

# Influence of temperature and stocking density during rearing on larval blue bream, *Ballerus ballerus* (L.)

Przemysław Piech, Roman Kujawa, Joanna Nowosad, Dariusz Kucharczyk, Katarzyna Targońska, Mariusz Szmyt

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Abstract. This study determined the effects of stocking density and water temperature on the rearing parameters and survival of larval blue bream under controlled conditions. Two experimental larval blue bream rearing variants were conducted. In the first experiment, larval blue bream were reared at different stocking densities of 20, 40, 80, and 120 indiv. dm<sup>-3</sup> at 25°C. In the second experiment, larvae were reared in water at 15, 20, 25, and 30°C at a stocking density of 40 individuals per dm<sup>3</sup>. The larval blue bream achieved the highest growth rate at the lowest density tested (20 indiv. dm<sup>-3</sup>; 69.4 mg at an average length of 26.8 mm) and at the highest temperature tested (30°C; 80.1 mg at an average length of 28.4 mm). The final larval survival rate during rearing at different temperatures ranged from 95.2 to 97.6%, while the final larval survival rate at different stocking densities ranged from 91.4 to 94.6%. The lowest growth and

P. Piech [E], R. Kujawa, K. Targońska, M. Szmyt Department of Ichthyology and Aquaculture, Faculty of Animal Bioengineering, University of Warmia and Mazury, 10-719 Olsztyn, Poland E-mail: przemyslaw.piech@uwm.edu.pl

J. Nowosad Department of Ichthyology, Hydrobiology and Aquatic Ecology, National Inland Fisheries Research Institute 10-719 Olsztyn, Poland

J. Nowosad, D. Kucharczyk Department of Research and Development, Chemprof, Gutkowo, 11-041 Olsztyn, Poland survival rates were recorded for larvae reared in water at  $15^{\circ}$ C and at a density of 120 indiv. dm<sup>-3</sup>.

**Keywords**: blue bream, growth rate, survival rate, controlled conditions, stocking material, cyprinid fish

## Introduction

Protecting the biodiversity of fish fauna has become a key element in preserving fish stocks in lentic and lotic inland waters. Biodiversity conservation is not possible without effective environmental protection, which refers to reducing progressive anthropogenic pressure such as water pollution, excessive land reclamation, riverbed aggregate extraction, etc. (Goryczko and Witkowski 2009). In situ and ex situ methods, from designating conservation sizes to establishing broodstocks, are also important elements of species conservation. This applies mainly to fishes that are threatened with extinction, commercially important for human consumption, or important to recreational fisheries (Szmyt et al. 2021). Maintaining fish populations at adequate levels in inland waters is supported by the production of stocking material in aquaculture (conservation aquaculture; Śliwiński et

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al. 2016, Zakęś et al. 2016). As a rapidly growing industry, aquaculture has become an essential tool in combatting the deteriorating state of the environment. As a result of the development of aquaculture, many mechanisms, including artificial spawning protocols, breeding protocols, and indoor cultured stock, have been developed to protect not only economically valuable species, but also species important for the biodiversity of reservoirs, including indicator species and endangered species (Radwan et al. 2002, Gorzel and Kornijów 2004, Kujawa et al., 2019, Kucharczyk et al. 2021, Kujawa and Piech, 2021). High costs of water, production, and staff salaries are often barriers to starting to farm/rear new species (Hakuć-Błażowska et al. 2010). Despite this, growing global awareness and concern for ecosystems provides an opportunity for farmers to introduce new species into aquaculture and to develop their rearing protocols for stocking purposes and supporting natural populations.

One such species is blue bream, Ballerus ballerus (L.), a rheophilous cyprinid that inhabits the lakes and lower reaches of the larger rivers of the North Sea and Baltic Sea catchments (Weser, Elbe, Oder, Vistula, up to the Neva Basin, southern Sweden and southern Finland), the Danube basin (from upper Austria, Lake Neusiedler and Platten), the northern tributaries of the Black Sea and the Sea of Azov (Dniester, Dnieper, Don), up to the Volga and the Urals (Hesse and Witkowski 1996, Tadajewska 2000). The body shape of blue bream resembles those of white bream, Blicca bjoerkna (L.), and white-eye bream, Ballerus sapa (Pallas), but unlike these species, it has a relatively high number of scales on the lateral line 62-77 (white bream 50-58; white-eye bream 47-52; Levin and Levina 2014). It feeds on cladocera and copepod zooplankton and benthos. Despite its negligible economic value, it is an important species in the food pyramid of aquatic habitat reservoirs, especially in the Oder River catchment.

Developing efficient protocols for the production of stocking material under controlled conditions requires intensive, wide-ranging research on important aspects of breeding. Good examples are recent studies on the effects of temperature, stocking density, and feeding regime on larval maraena whitefish (*Coregonus maraena* (Bloch) (Sebesta et al. 2018, Stejskal et al. 2021a, 2021b), which facilitated developing a complete protocol for producing stocking material of this species. A suitable reproduction method under controlled conditions using hormonal agents has already been successfully developed for blue bream, *Ballerus ballerus* (L.) (Piech and Kujawa 2021). A feeding model for this species has also been developed (Piech and Kujawa 2022), and the effects of different levels of water salinity on the breeding parameters of the larvae of this species have been investigated (Piech and Kujawa 2023).

The aim of this study was to investigate the effects of stocking density and water temperature on the growth and survival of larval blue bream, *B. ballerus*, under controlled conditions.

## Materials and Methods

The blue bream spawners were caught with gill-nets in mid-May during their natural spawning period in Lake Dąbie (northwest Poland). The larval blue bream originated from reproduction carried out under controlled conditions (Piech and Kujawa 2021).

