

Original scientific paper

# **EVALUATION OF THE BREEDING VALUE OF RAINBOW TROUT (ONCORHYNCHUS MYKKISS) FOR BODY DEVELOPMENT TRAITS USING A LINEAR MODEL**

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#### SUMMARY

The objective of this research is to assess the breeding value of male individuals and calculate heritability values for body weight and body length at 12 months of age in the rainbow trout population in the Republic of Serbia. The study utilized data on the physical development of trout obtained through the Center for Fisheries and Applied Hydrobiology "Mali Dunav", Experimental Estate "Radmilovac" of the Faculty of Agriculture, University of Belgrade (CEFAH) Breeding Program for rainbow trout in the Republic of Serbia. The fish were raised in individual tanks under tightly controlled conditions at the CEFAH, until they reached 5 months of age. At this point, the fish were marked (with PIT tags Norway) and transferred to commercial fish farms, where monitoring of their production results continued. To estimate the breeding value for body weight and body length of rainbow trout at 12 months of age, we analyzed data collected between 2016 and 2020, which included a total of 2014 individuals. The breeding values were estimated using a sire model. The mixed model incorporated fixed effects (spawning year and sex) and a random effect for the individual sire. The heritability estimate at 12 months of age was 0.44 for body weight, and 0.45 for body length. The breeding values were estimated for 107 sires, with body weight values ranging from -37.83 to +37.83 g and body length values ranging from -14.00 to +14.00 mm. The substantial variation observed within the studied rainbow trout population suggests the potential for further genetic improvement. The high heritability values confirm that further selection can be based on the phenotype. The inclusion of high-quality and validated males in breeding programs will contribute to the enhancement of production characteristics within the existing rainbow trout population in Serbia.

#### **INTRODUCTION**

The rainbow trout (*Oncorhynchus mykiss*) originates from North America and has successfully been cultivated worldwide (Marković & Mitrović-Tutundžić, 2003). Females can produce between 1300 to 2600 eggs for every kilogram of body weight, while males typically have around 10 million spermatozoa per mm<sup>3</sup> of sperm (Marković & Mitrović-Tutundžić, 2003). Trout readily adapts to new growing conditions. As the most widely cultivated coldwater freshwater fish globally, rainbow trout serves as a crucial model for various research fields, often earning the nickname "aquatic laboratory rats" (Palti et al., 2009). The paramount characteristic of trout farming is its high growth rate, which is challenging to maintain under unfavorable farming conditions (Sae-Lim et al., 2013). Trout production is rapidly expanding, and most breeders prefer using younger fish from their own broodstock. One





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#### Key words:

rainbow trout, phenotypic and genetic variability, heritability, breeding value

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significant advantage of raising rainbow trout is that a small number of sexually mature individuals can produce a large number of offspring for the next generation. However, small populations can lead to higher inbreeding rates (Yousefian & Nejati, 2008).

In recent years, programs for genetic improvement of domestic animals based on traditional selection have been highly successful, leading to significant productivity enhancements (Dedović, 2015). The success of selection primarily hinges on assessing the breeding value and employing alternative breeding techniques. Selection typically involves identifying individuals with the most favorable genotypes for reproduction and the generation of offspring. However, since the actual genetic values are often unknown, genotype assessment poses challenges (Dedović, 2015). In aquaculture, selection programs developed somewhat later due to a lack of estimated heritabilities, coefficients of variation, and correlations for economically significant traits (Marković et al., 2009). Unlike animal husbandry, where pedigree data is routinely recorded and represents only a minor portion of selection costs, aquaculture faces difficulties and expenses in collecting data, particularly when individuals lack chip codes, necessitating DNA analysis (Morton & Howarth, 2005). In aquaculture, families are frequently reared separately in separate tanks to maintain accurate pedigree records, potentially leading to a dominant environmental influence (Morton & Howarth, 2005). Implementing selection programs can yield substantial improvements in fish growth while reducing costs (Kause et al., 2006). In aquaculture, the primary goal of breeding programs is to enhance economically crucial traits like growth. Growth traits are the most common selection criteria due to their economic significance, ease of measurement, high phenotypic variation (approximately 30% coefficient of variation), and moderate to high heritability (Gjedrem, 1992; Hu et al., 2013). Planned breeding can result in a genetic gain of 10 to 20% per generation, surpassing gains in other farmed animals (Hershberger et al., 1990; Gjedrem & Thodesen, 2005; Gjedrem, 2010; Gjedrem et al., 2012). Successful selection accelerates genetic progress, leading to genetically superior animals (Spehar et al., 2011). Regarding genetic trait improvement in selection, body weight at 10 to 12 months is frequently included (Jobling, 2003).

