure between the sampling sites ($p=0.073$). Therefore, all the samples were pooled into one group, independently of the sex or the area that were captured.

The results of the age determination are shown in Table 1. The maximum age determined was 4+ and was attained by five female and six males. About 38% of the samples were classified in age class 1+ and 42% in class 2+. All values of K , L_{∞} and t_0 of the von Bertalanffy growth formula and the growth index φ' are presented in Table 2.

Table 1. Mean back-calculated total lengths (TL, mm) and 95% confidence intervals (mm) at age, annual increment and growth rate of males and females of the *B. strumicae* population in Nestos River. n - number of fish.

| Age (years) | n | Mean TL (mm) | Mean back-calculated TL at age | | | |
|--------------------|-----|--------------|--------------------------------|-------------|--------------|--------|
| | | | 1 | 2 | 3 | 4 |
| Female | | | | | | |
| 1+ | 26 | 70.76 | 50.75 | | | |
| 2+ | 22 | 88.01 | 54.85 | 76.76 | | |
| 3+ | 8 | 106.70 | 58.01 | 80.24 | 98.43 | |
| 4+ | 5 | 125.75 | 59.19 | 82.49 | 103.37 | 117.10 |
| Total individuals | 61 | | 61 | 36 | 14 | 5 |
| Mean | | | 55.70 | 79.83 | 100.90 | 117.10 |
| C.I. 95% | | | 50.75-59.19 | 76.76-82.49 | 98.43-103.37 | |
| Annual Increment | | | | 24.13 | 21.07 | 16.20 |
| Male | | | | | | |
| 1+ | 45 | 68.86 | 49.91 | | | |
| 2+ | 58 | 88.94 | 55.28 | 77.14 | | |
| 3+ | 15 | 107.07 | 57.30 | 79.43 | 98.24 | |
| 4+ | 6 | 125.88 | 59.17 | 82.21 | 103.78 | 114.77 |
| Total individuals | 124 | | 124 | 79 | 21 | 6 |
| Mean | | | 55.41 | 79.60 | 101.01 | 114.77 |
| C.I. 95% | | | 49.91-59.17 | 77.14-82.21 | 98.24-103.78 | |
| Annual Increment | | | | 24.18 | 21.41 | 13.76 |
| Male+Female | | | | | | |
| 1+ | 71 | 69.56 | 50.21 | | | |
| 2+ | 80 | 88.68 | 55.16 | 77.04 | | |
| 3+ | 23 | 106.94 | 57.54 | 79.72 | 98.31 | |
| 4+ | 11 | 125.82 | 59.18 | 82.34 | 103.59 | 115.83 |
| Total individuals | 185 | | 185 | 114 | 34 | 11 |
| Mean | | | 55.52 | 79.70 | 100.95 | 115.83 |
| C.I. 95% | | | 50.21-59.18 | 77.04-82.34 | 98.31-103.59 | |
| Annual Increment | | | | 24.17 | 21.25 | 14.88 |

Table 2. Parameters L_{∞} (TL, mm), K (year⁻¹), t₀ (year) of the von Bertalanffy growth equation and growth index $\varphi' = \text{Log}_{10}K + 2\text{Log}_{10}L_{\infty}$ for *B. strumicae* from Nestos River.

| Von Bertalanffy growth parameters | Estimate | Standard Error | Confidence Interval 95% | |
|-----------------------------------|----------|----------------|-------------------------|--------|
| | | | Lower | Upper |
| L_{∞} | 215.48 | 47.71 | 121.64 | 309.32 |
| K | 0.165 | 0.058 | 0.052 | 0.278 |
| t ₀ | -0.742 | 0.176 | -1.088 | -0.396 |
| φ' | 3.884 | | | |

Maturity

Proportion of mature male and female individuals of the species by size classes are presented in Fig. 2. The first mature males of *B. strumicae* appeared in the length class of 70 mm (Fig. 2a). The percentage of mature specimen increased steadily with body length, reaching the 50% in the length class of 100 mm. According to the logistic model all males with length greater than 120 mm were mature. The TL_{50} of males was 95.23 mm (SE=4.35) ($a=-7.731$, $b=0.0812$, $r^2=73.85$, $n=14$), which indicates a 2+ years old male fish.