#### Stocking density

Larval blue bream were reared in a RAS comprising 12 flow-through tanks designed for rearing larvae. Each tank had a working volume of 25 dm<sup>3</sup>. The larval blue bream (5 days post hatch (DPH); 7.6 mm average body length) were counted and placed in the tanks at densities of 20, 40, 80, and 120 indiv. dm<sup>-3</sup>. The stocking densities of single tanks were 500, 1,000, 2,000, and 3,000 larvae. The water temperature during rearing was  $25.0 \pm 0.2^{\circ}$ C. The stocking densities tested in this study were chosen based on Kujawa (2004).

#### Temperature

Each experimental system consisted of three rearing tanks, each of which had a working volume of 25

dm<sup>3</sup>. Larval blue bream, 5 DPH with an average body length of 7.6 mm, were counted and placed in each of the rearing tanks at a density of 40 indiv. dm<sup>-3</sup>. The stocking density of a single tank was 1,000 larvae. The water temperatures in the variants were 15.0, 20.0, 25.0, and 30.0  $\pm$  0.2°C. The temperatures tested in this study were chosen based on Kujawa et al. (2015).

Each of the experimental systems used to test the effects of temperature on the growth and survival of larval blue bream included a 200 dm<sup>3</sup> filter tank with a floating medium biological bed. The filter medium was bactoballs used in denitrification filters (AB Aqua Medic GmbH, Bissendorf, Germany). In the mini circuit, in which the effects of stocking density on larval blue bream growth and survival were tested, the filter tank had a capacity of 800 dm<sup>3</sup>. To ensure adequate oxygen conditions, a RESUN LP 100 membrane blower (Shenzhen XingRisheng Industrial Co. Ltd., Shenzhen, China) was also used. Air was distributed in the filter tanks using silicon tube diffusers (Akwatech Systemy Sp. K., Poznań, Poland). Aeration cubes were installed in each of the rearing tanks through which air from the blower was also pumped. A sprinkler system provided a continuous supply of oxygenated, purified water to the tanks.

The water oxygen content was measured with a YSI multiparameter meter (YSI Incorporated Brannum Lane Yellow Springs, Ohio, USA) and was within 6.5–7.8 mg  $O_2$  dm<sup>-3</sup>, the water pH range was 7.1–7.3. The ammonia and nitrite contents were measured in the morning with a HANNA INS HI 83200 with reagents (Hanna Instruments, Woonsocket, RI, USA), and neither was detected in the water during the experiment (Nowosad et al. 2013).

## Sample collection

Monitoring measurements of larval weight and total length were done every 4 d (on days 1, 5, 9, 13, 17, and 21 of rearing). Each sample comprised thirty individuals (n=30) caught randomly from each tank. These were collected in the morning before feeding. The fish were weighed to the nearest 0.1 mg and measured to the nearest 0.1 mm.

During the experiment, the number of dead larvae was counted daily and the final survival rate percentage was calculated. This was expressed as the ratio of the number of larvae at the end of rearing to the number of larvae at the start of the experiment. Cumulative mortality curves were plotted for each rearing variant based on daily counts of dead larvae. Average larval weight gain (mg) was calculated for larvae from each experimental group at the end of the three-week rearing period (Imanpoor et al. 2012). Measurements were used to calculate survival, the index of total length gain per unit time (ITL), specific growth rate (SGR), survival, and larval biomass (SBR) (Brown 1957, Peňáz et al. 1989). These values were used to calculate the relative growth rate (RGR) and the relative biomass growth rate (RBR) from the beginning of feeding to the end of the experiment (Myszkowski 1997). Growth rates (SGR and RGR) for body length were calculated in the same way.

## Fish feeding

The larval blue bream were fed manually *ad libitum* four times daily at three-hour intervals (08:00, 11:00, 14:00, 17:00; Piech and Kujawa 2022). The food was live brine shrimp, *Artemia* sp., nauplii (Ocean Nutrition Europe, Essen, Belgium) (Sorgeloos et al. 1977).

#### **Data Analysis**

The number of dead fish recorded daily were used to plot cumulative mortality curves for each fish rearing variant. To compare results, the relative final mean length (RFL), the relative final mean weight (RFW), and the relative final mean biomass (RFB) of the experimental fish were calculated, assuming that the length, weight, and biomass of the control fish (21 d feeding regime with live brine shrimp nauplii) at the end of the experiment was 100%. Duncan's multiple range test (P  $\leq$  0.05; Duncan 1955) was used to determine the significance of differences in mean

#### Table 1

Results of rearing larval blue bream (*B. ballerus*) at stocking densities of 20,40, 80, and 120 indiv. dm<sup>-3</sup>. Mean value  $\pm$  SD. Results in rows with the same letter index are not statistically significantly different ( $\alpha = 0.05$ )

	Experimental groups				
Parameter	20	40	80	120	
Initial mean body length (mm)	$7.6 \pm 0.1^{a}$	$7.6 \pm 0.1^{a}$	$7.6 \pm 0.1^{a}$	7.6±0.1 <sup>a</sup>	
Initial mean body weight (mg)	$2.2 \pm 0.1^{a}$	$2.2 \pm 0.1^{a}$	$2.2 \pm 0.1^{a}$	$2.2 \pm 0.1^{a}$	
Final mean body length (mm)	$26.8 \pm 0.9^{a}$	$24.6 \pm 0.7^{a}$	$22.4{\pm}0.9^b$	$20.1\!\pm\!1.2^{b}$	
Final mean body weight (mg)	$69.4 \pm 3.2^{a}$	$64.2 \pm 3.5^{a}$	$56.1 \pm 3.2^{b}$	$49.5 \pm 3.1^{\circ}$	
Initial stock (indiv.)	500	1000	1500	3000	
Final stock (indiv.)	$473.0 \pm 10.97$	$942.0 \pm 14.11$	$1397.0 \pm 37.0$	$2743.0 \pm 57.0$	
Survival (%)	$94.6 \pm 1.1^{a}$	$94.2 \pm 1.4^{a}$	$93.1 \pm 3.7^{a}$	$91.4 \pm 5.7^{a}$	
Increase in total length (ITL) (mm d <sup>-1</sup> )	$0.91{\pm}0.04^a$	$0.81{\pm}0.03^a$	$0.70 {\pm} 0.04^{ m b}$	$0.60 {\pm} 0.06^{\rm c}$	
Relative growth rate (RGR) for weight ( $\% d^{-1}$ )	$17.86 \pm 0.26^{a}$	$17.43 \pm 0.31^{a}$	$16.68 \pm 0.32^{b}$	$15.98 \pm 0.35^{c}$	
Relative growth rate (RGR) for length (% $d^{-1}$ )	$6.18 \pm 0.18^{a}$	$5.75 \pm 0.15^{a}$	$5.28 \pm 0.20^{b}$	$4.74 \pm 0.30^{b}$	
Relative growth rate (RBR) for biomass (% $d^{-1}$ )	$17.55 \pm 0.26^{a}$	$17.09 \pm 0.31^{b}$	$16.28 \pm 0.32^{c}$	$15.49 \pm 0.34^{d}$	
Biomass (g)	$1.31 {\pm} 0.06^{d}$	$2.42 \pm 0.13^{c}$	$3.13 \pm 0.18^{b}$	$5.43 \pm 0.34^{a}$	
Biomass (g dm <sup>-3</sup> )	32.83±1.51 <sup>d</sup>	$60.48 \pm 3.30^{\circ}$	$78.37 \pm 4.47^{b}$	$135.78 \pm 8.50^{a}$	