Breeders and animal farmers aim to rapidly improve the genetic traits of interest, where evaluating the breeding value constitutes the most intricate and critical aspect. Assessing the reproductive value of fish has a significant role in breeding and selection endeavors as it informs the selection of parent pairs based on their estimated value. An individual's breeding value represents the worth of the genes it will pass on to its offspring (Dedović, 2015). Evaluating an animal's breeding value encompasses a multitude of influences that cannot be directly measured, making the evaluation process highly complex. The objective of the breeding value estimation is to eliminate the impact of non-genetic factors and accurately estimate the additive genetic component of variance. The additive effect of a gene is the effect it has independently of other genes in the genotype, and it is the only aspect that is reliably passed on to offspring and can be precisely assessed through certain methods. For these reasons, the additive effect is of great importance as part of the total hereditary component in modern animal husbandry, as it best reflects the breeding value of the examined individual (Bogdanović et al., 1994). To assess the breeding value effectively, data must be available as a basis for evaluation, and the choice of the evaluation method depends on the data structure. A reliable breeding value assessment necessitates highly accurate measurement and recording of production and other assessed characteristics (Janković, 2017). The heritability value serves as a valuable indicator of the phenotypic expression of specific animal traits when selecting parent pairs for the next generation. The phenotype of an animal for a given trait is a reliable indicator of the animal's breeding value if the heritability value is known.

## MATERIAL AND METHODS

This research was carried out using individuals from the selection program conducted at the Center for Fisheries and Applied Hydrobiology "Mali Dunav," located at the "Radmilovac" Experimental Estate of the Faculty of Agriculture, University of Belgrade. The individuals used for implementing the selection program and obtaining the selected families of rainbow trout were collected from various fish farms in 2010. These fish farms are situated in different regions of Serbia: Lazić (Despotovac), Žubor (Despotovac), Bast komerc (Despotovac), Jablanica (Boljevac), Vrelo (Sokobanja), RM Stubica (Žagubica), Braduljica (Ivanjica), and Dini trade (Pirot). When forming the initial broodstock, the aim was to gather individuals with a diverse genetic structure from different locations. One of the criteria for selecting fish farms to establish the broodstock was that no fish from other fish farms had been introduced to these fish farms in the preceding 15 years.

After collecting the individuals, a crossbreeding scheme was devised, and spawning was conducted following the standard procedures (Marković & Mitrović-Tutundžić, 2003). Fertilized eggs were placed on incubation tub grids (15 l) in a darkened room. To protect the eggs from artificial lighting, the tubs were additionally covered. Water flow was adjusted to prevent movement of the planted eggs. The water temperature during the 30 to 32-day incubation period ranged from 9 to 11°C. When approximately two-thirds of the larvae began swimming, the process of

acclimatizing them to high-protein food commenced. Initially, larvae were fed every 15 minutes, with a gradual reduction in the number of daily meals as their body weight increased. A few days after the trout larvae started swimming, 100 larvae from each family were separated and transferred to individual tanks with a volume of about 120 l for further rearing. To track production results for each family, the individuals were marked (chipped). Chipping was done when the fish were five months old, or when they reached a body weight of approximately 5 gr. Up to 50 individuals with larger body weight were selected and marked out among the existing 100 individuals per family. An injector was used for marking, injecting a microchip into the abdominal cavity (PIT tags, Norway). Each fish received an identification number, consisting of a unique combination of 10 symbols, which were recorded in the database (BKTAGCOM, Norway) along with other production characteristics. Prior to being marked, the trout fry were anesthetized in a clove solution (manufactured by Probotanik) to reduce stress and facilitate body weight measurement using a precision scale (RADWAG THB-600, MAX 600 g, d=0.01 g, Poland) and body length measurement using an ichthyometer. After marking, the young fish were transported from the CEFAH to the raceway system at the "RM Stubica" company in Sisevac, where they were reared until 2016. After 2016, rearing continued at the "Bilans M" company fish farm in Perućac. The trout were fed with high-value protein food produced by Skreting (Italy), following the manufacturer's recommendations provided in tabular instructions. At the end of each year (in December), measurements of body weight and body length were taken for each individual, and these values were recorded in the database. At two years of age, in addition to the aforementioned basic data, data about the individual's sex, sexual maturity, and readiness for spawning were also entered into the database.