The first female mature appeared in the length class of 80 mm and all female individuals were mature at the length class of 130 mm (Fig. 2b). The

TL_{50} for the female specimen was equal to 112.54 mm (SE=4.97) ($a=-11.17$, $b=0.099$, $r^2=0.91$, $n=16$), and corresponded to the age of 3+.

Reproduction

The gonadosomatic index (GSI) indicates that the spawning period last for almost 2 months, starting on April (GSI=2.230), with a peak during May (GSI=7.204), while in June there is a slight decrease (GSI=6.451) (see Fig. 3). On July the index returned to a level slightly lower than that of April. The same pattern is also observed when observing the GSI of males and females separately, which presents a maximum value in May with the female specimens obtaining higher values than

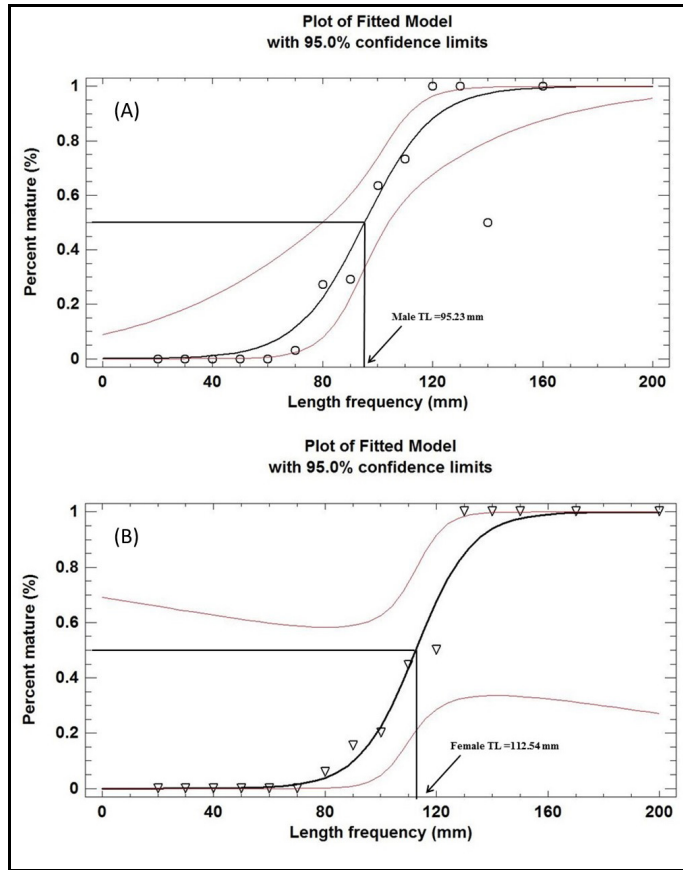


Figure 2. Proportion (%) of mature males (a) and females (b) of *B. strumicae* by size class (TL, mm) caught during the sampling period.

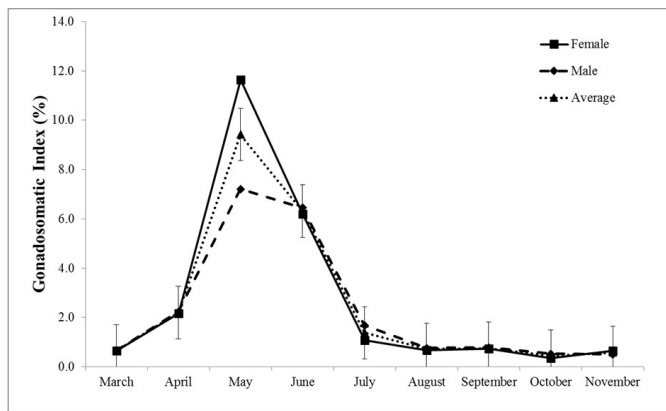


Figure 3. Monthly variation of Gonadosomatic index of *B. strumicae*.

males. The ANOVA test indicated the absence of any significant difference of the GSI values between the two sexes during the nine months of the study ($F=2.005, p=0.16$).

The absolute fecundity ranged between 1,981 and 14,280 oocytes, corresponding to specimens

with TL of 165 mm and 289 mm, respectively (Fig. 4). Relative fecundity was ranging between 35.85 eggs/g and 304.33 eggs/g, for specimens with TL of 293 mm and 211 mm, respectively.