lengths and weights among the fish from different experimental groups. Survival percentages were normalized (angular transformation), and differences were significant at  $\alpha$  = 0.05 (Sokal and Rohl 1981). The results were processed statistically in Excel 16.0 (Microsoft, Redmond, WA, USA) and Statistica 13.0 for Windows (StatSoft, Inc., Tulsa, OH, USA).

#### Results

The smallest larvae obtained were reared at a stocking density of 120 indiv. dm<sup>-3</sup>. On the last day of rearing, their average weight was 49.5  $\pm$  3.1 mg at an average length of 20.1  $\pm$  1.2 mm. At a stocking density of 80 indiv. dm<sup>-3</sup>, the average weight was 56.1  $\pm$ 3.2 mg at an average length of 22.4  $\pm$  0.9 mm. At a stocking density of 40 indiv. dm<sup>-3</sup>, the average weight of the larvae was 64.2  $\pm$  3.5 mg at an average length of 24.6  $\pm$  0.73 mm. The average weight of the largest larvae was 69.4  $\pm$  3.2 mg at an average length of 26.8  $\pm$  0.94 mm at a stocking density of 20 indiv. dm<sup>-3</sup> (Figs. 1-2).

The highest larval blue bream biomass of 135.78g (8.5 g dm<sup>-3</sup>) was obtained at the stocking density of 120 indiv. dm<sup>-3</sup>. The lowest larval biomass

was obtained at the stocking density of 20 indiv. dm<sup>-3</sup> at just 32.83 g (1.51 g dm<sup>-3</sup>). Table 1 presents the results of other breeding indicators studied such as RGR and ITL (Table 1).

During the 21-day rearing period, the lowest mortality was observed at the stocking density of 20 indiv. dm<sup>-3</sup> (Fig. 3). Nearly 94.6% of the individuals from the initial stock survived the experiment. The survival percentages at the stocking densities of 40 and 80 indiv. dm<sup>-3</sup> were slightly worse, but statistically insignificant, at 94.2 and 93.1%, respectively. The lowest larval survival rate was observed at the stocking density of 120 indiv. dm<sup>-3</sup> with 91.4% of larvae from the initial stock surviving (Figure 3). The larvae reared at a water temperature of 15°C were the smallest (Figs. 4-5).

On the last day of rearing, the average weight of larvae from the water temperature of  $15^{\circ}$ C was  $34.2 \pm 3.1$  mg at an average length of  $16.8 \pm 1.14$  mm. At a water of temperature  $20^{\circ}$ C, the average weight was  $51.3 \pm 0.1$  mg at an average length of  $21.1 \pm 0.73$  mm. At the water temperature of  $25^{\circ}$ C, the average larval weight was  $65.8 \pm 0.1$  mg at an average length of  $25.3 \pm 0.95$  mm. The highest average larval weight was  $80.1 \pm 0.1$  mg at an average length of  $28.4 \pm 1.4$ 



Figure 1. Increases in total lengths of larval blue bream (B. ballerus) reared at stocking densities of 20, 40, 80, and 120 indiv. dm<sup>-3</sup>.



Figure 2. Increases in the body weights of larval blue bream (*B. ballerus*) reared at stocking densities of 20, 40, 80, and 120 indiv. dm<sup>-3</sup>.

## Table 2

Results of rearing larval blue bream (*B. ballerus*) at water temperatures of 15, 20, 25, and 30°C  $\pm$  0.2°C. Mean value  $\pm$  SD. Results in rows with the same letter index are not statistically significantly different ( $\alpha = 0.05$ )