In order to estimate the breeding value for the traits of body weight and body length in rainbow trout, we used the data collected for the production traits (body weight and body length at 12 months – when trout begin to reach a size suitable for consumption and are most commercially valuable) from a database covering the period from 2016 to 2020. In this research, the breeding value of 107 individuals was evaluated. The number of individuals (offspring for which the breeding value of their fathers was estimated) for which body weight and body length were measured at 12 months during the mentioned research period was 2014.

Basic statistical indicators of phenotypic expression and variability of body weight and body length were calculated using standard statistical procedures within the SAS software package, employing the PROC MEANS procedure (SAS Inst., Inc., Cary, NC). Variance components were calculated using the REML method, assisted by the GLM procedure within the SAS software package. The following mixed model was employed:

$$Y_{ijkl} = \mu + G_i + P_j + T_k + o_l + e_{ijkl}$$

where:

 $Y_{ijkl}$  - phenotypic manifestation of the examined trait

 $\mu$  - general population average

G<sub>i</sub> - fixed effect of *i*-th year of birth

 $P_i$  - fixed effect of the *j*-th gender of the individual

 $T_k$  - fixed effect of the *k*-th tank in which the individuals were raised

 $o_1$  - random effect of the *l*-th father of the individual

e<sub>iikl</sub> - random error

The breeding value was estimated using the BLUP method with the sire model. Data coding and the breeding value assessment were performed using the PEST program package, while variance components were calculated using the VCE (version 6) program package.

The applied mixed model for the breeding value estimation had the following structure:

$$Y_{ijkl} = \mu + G_i + T_j + P_k + s_l + e_{ijkl}$$

where:

 $\begin{array}{l} Y_{ijkl}-\text{represents the observed trait} \\ \mu-\text{denotes the population average} \\ G_i-\text{accounts for the fixed effect of the spawning year} \\ T_j-\text{represents the fixed influence of the tank} \end{array}$ 

# $P_k$ – signifies the fixed influence of gender $s_1$ – corresponds to the random effect of the sire

 $e_{ijkl}$  – captures the random error.

## **RESULTS AND DISCUSSION**

Table 1 displays the phenotypic expression and variability in body weight and body length of rainbow trout at 12 months of age. A total of 2014 individuals were measured at this age. The average weight at 12 months was 283.70 g, with a standard deviation of 64.43 g and a coefficient of variation of 22.71%.

Table 1. Indicators of phenotypic expression and variability in body weight and body length at 12 months of age

Traits	Ν	x	Min	Max	$S^2$	Sd	Cv (%)
Body weight (g)	2014	283.70	50.00	513.00	4152.04	64.43	22.71
Body length (mm)	2014	291.80	160.00	355.00	629.18	25.08	8.59

The average body length at 12 months of age measured 291.80 mm. The standard deviation was 25.08, and the coefficient of variation stood at 8.59%. The coefficient of variation for body weight at 12 months of age is lower (22.71%) compared to the findings obtained by Ivanović (2005) and Gjedrem (1992) in their research ( $\approx$  30%). This observed variability underscores the significant phenotypic diversity within the examined traits and highlights the potential for genetic improvement.

Table 2 presents the F-values depicting the impact of the included factors on the traits of body weight and body length at 12 months of age.

Table 2. F-values for the included factors							
Traits	Spawning year	Tank	Sex				
Body weight (g)	66.05**	5.73 <sup>NZ</sup>	39.19**				
Body length (mm)	109.45**	4.74 <sup>NZ</sup>	2.46*				

Based on the results presented in Table 2, it can be affirmed that the influence of spawning year and sex on body weight and body length at 12 months of age was statistically significant. This finding aligns with similar conclusions reached by Ivanović (2005) and Mulder (2007). Interestingly, the pool where the fish were reared showed no statistical significance concerning the observed traits. This outcome was expected since the fish were raised in identical environmental conditions.