A seasonal variation in oocyte size was observed. Indeed, during the spring season (Fig. 5a),

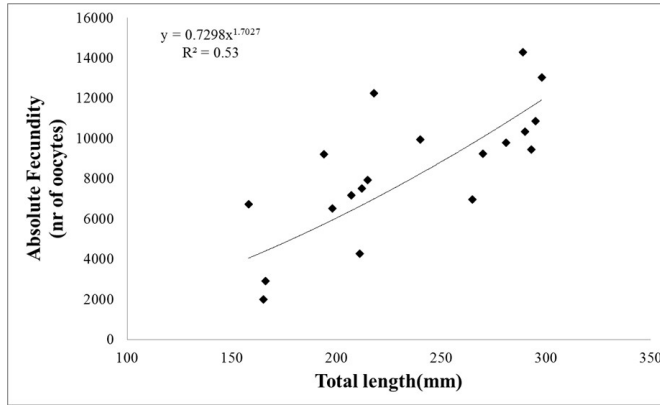


Figure 4. Relationship of absolute fecundity (F) as a function of total length (TL, mm) for the species *B. strumicae*.

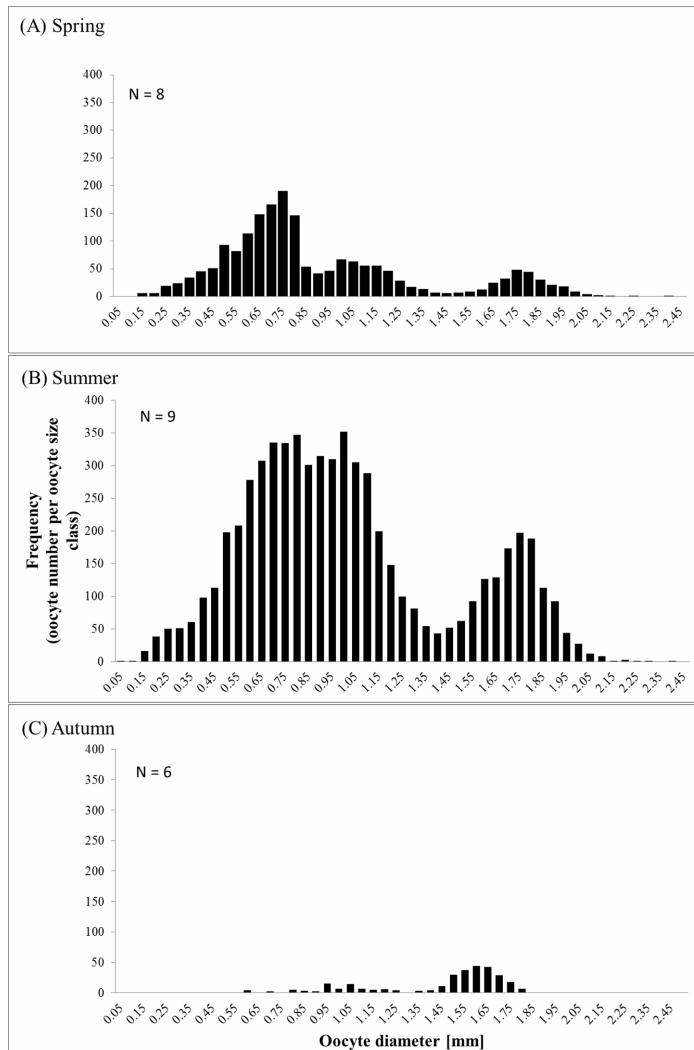


Figure 5. Seasonal variation of the size frequency profile of oocytes of *B. strumicae*. N=number of females sampled per season.

the majority of the oocytes with almost 60% was smaller than 0.8 mm, with mean diameter of 0.6 mm (SE=0.0044). Two more groups were observed, one group with mean diameter 1.05 mm (SE=0.007) and another with mean diameter 1.76 mm (SE=0.008). During summer there were only two groups of eggs (Fig. 5b) with significant increase of the eggs with diameter less than 1.45 mm (mean diameter 0.81 mm - SE=0.004 mm), while the second group had a mean diameter of 1.72 mm (SE=0.004). Finally, during autumn (Fig. 5c) there were also two groups, the first group with mean diameter of 0.97 mm (SE=0.019) while the second group presented a mean diameter of 1.59 mm (SE=0.008). However, the total number of oocytes with diameter over 1.05 mm was very small (267 oocytes) in contrast to the total number of oocytes of the same mean diameter during spring and summer.