	Experimental groups				
Parameter	15	20	25	30	
Initial mean body length (mm)	$7.6 \pm 0.1^{a}$	$7.6 \pm 0.1^{a}$	7.6±0.1 <sup>a</sup>	7.6±0.1 <sup>a</sup>	
Initial mean body weight (mg)	$2.2 \pm 0.1^{a}$	$2.2 \pm 0.1^{a}$	$2.2 \pm 0.1^{a}$	$2.2 \pm 0.1^{a}$	
Final mean body length (mm)	$16.8 \pm 1.14^{d}$	$21.1 \pm 0.73^{c}$	$25.3 \pm 0.95^{b}$	$28.4{\pm}1.4^{a}$	
Final mean body weight (mg)	$34.2 \pm 3.1^{d}$	$51.3 \pm 0.1^{c}$	$65.8\pm0.1^{\mathrm{b}}$	$80.1 \pm 0.1^{a}$	
Initial stock (indiv.)	1000	1000	1000	1000	
Final stock (indiv.)	$976.0 \pm 5.6$	$949.0 \pm 7.5$	$954.0 \pm 9.5$	$952.0 {\pm} 4.6$	
Survival (%)	$97.6 \pm 0.56^{a}$	$94.9 {\pm} 0.75^{ m b}$	$95.4 {\pm} 0.95^{ m b}$	$95.2 \pm 0.46^{b}$	
Increase in total length (ITL) (mm d <sup>-1</sup> )	$0.44{\pm}0.05^{d}$	$0.64 {\pm} 0.03^{c}$	$0.84{\pm}0.05^b$	$0.99 {\pm} 0.07^{\mathrm{a}}$	
Relative growth rate (RGR) for weight (% $d^{-1}$ )	$13.96 {\pm} 0.49^{d}$	$16.18 \pm 0.38^{c}$	$17.56 \pm 0.27^{b}$	$18.67 {\pm} 0.24^{a}$	
Relative growth rate (RGR) for length (% $d^{-1}$ )	$3.85 {\pm} 0.34^{d}$	$4.98{\pm}0.17^{\rm c}$	$5.89{\pm}0.19^b$	$6.48 {\pm} 0.25^{a}$	
Relative growth rate (RBR) for biomass (% $d^{-1}$ )	$13.83 \pm 0.49^{d}$	$15.89 \pm 0.38^{c}$	$17.30 \pm 0.27^{b}$	$18.39 {\pm} 0.24^{a}$	
Biomass (g)	$1.34 \pm 0.12^{d}$	$1.95 \pm 0.13^{c}$	$2.51{\pm}0.12^b$	$3.05 {\pm} 0.13^{a}$	
Biomass (g dm <sup>-3</sup> )	33.38±3.03 <sup>d</sup>	48.68±3.32 <sup>c</sup>	62.77±3.05 <sup>b</sup>	76.26±3.24 <sup>a</sup>	

mm at a water temperature of 30°C. The highest larval blue bream biomass of 76.26 (3.24 g dm<sup>-3</sup>) was obtained at a water temperature of 30°C, while the lowest of just 33.38 g (3.03 g dm<sup>-3</sup>) was obtained at a water temperature of 15°C (Table 2).

The lowest mortality observed was at a water temperature of 15°C during the 21-day rearing period (Figure 6), and almost 97.6% of the individuals from the initial stock survived. Slightly lower, but statistically insignificant, survival rates were observed at



Figure 3. Cumulative mortality of larval blue bream (B. ballerus) reared at stocking densities of 20, 40, 80, and 120 indiv. dm<sup>-3</sup>.



Figure 4. Increases in total lengths of larval blue bream (*B. ballerus*) reared in water temperatures of 15, 20, 25, and  $30 \pm 0.2^{\circ}$ C.



Figure 5. Increase in total weight of larval blue bream (*B. ballerus*) reared in water temperatures of 15, 20, 25, and  $30 \pm 0.2^{\circ}$ C.



Figure 6. Cumulative mortality of larval blue bream (B. ballerus) reared in water temperatures of 15, 20, 25, and 30 ± 0.2°C.

water temperatures of 25 and 30°C at 95.4 and 95.2%, respectively. The lowest larval survival rate was observed at a water temperature of 20°C, in which 94.9% of the larvae from the initial stock survived (Table 2).

## Discussion

The main role of aquaculture is to produce fish and aquatic invertebrates as sources of valuable, healthy food. Additionally, it is a conservation tool for species and specific populations. Consequently, breeding and rearing technologies have been or are being developed not only for fish species that are, or potentially could become, valuable raw materials for food production, but also for species the abundance of which in open waters is declining to the extent that they are threatened with extinction (Kujawa et al. 2019, Lepić et al. 2020, Kujawa and Piech 2021, Nowosad et al. 2021b, Szmyt et al. 2021). Researchers in this field are also interested in fish species that are of no economic importance, but are valuable components of aquatic ecosystems and their biodiversity. This was the case for several species of rheophilous cyprinid fishes that are of no economic importance and/or do not have a high conservation status, including: ide, Leuciscus idus (L.) (Cejko et al. 2010, Kupren et al. 2011, Targońska et al. 2011a, 2012, Kucharczyk et al. 2020); chub, Squalius cephalus (L.) (Krejszeff et al. 2008, Krejszeff et al. 2010, Kupren et al. 2011); common dace, Leuciscus leuciscus (L.) (Cejko et al, 2012, Nowosad et al. 2014, Targońska et al. 2015, Kucharczyk et al. 2019); barbel, Barbus barbus (L.) (Targońska et al. 2011b, Żarski et al. 2011, Nowosad et al. 2016, 2020, 2021, Prusińska et al. 2020); rudd, Scardinius erythrophthalmus (L.) (Kucharczyk et al. 1997a); and asp, *Leuciscus aspius* (L.) (Kujawa et al. 1997, Targońska et al. 2010).

For many fish species, variation in individual growth can lead to changes in population productivity since size affects survival, fecundity, and reproduction. Variation in growth results from a number of aspects that are dependent and independent of stocking density (Matthias et al. 2018). This is important for intensive rearing under controlled conditions (Karakatsouli et al. 2007, 2015), and particularly so for larvae. Aste et al. (2009) investigated the effects larval African catfish (Clarias gariepinus on (Burchell)) stocking density in RAS on rearing parameters ranging from 1 to 25 individuals per l, and they report that the optimum density level was 15 larvae per l. Santos et al. (2020) reared larval cachama (Colossoma macropomum (Cuvier)) in a RAS and analyzed the effect of initial stocking densities of 10, 30, and 50 indiv. dm<sup>-3</sup> on rearing parameters. They found it was possible to rear this species successfully at the highest stocking density. Szkudlarek and Zakeś (2007) analyzed the effects of stocking density on larval pikeperch, Sander lucioperca (L.), growth and survival and found that increasing maximum larval density (100 indiv. dm<sup>-3</sup>, range 25-100) between 4 and 18 DPH had a favorable effect on survival. In further larval pikeperch rearing at densities of 6 to 15 indiv. dm<sup>-3</sup>, the highest survival and growth rates were obtained at the highest density. However, the proper density depends on the species, initial larval size, feeding regime, food type, and water temperature. One example is the mass rearing of larval burbot (Lota lota (L.)) at a density of 1,000 indiv. dm<sup>-3</sup> (Żarski et al. 2009).

