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# Comparative analysis of morphometric parameters and conditions factors of grayling 3+ (*Thymallus thymallus*) from the Krušnica River and the Martin Brod fish farm

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**Abstract**
The morphometric characteristics of fish are used to determine the difference between fish populations, and the fitness factor is one of the ways to monitor the influence of environmental factors on the general condition of fish. The aim of this study was to determine the morphometric characteristics and the fitness factor of graylings in their natural habitat and in fish farms. In this paper, a comparative analysis of morphometric characteristics and fitness factors of grayling individuals aged 3+ from the Krušnica River and the Martin Brod fish farm was performed. A total of 134 individuals aged 3+ were caught, ie 34 individuals from the Krušnica River and 100 individuals from the Martin Brod fish farm. The following parameters were analyzed in all individuals: total length, total length, weight, age structure, sex and fitness factor. Based on the obtained results, statistically significant differences were recorded for the total length, standard length, weight and fitness factor. In the Krušnica River, out of a total of 34 grayling, 14 grayling were males and 20 were females, while in the Martin Brod fish farm, 81 females and 19 males were determined.

**Keywords:** morphometric parameters, conditions factor, grayling, krušnica, martin brod

## Introduction
Research on fish populations is important for a variety of perspectives, including ecology, evolution, Behavior, conservation, and water management [1]. Using the morphometric characteristics of fish, it is possible to identify differences between fish populations [2], and the same parameters are used to determine fish [3]. Morphometric characteristics are progressive parameters that describe aspects of body shape [4]. Morphological plasticity represents the adaptability of fish to the complex and different ecological conditions in which fish are found [5]. A set of ecological factors, such as temperature, salinity, dissolved oxygen, radiation, water depth, and current flow, influence morphological variations between fish [6, 7, 8, 9]. In addition to numerous studies on wild specimens [10, 11, 12, 13, 14, 15, 16], many authors have investigated how the environment in which oily fish are raised in fish farms affects their morphology [17, 18, 19, 20, 21]. In view of the fact that the phenotypic characteristics of captive fish show a high degree of divergence, it is possible to find individuals with an extreme phenotype in fish farms [22]. As an indicator of general health status at the individual or population level, the fitness factor is used [23]. Fitness factor values represent the physical condition of fish and are a suitable parameter for comparing individuals of the same fish species, and are also a good indicator of differences between sexes, seasons, and sampling site [24]. Distribution of European grayling extends from the north of England and southern Scandinavia to the Loire River in southern France in the west, and reaches the Po River Basin in the south, and is present in the Dunav River Basin, while the eastern border of its natural habitat is the Urals in Russia [25]. Many grayling populations are highly endangered or eradicated, increasing the need to understand the ecological requirements of this species [26]. The causes of declining grayling populations are numerous, such as habitat destruction, watercourse regulation, Various pollutants, Introduction of allochthonous species, fishing, and environmental degradation [27]. For this reason, graylings are largely protected in Europe and are listed in the Council Directive 92/43 / EEC on the conservation of natural habitats and of wild fauna and flora - Annex V, as well as the Berne Convention - Annex III [25]. Based on documented literature data, the aim of this study was to determine the morphometric characteristics and condition of graylings from their natural habitat and fish farms, and to determine whether the environment inhabited by the grayling affects his morphometric parameters.

## Materials and methods
### Locality
Krušnica River, only 6.5 m long, is located in the northwestern part of Bosnia and Herzegovina and is a right tributary of the Una River. The Krušnica spring is located 5 km from Bosanska Krupa, with the northernmost point at 44o 50 '41.4' 'north latitude and the southernmost point at 16o10'5.8' 'south latitude (Fig. 1). One of the largest full-system salmonid ponds is located upstream of the bridge on the Unac River in Martin Brod itself with the northernmost point at 44o29'17 " north latitude and the southernmost point at 16o8'28 " south latitude (Fig. 1).
The pond was built in 1985 and has 100 pools, 10,000m2. Production of juvenile grayling began in 2001, and until then, juvenile rainbow trout and juvenile brown trout have been farmed.
Major headings are to be column centered in a bold font without underline. They need be numbered. "2. Headings and Footnotes" at the top of this paragraph is a major heading.