As part of our research, we calculated the heritability for body weight and body length at 12 months of age. Table 3 provides details on the variance components along with the calculated heritability for the observed traits.

Table 3. Values of variance components and heritability						
Traits	Sire variance	Additive genetic variance	Environmental variance	Phenotypic variance	Heritability	Standard error of heritability
Body weight (g)	519.00	2076.02	2611.74	4687.76	0.44	0.01
Body length (mm)	78.64	314.59	374.96	689.55	0.45	0.01

The heritability values for body weight and body length at 12 months of age are notably high, at 0.44 and 0.45, respectively. These heritability values indicate a strong connection between an individual's phenotype and genotype, allowing for further selection based on phenotype through individual selection, as demonstrated in studies by Kause et al. (2002, 2007), Hu et al. (2013), and Sae-Lim et al. (2013). The approximately equal heritability values for body weight and body length are justified by a high positive correlation between these two traits. Heritability values exceeding 0.40 affirm that the phenotype serves as a reliable indicator of the breeding value, as corroborated by Cassell (2009).

When assessing the breeding value, it becomes imperative to rank and select individuals with the most favorable values. Precise and dependable breeding values necessitate the accurate application of genetic parameters and measurements obtained from the animals, as well as the performance results of their ancestors, relatives, and descendants, as elucidated by Đedović (2015). The breeding value pertains to the worth of an individual for a specific trait within a breeding program, revealing the additive value of the genes that the individual will pass on to

its offspring. This is the primary reason why the selection of parental pairs is grounded in the breeding value of the individual, as emphasized by Janković (2017).

Given that this research assessment included 107 individuals, Table 4 displays the estimated reproductive values for body weight and body length at 12 months of age for the ten best-ranked and ten worst-ranked individuals.

Table 4. Estimated breeding values of male rainbow trout at 12 months of age					
Traits -	Top ranke	ed individuals	Worst ranked individuals		
Traits	ID number	Breeding value	ID number	Breeding value	
	487	37.835	409	-37.835	
	413	35.596	502	-33.261	
	421	31.275	491	-31.977	
	484	28.609	501	-31.275	
Dedrerselaht (a)	478	28.271	404	-28.609	
Body weight (g)	327	26.558	489	-26.558	
	422	26.077	388	-26.14	
	445	25.169	415	-25.169	
	391	24.233	470	-24.233	
	402	20.468	504	-20.554	
	478	14.003	470	-14.003	
	422	11.292	502	-11.657	
	413	11.274	399	-11.595	
	421	10.22	501	-10.22	
Dedaulan ath (ama)	487	10.072	409	-10.072	
Body length (mm)	473	9.677	388	-9.777	
	475	8.685	394	-9.677	
	445	8.417	504	-9.435	
	397	8.082	341	-8.685	
	477	6.542	415	-8.417	

The obtained results reveal that the male with the number 487 exhibited the highest breeding value for body weight, while the male numbered 478 had the highest breeding value for body length. Progeny of these two highly ranked males outperformed others in terms of body weight at 12 months, and this trend extends to the rest of the males within the group of best-ranked sires based on their estimated breeding values. Conversely, the male with the number 409 ranked the lowest for body weight, and male 470 ranked the lowest for body length. Since the breeding values are estimated based on progeny, it is considered that the accuracy of these values is high, as corroborated by Sae-Lim et al. (2017) in their research.

# CONCLUSION

Based on the thorough examination and analysis of the obtained results concerning the phenotypic expression and variability of fish body weight, it has been established that there are significant variations within the examined population of rainbow trout. This underscores the potential for their further genetic improvement. High heritability values suggest that further selection can be based on phenotypic characteristics. Incorporating high-quality and proven males into breeding programs will substantially contribute to enhancing the production traits of individuals within the population. Assessing the breeding value is particularly crucial for males, given their capacity to produce a large number of offspring through artificial spawning techniques. The ranking of males assumes significance as it aids in the selection of suitable sires, expediting the realization of breeding objectives. By identifying superior animals to serve as parents for the next generation, breeding programs can achieve maximum genetic progress. The cornerstone of this process lies in the swift and dependable assessment of the breeding value of potential selection candidates.

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