Condition Factor

Fluctuation of the condition factor Fulton (K) can be observed for both female and male samples over the time. Despite two peaks observed, April (K=1.517) and September (K=1.637), ANOVA test did not indicate any significant differentiation among the months (F=2.235, p=0.13). For the males specimen, K values ranged from 0.903 (November) to 1.434 (April). On the other hand for females, it ranges between 0.903 (November) and 1.922 (September).

Diet & Vacuity Index

Twelve different food items were identified and classified into six major categories: plant detritus, insects (e.g. larvae of chironomidae, amphipoda, ephemeroptera, trichoptera), and arachnids. The ANOSIM analysis revealed the similarity of the diet composition between the sexes (Global R=0, p=0.5) and between all sampling sites (Global R=0, p=0.6).

The dominant food items were the chironomid larvae (60.40%), followed by the ephemeroptera (54.46%), trichoptera (38.61%) and plant detritus (31.68%) (Table 3). To be noted that over 50% of the stomachs presented sediment together with dominant food items.

The chironomid larvae were the main food source for *B. strumicae* during spring, summer and autumn (Table 4). During spring, the diet was completed by ephemeroptera (9.79%) and the amphipods (8.88%) (Table 4). During summer, trichoptera (12.70%) and ephemeroptera (12.74%)

Table 3. Overall diet composition of *B. strumicae* presented as frequency of occurrence (%FO) and number (%N).

| Food items | % FO | % N |
|--------------------------|-------|-------|
| Insects | | |
| 1 Chironomidae (larvae) | 60.40 | 39.98 |
| 2 Ephemeroptera (larvae) | 54.46 | 10.01 |
| 3 Trichoptera | 38.61 | 13.36 |
| 4 Amphipoda | 16.83 | 4.69 |
| 5 Trichoptera (larvae) | 1.98 | 0.42 |
| 6 Coleoptera | 0.99 | 0.03 |
| Diptera larvae | | |
| 7 Similidae | 10.89 | 0.84 |
| 8 Crane fly | 1.98 | 0.38 |
| 9 Sediment | 52.47 | |
| 10 Plant detritus | 31.68 | |
| 11 Unidentified | 10.89 | 5.37 |
| 12 Flying insects | 6.93 | 2.82 |
| 13 Ants | 1.98 | 0.09 |
| 14 Arachnida | 0.99 | 1.11 |

Table 4. Seasonal variation in percentage of *B. strumicae* dietary composition.

| Food items | Season | | |
|--------------------------|--------|--------|--------|
| | Spring | Summer | Autumn |
| Insects | | | |
| 1 Chironomidae (larvae) | 35.35 | 54.02 | 30.57 |
| 2 Ephemeroptera (larvae) | 9.79 | 12.74 | 7.51 |
| 3 Trichoptera | 4.47 | 12.70 | 22.92 |
| 4 Amphipoda | 8.88 | 2.72 | 2.47 |
| 5 Trichoptera (larvae) | | 0.13 | 1.13 |
| 6 Coleoptera | | 0.09 | |
| Diptera larvae | | | |
| 7 Similidae | 1.06 | 1.05 | 0.39 |
| 8 Crane fly | | 1.15 | |
| 9 Plant detritus | 11.11 | 3.63 | 1.37 |
| 10 Flying insects | | 8.17 | 0.30 |
| 11 Ants | | 0.26 | |
| 12 Arachnida | | 3.33 | |

were the second most abundant food items (Table 4). The number of the trichoptera increased (22.92%) during autumn (Table 4).

Regarding the Vacuity Index, from all specimens analyzed, 99 stomachs were completely empty, corresponding to a Vacuity Index of 49.5%. Furthermore, the majority of the empty stomachs were counted in autumn (52.27%), while in the other two seasons, this percentage ranged at a lower level, 48.28% during spring and 48.82% during summer.