In the present study on larval blue bream, the larvae grew fastest at the lowest density (20 indiv. dm<sup>-3</sup>). In this group, they had the highest, statistically significant RGR values for weight and length (%-d<sup>-1</sup>) and the highest mean length and weight at the end of rearing. Similar observations were recorded for asp, chub, and ide, which also had the highest weights and total lengths at the lowest stocking densities tested (Żarski et al. 2008, Kupren et al. 2011). Statistically significant differences in the mean weights of larvae from different test groups appeared on 17 d of rearing, while differences in length were noted slightly earlier on 13 d of rearing. However, Kupren et al. (2011) reported differences in larval lengths in the first (asp) or third (ide, chub) weeks of rearing. The survival rate of larval blue bream after the end of rearing in all density variants was high and ranged from 91.4 to 94.6%, and no statistical differences were found among the experimental groups. Kupren et al. (2011) reported similar results and found that stocking density had no significant effect on larval asp, chub, or ide mortality, and they demonstrated that the larvae of all three species could be reared at densities of up to 400 indiv. dm<sup>-3</sup> with a slight reduction in growth rates and no negative effects on survival or development.

In addition to determining the correct stocking density and food for larval fish, determining optimum rearing temperature is important. This temperature should be one at which the larvae have a high survival rate while they simultaneously grow to similar individual sizes. Research continues to be conducted on determining the ranges of optimum temperatures for embryonic and larval development in fishes. This is because it is one of the most important abiotic factors influencing larval growth rates, growth, and survival during intensive rearing under controlled conditions. Studies conducted on cyprinid fish such as asp, sichel (Pelecus cultratus (L.)), and common bream (Abramis brama (L.) have shown that, in many cases, the optimal temperature range for their rearing under controlled conditions is between 25°C and sometimes even 30°C (Kucharczyk et al. 1997a, 1998, Kujawa et al. 1997, 2015). These temperatures are often significantly higher than those occurring during embryonic and larval development in the wild (Kujawa et al. 1997, Kupren et al. 2011). Learning about the aspects of temperature effects on development, survival, and the possibility of developmental deformities is extremely important for rearing efficiency and the quality of stocking material (e.g., Kupren et al. 2010, 2011, Łączyńska et al. 2016, Nowosad et al. 2021b).

In the present study, as for most of cyprinid fishes studied, temperatures in the range of 25–30 °C can be considered optimal. These are very similar to

the results obtained during the rearing of larval sichel (Kujawa et al. 2015) and common bream (Kucharczyk et al. 1997b, 1998) and marginally higher, although they overlap, with those for asp (Kujawa et al. 1997), ide, dace, and chub (Kupren et al. 2010, 2011). For other freshwater fish species that often live in the same area as blue bream, such as European whitefish, the optimum temperatures for larval development are much lower in the 15-19°C range (Sebesta et al. 2018). However, a similar pattern is also observed here in that the optimum temperatures for larvae reared under controlled conditions are higher than those found in the wild. Regardless of the species studied, this indicates that larvae exhibit extremely high plasticity with regard to thermal conditions. It is clear, however, that these conditions cannot be compared indiscriminately. Optimal thermal conditions are generally applied in RAS during fish reproduction and embryonic and larval development (e.g., for ide and dace 12-14°C - Targońska et al. 2011, Cejko et al. 2012, Nowosad et al. 2014, Kucharczyk et al. 2019, 2020). However, temperature fluctuations do occur in the wild, especially in lotic waters and in lake shallows, which are, after all, the usual spawning grounds of these fishes and the rearing sites of their juveniles. Studies conducted under controlled conditions and in the natural environment indicate that large temperature fluctuations, even within the range of optimum temperatures, can disrupt spawning, reduce fecundity and gamete quality, cause high embryo and larval mortality, and the occurrence of developmental deformities (Kupren et al. 2010, 2011, Nowosad et al. 2014, Targońska et al. 2014). However, this provides very important information for producers of stocking material; namely, that while larval rearing temperatures do not have to hit the optimal point precisely, they should be fairly constant.

The present study confirmed that larval blue bream are highly tolerant of high stocking densities and high temperatures that are optimal for rheophilic fish. The best larval blue bream rearing results and the lowest mortality rates were obtained when this species was reared at the lowest stocking density of 20 indiv. dm<sup>-3</sup>. The highest temperature tested

(30°C) was optimal and produced the highest larval blue bream survival and growth rates. For large-scale blue bream stocking material production, it is recommended to apply lower densities and a temperature of 25° to obtain optimal rearing results both biologically and to lower costs of heating water in the RAS.

Author contributions. P.P. and R.K. designed the study. P.P. conducted the field investigations and provided environmental data. R.K. managed the database and did some of the analyses. R.K. conducted the statistical analyses. P.P, R.K., J.N, D.K., K.T., and M.T. did some of the data analyses and drafted the manuscript. All authors contributed to writing the manuscript and approved the final version that was submitted.

