



## Sexual Cycle of the Blue Bream (*Ballerus ballerus*) from the Lower Oder River and Dąbie Lake (NW Poland)

Józef Domagała<sup>1</sup>, Katarzyna Dziewulska<sup>1</sup>, Lucyna Kirczuk<sup>1,\*</sup>, Małgorzata Pilecka-Rapacz<sup>1</sup>

<sup>1</sup> Department of General Zoology, University of Szczecin, Felczaka 3c street, 71-415 Szczecin, Poland.

\* Corresponding Author: Tel.: +48.91 4441618; Fax: +48.91 4441623;  
E-mail: lucyna.kirczuk@univ.szczecin.pl

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### Abstract

Histological studies of the blue bream (*Ballerus ballerus* L., 1758) (Actinopterygii, Cypriniformes, Cyprinidae) gonad maturation have not been conducted to date. The aim of this study was to analyse the annual gonad development cycle of the blue bream from the Oder River and Lake Dąbie in north-western Poland.

The spawning period of the blue bream, a single-portion spawner, was short, lasted approximately two weeks and occurred in the first half of April in Lake Dąbie, and in the second half of April in the Oder River. The blue bream male gonads reached maturity stage IV before the winter and remained at this stage until early spring. The gonads contained cells at all spermatogenic stages and the tubule lumen was filled with numerous spermatozoa. The males from Lake Dąbie finalized their spermatogenesis in April slightly faster than those from the Oder River. Some individuals had gonad filled with large amounts of spermatozoa even in May and June.

**Keywords:** Blue bream, *Ballerus ballerus*, reproductive cycle, gonad.

### Introduction

The blue bream (zope) is a species occurring in the waters of central and eastern Europe. It is most common in the waters of south-eastern Europe. The species inhabits lower parts of rivers, some of the European lakes and even brackish marine bays. It prefers calm sites in rivers, as well as slow-moving or still water (Tadajewska, 2000; Kottelat and Freyhof, 2007).

Few studies of the blue bream biology are available and regard nutrition, condition, chemical composition of the meat (Kompowski, 1971a,b; Treer *et al.*, 2009; Živković *et al.*, 2013), morphology (Krzykowski *et al.*, 1996; Szlachciak, 2005), and parasites (Wierzbička, 1977, 1998; Pietrock *et al.*, 1999; Pietrock and Scholz, 2000), as well as hybrids (Sly'anko and Sly'anko, 2010) in the population from the Danube River (Živković *et al.*, 2013), the lower part of the Oder River, Lake Dąbie and Szczecin Lagoon (Karabanowicz and Kompowski, 1994; Krzykowski *et al.*, 1996; Kompowski and Błaszczuk, 1997; Pietrock *et al.*, 1999; Pietrock and Scholtz, 2000; Szlachciak 2005), a region of Croatia (Treer *et al.*, 2009), Rybinski Dam Reservoir and Kuibyshev Reservoir on the Volga River (Chašem 1969;

Strelnikov *et al.*, 1983; Kuznetsov, 1990), and Lake Vedlozero in Karelia, Russia (Aleksandrova 1966).

The blue bream is rarely found in Poland. The largest populations have been observed since the mid-1960s in the north-western part of the country, Szczecin Lagoon, Lake Dąbie and the lower Oder River (Kompowski, 1971a,b, 1991). The blue bream becomes sexually mature at the age of 3–5 years, with a minimum standard length of 24.1 cm in female and 20.6 cm in male (Karabanowicz and Kompowski, 1994; Kompowski and Błaszczuk, 1997). The reproduction of the blue bream takes place at a water temperature above 12°C (Kompowski 1991). The females lay eggs once in a season. In Poland, spawning takes place in the spring, from the first decade of April to early May (Kompowski and Błaszczuk, 1997), and in the case of exceptionally low temperatures it may occur in the second half of May (Karabanowicz and Kompowski, 1994).