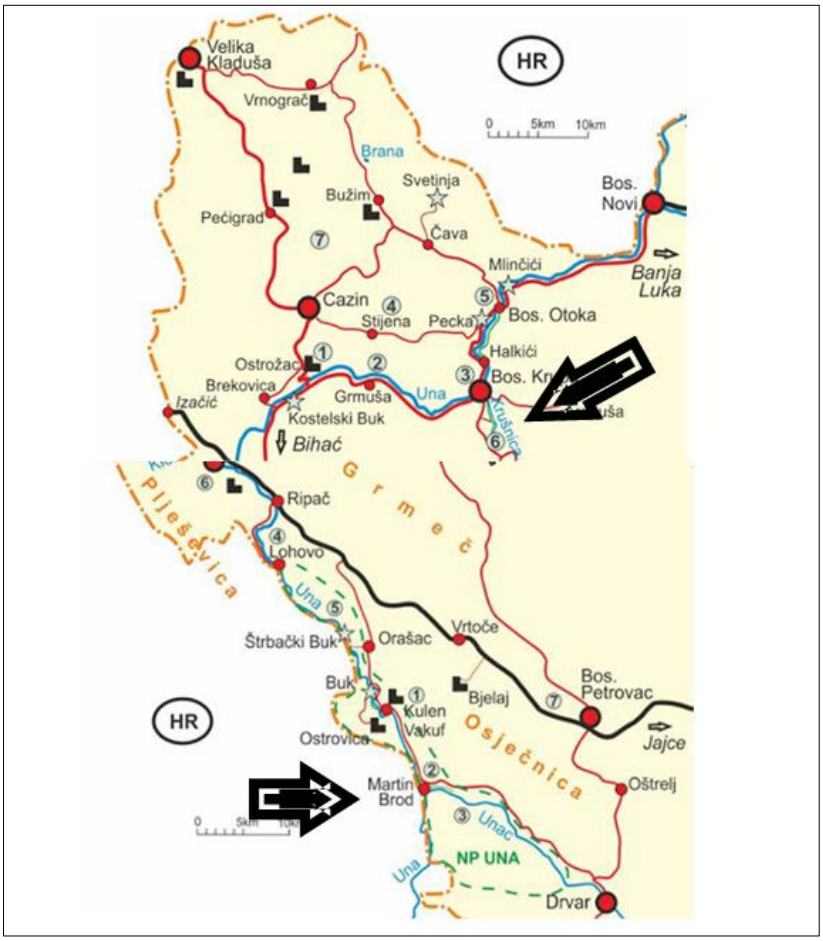


**Fig 1:** Krušnica River and Martin Brod fish farm

**Fishing**

The catch of grayling from the Krušnica River was done in accordance with the current legislation on freshwater fisheries in Bosnia and Herzegovina. For this reason, fishing on the Krušnica River was carried out from May to December 2006 and 2007 using the fly fishing technique. The accessories used were a fly rod, action 5, Half-twelve-Shakespeare, a Shakespeare wheel as a roll and a rope of the DTF-6-Cortland type. Sampling of fish at the Martin Brod pond was done in May and June 2006 and November 2007, and a hand bag with different eyelet diameters was used when taking the fish.

**Morphometric parameters**

In all analyzed individuals, body weight, total and standard length, age and sex were determined. The total and standard length of the caught individuals was determined with the help of an ichthyometer, and the mass of the caught individuals was determined with the help of a laboratory scale of the type "Sartorius BL 3100" with a capacity of 3,100 g and an accuracy of d = 0.1 g. According to [28] and [29], the age structure of ichthyopopulation of an aquatic biotope is determined by determining scleritic rings on the scales. The scales were cleaned with a mild solution of KOH, and then permanent preparations were made for each individual separately. After that, using a monoocular laboratory magnifier type "Optus" Leuchtlupe, Art. Nr. 96-17000, with 2x and 4x magnification and "Zuzi" binocular microscope Mod. 50137007 N0, serie A44253, "Auxilab" - Spain, with magnification of the eyepiece WF 10x, and magnification of the lens 4x, 10x, 40x, scleritic rings on the scales were counted and the age of the caught individuals was read. Gender determination was performed by gonadal dissection. Fulton 's nutritional coefficient in all individuals was analyzed as a condition factor in the work. The Fulton coefficient is calculated by the formula:

$K = W * 100 / L^3$ where

W-mass of fish in grams,

L-standard length of fish in centimeters [24].

**Statistical analysis**

Data were processed using Exel 2010 to determine the mean and range. For the analysis of the variance test, t tests (two samples) were used whose confidence interval was 95% and absolute precision 5%.

**Results and Discussion**

In this research, a total of 134 grayling from the Krušnica River and the Martin Brod fish farm were analyzed. For the purpose of this study, only individuals aged 3+ were considered. The average standard length of grayling individuals from Krušnica was 27.51 ± 0.02 cm, while the total length was 31.76 ± 0.02 cm and the body weight 330.29 ± 0.63 g. In grayling individuals from the Martin Brod fish farm recorded a higher value of standard (28.33 ± 0.01 cm) and total (33.05 ± 0.01 cm) length, while the mean value of mass (308.06 ± 0.43 g) was lower compared to grayling individuals from the Krušnica River (Table 1). The fitness factor was slightly higher in individuals from the Krušnica River (1.62 ± 0.00) compared to individuals from fish farms (1.35 ± 0.00). The sex of all caught individuals was determined, and the ratio of individuals in the Krušnica River

was 14:20 in favor of females, and in the Martin Brod fish farm 19:81 also in favor of females. The T test (two samples) was used to analyze the difference between individuals from the Krušnica River and the Martin Brod fish farm, and significant differences were recorded for standard length, total length, weight and fitness factor.