#### ORCID iD

Przemysław Piech 🕑 https://orcid.org/0000-0003-0538-5758
Roman Kujawa (D) https://orcid.org/0000-0003-0925-1204
Joanna Nowosad (D) https://orcid.org/0000-0001-9491-0141
Dariusz Kucharczyk i https://orcid.org/0000-0002-0889-0656
Katarzyna Targońska D https://orcid.org/0000-0002-6273-2587
Mariusz Szmyt D https://orcid.org/0000-0001-8799-9088

## References

- Atse, B.C., Konan, K.J., Alla, Y.L., Pangini, K. (2009). Effect of rearing density and feeding regimes on growth and survival of African catfish, *Heterobranchus longifilis* (Valenceinnes, 1840) Larvae in a closed recirculating aquaculture system. Journal of Applied Aquaculture, 21, 183–195.
- Brown, M. E. (1957). Experimental studies on growth. In: The physiology of fishes. (Ed.) M.E. Brown, Academic Press, New York, 361–400.
- Cejko, B. I., Kowalski, R. K., Kucharczyk, D., Targońska, K., Krejszeff, S., Żarski, D., Glogowski, J. (2010). Influence of the length of time after hormonal stimulation on selected parameters of milt of ide *Leuciscus idus* L. Aquaculture Research, 41, 804–813.
- Cejko, B. I., Targońska, K., Kowalski, R. K., Żarski, D., Sarosiek, B., Kucharczyk, D., Glogowski, J. (2012). The effectiveness of hormonal preparations (Ovopel, Ovaprim, LHRH a, hCG and CPE) in stimulating spermiation in dace *Leuciscus leuciscus* (L.). Journal of Applied Ichthyology, 28, 873–877.
- Duncan, D. B. (1955). Multiple range and multiple F-test. Biometrics, 11, 1-42.

- Goryczko, K., Witkowski, A. (2009). Stocking management and environmental protection. Chrońmy Przyrodę Ojczystą, 65, 93–98 (in Polish).
- Gorzel, M., Kornijów, R. (2004). Biological methods for assessing river water quality. Kosmos, 53, 183–191 (in Polish).
- Hakuć-Błażowska, A., Kupren, K., Turkowski, K., Targońska, K., Żarski, D., Kucharczyk, D. (2010). A comparison of the economic effectiveness of various spawning agents for stimulating the reproduction of the cultured and wild forms of the common barbel *Barbus barbus* (L.). Polish Journal of Natural Sciences, 25, 272–286.
- Heese, T., Witkowski, A. (1996). Conservation of rare and threatened fish species in Poland: Current state and perspectives. Zoologica Poloniae, 41(Supl.), 5–6 (in Polish).
- Imanpoor, M. R., Najafi, E., Kabir, M. (2012). Effects of different salinity and temperatures on the growth, survival, haematocrit and blood biochemistry of Goldfish (*Carassius auratus*). Aquaculture Research, 43, 332–338.
- Karakatsouli, N., Papoutsoglou, S. E., Manolessos, G. (2007). Combined effects of rearing density and tank colour on the growth and welfare of juvenile white sea bream *Diplodus sargus* L. in a recirculating water system. Aquaculture Research, 38, 1152–1160.
- Karakatsouli, N., Kassianos, N., Papoutsoglou, S.E. (2015). Effects of rearing density and tank colour on juvenile sharpsnout seabream (*Diplodus puntazzo*) growth performance. Aquaculture International, 23, 943–953.
- Krejszeff, S., Kucharczyk, D., Kupren, K., Targońska, K., Mamcarz, A., Kujawa, R., Kaczkowski, Z., Ratajski, S. (2008). Reproduction of chub, *Leuciscus cephalus* L., under controlled conditions. Aquaculture Research, 39, 907–912.
- Krejszeff, S., Targońska, K., Żarski, D., Kucharczyk, D. (2010). Artificial reproduction of two different spawn-forms of the chub. Reproductive Biology, 10, 67–74.
- Kucharczyk, D., Nowosad, J., Kucharczyk, D. J., Kupren, K., Targońska, K., Wyszomirska, E., Kujawa, R. (2019). Out-of-season artificial reproduction of common dace (*Leuciscus leuciscus* L.) under controlled conditions. Animal Reproduction Science, 202, 21–25.
- Kucharczyk, D., Nowosad, J., Wyszomirska, E., Cejko, B. I., Arciuch-Rutkowska, M., Juchno, D., Boroń, A. (2020). Comparison of artificial spawning effectiveness of hCG, CPH and GnRHa in combination with dopamine inhibitors in a wild strain of ide *Leuciscus idus* (L.) in hatchery conditions. Animal Reproduction Science, 221, 106543.
- Kucharczyk, D., Kujawa, R., Mamcarz, A., Wyszomirska, E. (1997a). Induced spawning in rudd (*Scardinius erythtrophthalmus* L.). Polish Archives of Hydrobiology, 44, 209–213.
- Kucharczyk, D., Luczynski, M., Kujawa, R., Czerkies, P. (1997b). Effect of temperature on embryonic and larval

development of bream (*Abramis brama* L.). Aquatic Sciences, 59, 214–224.

- Kucharczyk, D., Luczynski, M., Kujawa, R., Kaminski, R., Ulikowski, D., Brzuzan, P. (1998). Influences of temperature and food on early development of bream (*Abramis brama* L.). Archiv für Hydrobiologie, 141, 243–256.
- Kucharczyk, D., Malinovskyi, O., Nowosad, J., Kowalska, A., Cejko, B. I. (2021). Comparison of responses to artificial spawning of ruffe (*Gymnocephalus cernua*) specimens captured from their natural habitat to those produced in cultured conditions. Animal Reproduction Science, 225, 106684.
- Kujawa, R., Mamcarz, A., Kucharczyk, D. (1997). Effect of temperature on embryonic development of asp (*Aspius aspius* L.). Polish Archives of Hydrobiology, 44, 139–143.
- Kujawa, R. (2004). Biological foundations for rearing rheophilic cyprinid species under controlled conditions – Rozprawy i monografie 88, Wyd. UWM, Olsztyn, 88 p. (in Polish).
- Kujawa, R., Furgała-Selezniow, G., Mamcarz, A., Lach, M., Kucharczyk, D. (2015). Influence of temperature on the growth and survivability of sichel larvae *Pelecus cultratus* reared under controlled conditions. Ichthyological Research, 62, 163–170.
- Kujawa, R., Cejko, B. I., Fopp-Bayat, D., Judycka, S., Glińska-Lewczuk, K., Timofte, C. M., Nowosad, J., Kucharczyk, D. (2019). Reproduction of endangered river lamprey (*Lampetra fluviatilis*) in controlled conditions. Animal reproduction science, 203, 75–83.
- Kujawa, R., Piech, P. (2021). Rearing of Bitterling (*Rhodeus amarus*) Larvae and Fry under Controlled Conditions for the Restitution of Endangered Populations. Animals, 11, 3534.
- Kupren, K., Mamcarz, A., Kucharczyk, D. (2010). Effects of temperature on survival, deformations rate and selected parameters of newly hatched larvae of three rheophilic cyprinids (genus *Leuciscus*). Polish Journal of Natural Sciences, 25, 299–312.
- Kupren, K., Żarski, D., Krejszeff, S., Kucharczyk, D. Targońska, K. (2011). Effect of stocking density on growth, survival and development of asp *Aspius aspius* (L.), ide *Leuciscus idus* (L.) and chub *Leuciscus cephalus* (L.) larvae during initial rearing under laboratory conditions. Italian Journal of Animal Science, 10, e34.
- Lepič, P., Blecha, M., Kozák, P. (2020). Intensive winter culture of *Chondrostoma nasus* (Linnaeus, 1758) and *Vimba vimba* (Linnaeus, 1758) for spring restocking. Turkish Journal of Fisheries and Aquatic Sciences, 20, 97–102.
- Levin, B. A., Levina, M. A. (2014). Poly- and oligomerization of scales in the blue bream *Ballerus ballerus (Cyprinidae*) as a consequence of thyroid status regulation. Journal of Applied Ichthyology, 30, 809–813.