To date, the annual reproductive cycle of the blue bream has only been described on the basis of gonad morphology and GSI value fluctuations (Karabinowicz and Kompowski, 1994). No reports of histological studies of the blue bream gonad maturation can be found in the literature. The aim of this study was to analyse the annual gonad development cycle of the blue bream of both sexes

inhibiting the lower Oder River and Lake Dąbie in north-western Poland.

## Material and Methods

### Study Areas and Fish Sampling

The investigated material was selected from the adult males and females of blue bream (*Ballerus ballerus* L., 1758) (Actinopterygii, Cypriniformes, Cyprinidae) from the populations inhabiting (1) the East Oder River in the lower section; (2) Lake Dąbie, southern part of the flow-through lake (Figure 1). The fish were caught as bycatch of commercial fishing by gillnet between September 2009 and August 2010, from one to four times a month at each site. Attempts to catch the blue bream in the Warm Canal were conducted. Into the canal, the post-cooling water of the “Dolna Odra” power plant (53°11'N, 14°29'E) is discharged. The effective water temperature is 6–8°C higher than that of the neighboring Oder River (Domagała et al. 2015a). The Warm Canal is open and fish can freely move in or out of it into the Oder River. Only 8 females and 15 males of the blue bream were collected in the area, as the species avoids thermally polluted environment.

The age was determined based on the analysis of rings on the collected scales Kompowski (1971b, 1991). In the laboratory, the total length (TL) and standard length (ST) of the fish were measured to the accuracy of 0.1 cm. The fish were weighed on an electronic scale to the nearest 0.1 g. Subsequently, the gonads were prepared and weighed to the nearest 0.1 mg, and fixed in Bouin fluid. The Fulton Index ( $C_F$ ) and GSI were calculated as follows:

$$C_F = 100W_g \times TL^{-3} \text{ and}$$

$$GSI = 100W_g \times W_b^{-1}$$

where: TL is the total length of fish (cm),  $W_g$  is

the gonad weight (g),  $W_b$  is the total fish weight (g).

### Histological Analysis of Gonad

All gonads were included in the histological analysis. For the analysis, fragments of 0.5 cm in length were excised from the middle part of the gonad. Standard paraffin technique and Heidenhain's iron hematoxylin staining were used. Histological slides of 5  $\mu$ m in thickness were made. Between 50 and 100 slides were made from each female gonad, 25 from each male. The specimens were evaluated under a Nikon Eclipse 80i microscope with a maximum magnification of 1000 $\times$ .

Histological analysis was performed to determine the developmental stage of the blue bream gonads using the modified 6-grade scales proposed by Domagała et al. (2013, 2015b) (Table 1, Table 2).

### Statistical Analysis

The nonparametric Mann–Whitney U test or Kruskal–Wallis test was used to compare the following characteristics of the fish between the investigated sites: TL, SL, fish weight,  $C_F$ , GSI, sizes of oocytes at different stages. The significance of differences in the number of female and male was tested with the chi-square test. All analyses were performed at the significance level of 0.05 using the Statistica v.12 software (StatSoft, Inc.).

## Results

### Principal Somatic Parameters of Fish

As many as 128 females and 81 males were caught in the Oder River, while 129 females and 97 males were caught in Lake Dąbie (Table 3). The caught fish were adult, aged from 3+ to 11+.

The number of the blue bream males collected from the Oder River and Lake Dąbie was significantly



**Figure 1.** Map of Poland and sampling locations: Oder River from which water is supplied to the power plant “Dolna Odra” (PP), Warm Canal which is discharged cooling water (CCW) and Dąbie Lake.