The relationship among the preys, as presented in the Costello diagram (Fig. 6) provided some aspects of the species feeding strategy. Chironomids larvae were the most important type of

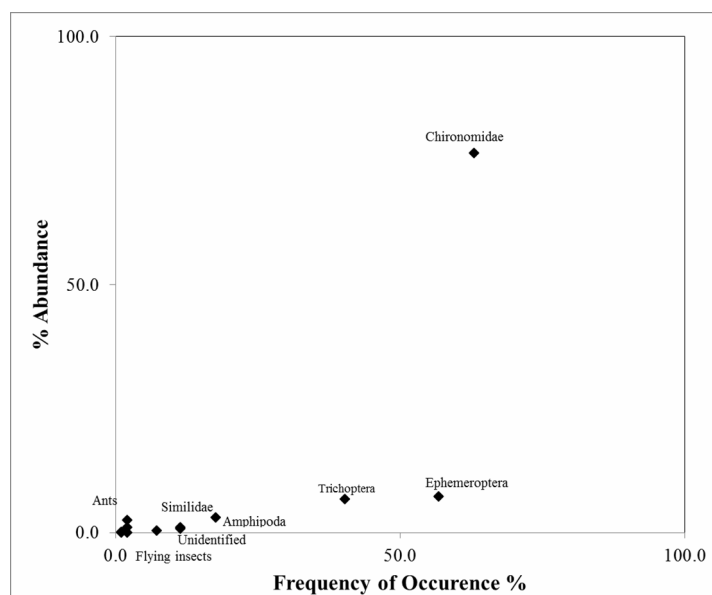


Figure 6. Plots based on the Costello graphical method, presenting the relationship among prey abundance and the frequency of occurrence (%FO) of food categories of *B. strumicae*.

food item both by number and by occurrence. Moreover, trichoptera and ephemeroptera were important food items by occurrence and less by number. Finally, there were food items such as amphipoda, flying insects, or ants, presenting less interest for *B. strumicae* diet. As it can be observed from the Costello diagram (Fig. 6), it seems that *B. strumicae* has a high degree of specialization in predation of the chironomidae, following a more general feeding strategy of capturing other types of food items, such as diptera larvae, coleoptera, amphipods, flying insects.

Discussion

Sex ratio

The sex ratio obtained in the present study was 1.66 : 1 (upstream) and 2.33 : 1 (downstream), with the males being, in both cases, the dominate sex. These findings were similar to the findings of Vasiliou & Economidis (2005), who obtained a ratio of 1.28 : 1 in favour of the males of *B. cyclolepis* from Doirani's Stream. However, there are cases like the one reported by Zutinic et al. (2014), who obtained a sex ratio equal to 1 : 1 for the population of *B. balcanicus* in Central Croatia, while the dominance of the females was reported by Vitali & Braghieri (1984) for the population of *B. barbus plebejus* in River Po, Italy. The aforementioned observed variation of the sex ratio might be the re-

sult of the environmental sex determination that is the diverse environmental conditions prevailed in the habitats during the birth and growth of the fishes promote the evolvement of one sex over the other (Kraak & Pen 2002).

Age & Growth

The maximum age observed in the Nestos' population was 4+, corresponding to five female and six male specimens (Table 1). Other species showed similar results, such as *B. cyclolepis* from Doirani stream where male barbels reached 5+ and females 9+ (Vasiliou & Economidis 2005). *Barbus strumicae* from River Dzerman and Dospat dam (belonging to the Nestos drainage, Bulgaria) recorded a maximum age of 6+ years (Dikov & Zhivkov 1985). Regarding *B. balcanicus* in river Ilova, the maximum age recorded by Žutinić et al. (2014) was 3+ for the female specimens and 2+ for the male specimens, while the maximum recorded age for the population of *B. barbus plebejus* in River Po was 8 (Vitali & Braghieri 1984).

The annual increment obtained in the current study was similar to the increment determined by Del Mar Torralva et al. (1997) and Herrera & Fernández-Delgado (1992) for the undisturbed populations of *B. sclateri* and *B. bocagei sclateri*, respectively. One reason that could explain this similarity could be the quite sort time that past since the construction of the dams in Nestos River (almost 18 years), which did not favour the differ-

entiation of the species' growth rate.