- Łączyńska, B., Palińska-Żarska, K., Nowosad, J., Biłas, M., Krejszeff, S., Müller, T., Kucharczyk, D., Żarski, D. (2016). Effect of age, size and digestive tract development on weaning effectiveness in crucian carp, *Carassius carassius* (Linnaeus, 1758). Journal of Applied Ichthyology, 32, 866–872.
- Matthias, B. G., Ahrens, R. N. M., Allen M. S., Tuten, T., Siders, Z. A., Wilson, K. L. (2018). Understanding the effects of density and environmental variability on the process of fish growth. Fisheries Research, 198, 209–219.
- Myszkowski, L. (1997). Pitfalls of using growth rate coefficients. Polskie Archiwum Hydrobiologii, 44, 389–396,
- Nowosad, J., Żarski, D., Biłas, M., Dryl, K., Krejszeff, S., Kucharczyk, D. (2013). Dynamics of ammonia excretion in juvenile common tench, *Tinca tinca* (L.), during intensive rearing under controlled conditions. Aquaculture International, 21, 629–637.
- Nowosad, J., Targońska, K., Chwaluczyk, R., Kaszubowski, R., Kucharczyk, D. (2014). Effect of temperature on the effectiveness of artificial reproduction of dace [*Cyprinidae* (*Leuciscus leuciscus* (L.))] under laboratory and field conditions. Journal of Thermal Biology, 45, 62–68.
- Nowosad, J., Kucharczyk, D., Targońska, K., Wyszomirska, E., Chwaluczyk, R., Kupren, K. (2016). The Synergistic Effect of Temperature and Hormonal Stimulation on Spawning Efficiency of Common Barbel, *Barbus barbus* L. Turkish Journal of Fisheries and Aquatic Sciences, 16, 517–524.
- Nowosad, J., Dryl, K., Kupren, K., Kucharczyk, D. (2020). Inhibiting the influence of ovarian fluid on spermatozoa activation and spermatozoa kinetic characteristics in the common barbel *Barbus barbus*. Theriogenology, 158, 250–257.
- Nowosad, J., Kupren, K., Biegaj, M., Kucharczyk, D. (2021a). Allometric and ontogenetic larval development of common barbel during rearing under optimal conditions. Animal, 15, 100107.
- Nowosad, J., Kucharczyk, D., Sikora, M., Kupren, K. (2021b). Optimization of barbel (*Barbus barbus* L.) fertilization and effects of ovarian fluid when there are controlled conditions for gamete activations. Animal Reproduction Science, 224, 106652.
- Peňáz, M., Prokeš, M., Kouřil, J., Hamáčková, J. (1989). Influence of water temperature on the early development and growth of the tench, *Tinca tinca*. Folia Zoologica, 38, 275–287.
- Piech, P., Kujawa, R. (2021). Artificial reproduction of blue bream (*Ballerus ballerus* L.) as a conservative method under controlled conditions. Animals, 11, 1326.
- Piech, P., Kujawa, R. (2022). Impact of replacing natural food with commercial feed on the growth and survival of blue

bream larvae (*Ballerus ballerus*) under controlled conditions. Fisheries & Aquatic Life 30, 192–201.