**Table 1:** Morphometric parameters and condition factor of grayling from Krušnica River and Martin Brod fish farm

| | ***Krušnica*** | | ***Martin Brod*** | | ***Sig.*** |
|---|---|---|---|---|---|
| | **Mean ± stdv** | **Range** | **Mean ± stdv** | **Range** | |
| SL (cm) | 27,51 ± 0,02 | 21,00-32,00 | 28,33±0,01 | 25,00-32,00 | 0,000* |
| TL (cm) | 31,76 ± 0,02 | 26,00-37,00 | 33,05±0,01 | 29,00-37,00 | 0,001* |
| W (g) | 330,29 ±0,63 | 200,00-475,00 | 308,06±0,43 | 200,20-418,50 | 0,005* |
| CF | 1, 62 ± 0, 00. | 1,11-2,88 | 1, 35 ± 0,00. | 1,01-1,64 | 0,000* |

Morphometric methods determine the dimensions of individual parts of the body, as well as the relationships between them and between different populations. In order to ensure the reliability of the comparisons, it is necessary to take into account the age and sex of the compared fish. This classical method of the ratio of individual body lengths has proven to be particularly suitable for determining the ecological effects on fish morphology [30, 31]. According to the available literature data, there are numerous studies on grayling in the natural habitat, but very little information on grayling grown under controlled conditions, and for this reason this research is very important. The data obtained by this research differ to a lesser extent in relation to the data of previous research. The average body length of grayling aged 3+ from the Balkan population was about 24.0 and 30.0 cm [12], which is less than the average body length of grayling individuals from the Krušnica River and the Martin Brod fish farm. Specimens of grayling from the Caspian population with an average length of 18.0 and 24.0 cm, also aged 3+ are much smaller compared to our research [32]. The populations of grayling from the rivers Lim, Sava Bohinjka and Una, regardless of their natural habitat or fish farm, have a similar size [33], which is in line with our results. The recorded average lengths of European grayling in the rivers of Ukraine Lyutyanki (18.7 cm), Shypitu (13.2 cm) and Pesyi (22.1 cm) are much lower than the average lengths of grayling in our studies [34]. Three-year-old grayling from the rivers Drina, Bosna and Vrbas can reach a length of 27.2-37.5 cm [35], which is quite similar to the mean values of body length of grayling from our research. In the research conducted [36] in the river Kupa, the recorded values of the total length of grayling ranged from 17.0 to 41.0 cm, with different values in relation to the results of our research. According to [37], a 15-month-old grayling in the Unac River can reach a length of 28.0 cm, equal to the smallest specimens from our research, and the growth intensity of freshly stocked grayling in the Unac River is equal to the growth of a three-year-old grayling in a fish farm. The average weight of grayling from the river Krušnica was 330.29 g, and from the fish farm Martin Brod 308.06 g and much higher average values of body weight of grayling compared to the recorded value in the following rivers Lyutyanki 80.2 g, Shypitu 30.9 g and Pesyi 119, 9 g [34]. The mass of grayling in the river Kupa ranged from 40 to 700 g [36], and is much higher than in our research, but it is important to note that for the purpose of this research we used only individuals of age class 3+, which is probably the reason such a deviation from the literature data. Based on the values of fitness factors, it can be concluded that the conditions for the growth and development of grayling are much more favorable in the natural environment, ie in the river Krušnica in relation to the fish farm Martin Brod. The values of the fitness factor of grayling from the river Kupa (1,094) are lower in relation to the values of the fitness factor of individuals from the river Krušnica and the fish farm Martin Brod. Similar values of fitness factors as in our study were recorded for grayling individuals in the Turiec River (1.55) and in the Slana River (1.70) [38, 39]. Much more unfavorable living conditions for grayling were recorded in the Pomerania River, since the fitness factor (1.00) was much lower compared to the results of our research. In the river Una, the recorded value of the condition factor for grayling was 1.09 and in the river Sana 0.84 [40], which are much lower values compared to the values obtained by this research.

## Conclusion

The natural habitat is a far better environment for the growth and development of grayling as opposed to the breeding of this species in fish farms. Regardless of the fact that grayling individuals receive continuous food in the fish farm, grayling individuals from the Krušnica River had a higher average body weight as well as a higher fitness factor. Therefore, grayling is a species that requires very specific conditions and conditions that should be approximately similar to the natural environment for breeding in fish farms.

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