According to Przybylski et al. (2004) and Vasiliou & Economidis (2005), the dam lakes provide to the fish species a more stable environment from a physico-chemical perspective, in contrast to the highly variable downstream environment. Thus, it would be expected that the population of *B. strumicae* inhabiting the upstream part of the river to exhibit better growth performance than the downstream. However, no such differentiation was observed in the case of *B. strumicae* in Nestos River. This population presented a moderate growth rate ($K=0.165$) when compared to the Dzerman River ($K=0.293$) (Dikov & Zhivkov 1985) or to the population of *B. cyclolepis* from Doirani stream which presented the lowest growth rate ($K=0.112$) (Vasiliou & Economidis 2005). Similar results were reported for *B. balcanicus* inhabiting Ilova River and its tributaries, with the growth rate range from 1.12 to 1.28 (Žutinić et al. 2014). Bobori et al. (2006) indicated that species with low K , attained the L_{∞} slower and therefore tended to have longer lifespan than those with higher growth rate. The K value obtained in the present study for *B. strumicae* should be the result of optimum conditions prevailing in both the upstream and downstream part of the river.

Reproduction

Regarding the reproduction of the species, it was observed that the population of *B. strumicae* in Nestos River attained maturity at the same age that *B. sclateri*, as it is reported by Herrera & Fernández-Delgado (1992). According to the aforementioned authors, males of *B. sclateri* attained maturity earlier than the females, during their third year (2+) in contrast to the 4+ or 5+ years of the female. In the present study, the maturity and the spawning time of *B. strumicae* were in agreement with the results of Herrera & Fernández-Delgado (1992) on *B. sclateri*, and Vasiliou & Economidis (2005) on *B. peloponnesius* and *B. cyclolepis*. Moreover, Žutinić et al. (2014) reported that both males and females of the species *B. balcanicus* mature during their first year (0+), since the observed mature male specimens at a size of 67 mm and mature females at size of 82 mm.

All three species, *B. balcanicus*, *B. cyclolepis*, and *B. peloponnesius*, presented similar period for reproduction from late March-beginning of April until mid-July (Vasiliou & Economidis 2005, IUCN 2013). Similar period was presented also by Gona-

dosomatic Index variation in the present study. Fluctuation of the GSI index indicated that both female and male *B. strumicae* have the same reproduction period, with higher values of GSI obtained by the females.

The study of the size-frequency profile of the oocytes was similar to the one observed by Herrera & Fernández-Delgado (1992) and Vasiliou & Economidis (2005). However, the small number of oocytes with diameter > 1.05 mm (less than 300) in the samples captured in autumn, represents oocytes that probably were not released during the spawning period and probably undergo the process of atresia (Lefler et al. 2008). This fact indicates an extensive period of oocyte release, which is completed in summer.

Condition factor

Despite the slight variation of the condition factor, there is a seasonal variability, with two maxima, one for the males in mid-spring (April, 0.178) and early fall for the female (September, 0.164). According to Bagenal & Tesch (1978), the condition factor is affected by various factors such as nutritional or ecological but variation is also observed during the spawning period. Therefore, the first peak of maximum value of the factor, in April and presented by males, coincided with the start of the breeding season. The second peak, observed in September, could be associated with the start of the autumn season and the preparation of the species for the forthcoming winter season. Vasiliou & Economidis (2005), who also observed the presence of two peaks, attributed the increase of the Fulton's factor to the favourable conditions. On the other hand the decrease of the factor during summer is attributed to the attenuation of the fishes due to the spawning and the unfavourable environmental conditions (Encina & Granado-Lorencio, 1997, Vila-Gispert & Moreno-Amich 2001). Consequently, the increase of the factor in autumn, as it was observed in the current study, could be attributed to the favourable environmental conditions prevailed in autumn, such as increase of water flow and decrease of the temperature.

Diet & Vacuity Index

The diet composition of the Nestos' population was similar to that of *B. barbus* (Piria et al. 2005), *B. cyclolepis* (Rozdina et al. 2008) and *B. bocagei* (Magalhães 1993, Collares-Pereira et al. 1996). Despite the number of food items identified was 12, the

dietary spectrum was quite narrow since the species displayed a preference to the larvae of chironomids and ephemeroptera. As it was observed, the species presents a high specialization in the predation of the chironomidae larvae, which it appears to be its main food source throughout the year. To report the high abundance of plant detritus in the gut content, an item also observed in other species of the genus *Barbus* (see authors above listed). The ability of the species to consume plant detritus is an advantage compared to other benthic feeders in cases of low abundance of other types of preys (Magalhães 1992).

The present results provide the first insights into the biology of *B. strumicae* population of Nestos River. It must be noted that the species population appears to be in a good condition. Therefore, it is not required the establishment of any special management and protection plan. However, due to the special conditions prevailing in the downstream part of the river (function of HPPs) a continuous monitoring of the population is advised.

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