**Table 1.** Maturity stages of female gonads in ruffe used in this study for blue bream (*Ballerus ballerus* L.)

Developmental stage	Period name	Description
I (cell sex differentiation)	Oogonial period	Stages of early prophase of meiosis, pre-meiotic transformation of nucleus
II	Protoplasmatic growth (pre-vitellogenesis)	Oocytes at different stages of protoplasmatic growth and oogonia making storage compounds for the next spawning
III	Trophoplasmatic growth (vitellogenesis)	Vacuolisation of cytoplasm; accumulation of yolk;
IV		Oocyte filled with yolk; beginning of oocyte polarisation
V	Maturation	Oocytes are liberated from the folliculi areola and dropped to body cavity; broken areola remain in the tissue of the ovary; free oocytes are present in the ovary; spawn is released from the sex opening
VI	Post-spawning ovary	Folliculi areola broken, small number of oocytes in Stage 4 stadium, oogonia and oocytes in protoplasmatic growth. After resorption of folliculi areola the ovary reaches Stage 2, or beginning of Stage 3 or even 4

Reprinted from Journal of Freshwater Ecology, 2013, Domagala, J., Kirczuk, L. and Pilecka-Rapacz, M. Annual development cycle of gonads of Eurasian ruffe (*Gymnocephalus cernuus* L.) females from lower Odra River sections differing in the influence of cooling water. 28: 423-437.

**Table 2.** The maturity scale of the blue bream (*Ballerus ballerus*) testis SG A - spermatogonia A; SG B- spermatogonia B; SC I- primary spermatocyte; SC II- secondary spermatocyte; SD - spermatid; SZ - spermatozoa

Cell	SG B	SC I	SC II	SD	SZ	SZ
Stage	A	B				in spermatic duct
I	+	-	-	-	-	-
II II <sub>E</sub>	+	+	-	-	-	[+]
II <sub>L</sub>	+	+	-	-	-	[+]
III III <sub>E</sub>	+	+	+	-	-	-
III <sub>L</sub>	+	+	+	+	+	-
IV	+	+	+	+	+	+
V	+	-	-	-	-	+
V-II	+	*	-	-	-	+
VI*	+	-	-	-	-	(+)
VI-II	+	*	-	-	-	(+)

II<sub>E</sub> - cysts containing less than 10 SG B

II<sub>L</sub> - cysts containing more than 10 SG B

[+] - spermatozoa left from the previous cycle

(+) - residual spermatozoa

• - SZ occupy less than 50% of gonad cross-section;

\* - cells of the subsequent spermatogenic cycle

**Table 3.** Number of the blue bream (*Ballerus ballerus* L.) used in the histological study

Sampling location	Sex	Number of fish per month												Total
		Jan	Feb	Mar	Apr	May	Jun	Jul	Aug	Sep	Oct	Nov	Dec	
Oder River	♀	10	8	12	19	17	15	9	6	6	8	8	10	128
	♂	7	5	11	15	6	8	6	5	6	7	1	4	81
Lake Dąbie	♀	5	9	10	32	12	12	7	7	17	6	6	6	129
	♂	5	5	9	6	20	22	6	6	8	6	1	3	97

lower than that of the females (chi-square test,  $\chi^2=10.57$ ,  $df = 1$ ,  $P<0.05$  and  $\chi^2=4.53$ ,  $df = 1$ ,  $P<0.05$ , respectively) (Table 3). The length and weight of the studied blue bream differ significantly (Kruskal–Wallis test,  $H = 166.23$ ,  $df=3$ ,  $P<0.05$  and  $H=54.91$ ,  $df=3$ ;  $P<0.05$ , respectively). Total body length and

weight of the females caught in particular locations were significantly higher than those of the males collected at the same sites (Kruskal–Wallis test,  $P<0.05$ ;  $P<0.05$  and  $P<0.05$ ;  $P<0.05$ , respectively). The length and weight of the females from the Oder River and Lake Dąbie differed significantly (Kruskal–

Wallis test,  $P < 0.05$  and  $< 0.05$ , respectively). The length of the males did not show statistically significant differences between the studied sites, while the weight of the males from the Oder River was significantly higher than that of the males from Lake Dąbie (Kruskal–Wallis test,  $P = 0.15$  and  $P < 0.05$ , respectively). The condition factor was similar in both sexes and in all studied locations (Kruskal–Wallis test,  $H = 1.95$ ,  $df = 1$ ,  $P > 0.05$ ;  $H = 3.28$ ,  $df = 3$ ,  $P > 0.05$ ) (Table 4).