- Piech, P., Kujawa, R. (2023). Effect of different water salinities on the larvae of the blue bream *Ballerus ballerus* (Linnaeus, 1758) during rearing. Animals, 13(7), 1245.
- Prusińska, M., Nowosad, J., Jarmołowicz, S., Mikiewicz, M., Duda, A., Wiszniewski, G., Sikora, M., Biegaj, M., Samselska, A., Arciuch-Rutkowska, M., Targońska, K., Otrocka-Domagała, I., Kucharczyk, D. (2020). Effect of feeding barbel larvae (*Barbus barbus* (L, 1758)) *Artemia* sp. nauplii enriched with PUFAs on their growth and survival rate, blood composition, alimentary tract histological structure and body chemical composition. Aquaculture Reports, 18, 100492.
- Radwan, S., Mieczan, T., Plaska, W., Wojciechowska, W., Sender, J., Jaszczenko, P. (2002). The aquatic ecosystems of the Polesia region: Current state and directions of changes.. Acta Agrophysica, 66, 89–120 (in Polish).
- Santos, E. L. R., Rezende, F. P., Moron, S. E. (2020). Stress-related physiological and histological responses of tambaqui (*Colossoma macropomum*) to transportation in water with tea tree and clove essential oil anesthetics. Aquaculture, 523, 735164.
- Sebesta, R., Kucharczyk, D., Nowosad, J., Sikora, M., Stejskal, V. (2018). Effect of temperature on growth and survival of maraena whitefish *Coregonus maraena* (Bloch 1779) larvae in controlled conditions. Aquaculture Research, 49, 3151–3157.
- Sokal, R. R., Rohl, F. J. (1981). Biometry: The Principles and Practice of Statistics in Biological Research, ed. 2. San Francisco. W. H. Freeman, 733.
- Sorgeloos, P., Bossuyt, E., Lavińa, E., Baeza-Mesa, M., Persoone, G. (1977). Decapsulation of Artemia cysts: a simple technique for the improvement of the use of brine shrimp in aquaculture. Aquaculture, 12, 311–315.
- Stejskal, V., Matousek, J., Sebesta, R., Nowosad, J., Sikora, M., Kucharczyk, D. (2021a). Stocking density effect on survival and growth of early life stages of maraena whitefish, *Coregonus maraena* (Actinopterygii: Salmoniformes: *Salmonidae*). Acta Ichthyologica et Piscatoria, 51, 139–144.
- Stejskal, V., Gebauer, T., Sebesta, R., Nowosad, J., Sikora, M., Biegaj, M., Kucharczyk, D. (2021b). Effect of feeding strategy on survival, growth, intestine development, and liver status of maraena whitefish *Coregonus maraena* larvae. Journal of the World Aquaculture Society, 52, 829–842.
- Szkudlarek, M., Zakęś, Z. (2007). Effect of stocking density on survival and growth performance of pikeperch, *Sander lucioperca* (L.), larvae under controlled conditions. Aquaculture International, 15, 67–81.
- Szmyt, M., Piech, P., Panasiewicz, G., Niewiadomski, P., Pietrzak-Fiećko, R. (2021). Gonadal steroid fluctuations and reproduction results of European grayling

(*Thymallus thymallus* L.) from broodstock farmed in a recirculation aquaculture system. Animal Reproduction Science, 224, 106648.

- Śliwiński, J., Ostaszewska, T., Kamaszewska, M. (2016). Aquaculture as a tool for improving the status of rheophilous cyprinids in Poland: State of and perspectives for research.. Przegląd Naukowy Inżynieria i Kształtowanie Środowiska, 25, 239–248 (in Polish).
- Tadajewska M. (2000). Blue bream, Abramis ballerus. In: The Freshwater Fishes of Poland (Ed.) M. Brylińska. Wydawnictwo Naukowe PWN, Warszawa: 257–261 (in Polish).
- Targońska, K., Kucharczyk, D., Kujawa, R., Mamcarz, A., Żarski, D. (2010). Controlled reproduction of asp, *Aspius aspius* (L.) using luteinizing hormone releasing hormone (LHRH) analogues with dopamine inhibitors. Aquaculture, 306, 407–410.
- Targońska, K., Kucharczyk, D., Żarski, D., Cejko, B., Krejszeff, S., Kupren, K., Król, R., Dryl, K., Kowalski, K., Glogowski, J. (2011b). Artificial reproduction of wild and cultured barbel (*Barbus barbus, Cyprinidae*) under controlled conditions. Acta Veterinaria Hungarica, 59, 363–372.
- Targońska, K., Kupren, K., Żarski, D., Król, R., Kucharczyk, D. (2011a). Influence of thermal conditions on successful ide (*Leuciscus idus* L.) artificial reproduction during spawning season. Italian Journal of Animal Science, 10(4), e50.
- Targońska, K., Żarski, D., Krejszeff S., Kucharczyk, D. (2012). Influence of age of wild ide *Leuciscus idus* (L.) female on spawning effectiveness under controlled conditions. Italian Journal of Animal Science, 11, 342–346.
- Targońska, K., Kupren, K., Palińska-Żarska, K., Mamcarz, A., Kujawa, R., Skrzypczak, A., Furgała-Selezniow, G., Czarkowski, T.K., Hakuć-Błażowska, A., Kucharczyk, D.

(2014). Influence of temperature during four following spawning seasons on the spawning effectiveness of common bream, *Abramis brama* (L.) under natural and controlled conditions. Journal of Thermal Biology, 39, 17–23.

- Targońska, K., Kupren, K., Kujawa, R., Mamcarz, A., Kaczkowski, Z., Glogowski, J., Kowalski, R.K., Żarski, D., Wyszomirska, E., Kucharczyk, D. (2015). Artificial reproduction of different dace, *Leuciscus leuciscus* (L.) populations as a method for biodiversity preservation. Turkish Journal of Fisheries and Aquatic Sciences, 15, 477–485.
- Zakęś, Z., Wunderlich, K., Szczepkowski, M., Szczepkowska, B. (2016). Rearing quality of of lake whitefish stocking material produced in aquaculture facilities with recirculating aquaculture system technology. Management of fish farms in 2016: Economic, legal, and ecological conditions, 155 (in Polish).
- Żarski, D., Kucharczyk, D., Kwiatkowski, M., Targońska, K., Kupren, K., Krejszeff, S., Jamróz, M., Hakuć-Błażowska, A., Kujawa, R., Mamcarz, A. (2008). The effect of stocking density on the growth and survival of larval asp, *Aspius aspius* (L.), and European chub, *Leuciscus cephalus* (L.), during rearing under controlled conditions. Fisheries & Aquatic Life, 16, 371–381.
- Żarski, D., Sasinowski, W., Kucharczyk, D., Kwiatkowski, M., Krejszeff, S., Targońska, K. (2009). Mass initial rearing of burbot *Lota lota* (L.) larvae under controlled conditions. Polish Journal of Natural Sciences 24, 76–84.
- Żarski, D., Kupren, K., Targońska, K., Krejszeff, S., Furgała-Selezniow, G., Kucharczyk, D. (2011). The effect of initial larval stocking density on growth and survival in common barbel *Barbus barbus* (L.). Journal of Applied Ichthyology, 27, 1155–1158.