#### Variation of GSI in Females

The GSI of the females from both sites was the highest from December to March (Kruskal–Wallis test,  $H = 55.75$ ,  $df = 11$ ,  $P < 0.05$  and  $H = 66.51$ ,  $df = 11$ ,  $P < 0.05$ ) (Figure 2a). In March, the females from both sites could be divided into 2 groups: those whose gonads would not mature in that season, with a GSI of  $2.93 \pm 1.74$  (average  $\pm$  SD) and  $0.72 \pm 1.11$  for the individuals from the Oder River and Lake Dąbie, respectively, and those with gonads preparing for spawning, with a GSI of  $15.64 \pm 2.44$  and  $11.06 \pm 2.46$ , respectively (Mann–Whitney U test,  $U = 20$ ,  $P < 0.05$ ). In April and May, 2 groups of females could be distinguished as well: those with gonads preparing for spawning and those with post-spawning gonads. In April, the GSI in the pre-spawning individuals from the Oder River and Lake Dąbie was  $11.36 \pm 3.31$  and  $12.98 \pm 5.49$ , respectively, while that in the post-spawning individuals was  $1.44 \pm 0.75$  and  $1.13 \pm 0.73$ , respectively (Mann–Whitney U test,  $U = 54$ ,  $P > 0.05$ ). In May, all females from the Oder River had post-spawning gonads (GSI  $1.96 \pm 0.96$ ), while the females from Lake Dąbie had either spawning gonads (GSI

$7.07 \pm 1.18$ ) or post-spawning gonads (GSI  $2.71 \pm 3.10$ ). The GSI value of the females from the two natural locations did not differ in a significant manner except March (Mann–Whitney U test,  $U = 44.0$ ,  $P > 0.05$ ). (Figure 2a). GSI in the female blue brems was the lowest after spawning, from May to July, with an average GSI value of  $1.62 \pm 0.82$  and  $1.47 \pm 0.72$  in the fish from the Oder River and Lake Dąbie, respectively, in July (Kruskal–Wallis test,  $P < 0.05$ ). From August, a gradual increase in the GSI value was observed at both sites. The changes in the GSI value depending on the stage of ovary maturity are presented in Table 5.

#### Variation of GSI in Males

In particular months the GSI value of the males from the two natural locations did not differ in a significant manner except April (Mann–Whitney U test,  $U = 7.0$ ;  $P < 0.05$ ) (Figure 2b). The highest GSI values were observed in the blue bream prior to spawning, when all male were at maturity stage IV (Kruskal–Wallis test,  $H = 93.91$ ,  $df = 9$ ,  $P < 0.05$  and  $H = 81.88$ ,  $df = 9$ ,  $P < 0.05$ ). The mean GSI value in the mature individuals was  $2.0 \pm 0.54$ . A significant reduction in GSI value was observed in May–June (Kruskal–Wallis test,  $P < 0.05$ ). The lowest GSI values were observed from May to August (average  $0.25 \pm 0.28$ ) (Kruskal–Wallis test,  $P < 0.05$ ). In these months, the individuals were at stage I, II, (substages II<sub>E</sub> and II<sub>L</sub>), V and VI–II. From September, the GSI values slowly increased until the following spring, i.e. the subsequent reproductive season (Figure 2b). The changes in the GSI value depending on the stage of testis maturity are presented in Table 5.

**Table 4.** Characteristics of the blue bream (*Ballerus ballerus* L.) examined, mean  $\pm$  SD and range. Values marked with different letters (a,b,c) shows significant differences between the features ( $P < 0.05$ ; ANOVA Kruskal–Wallis test)

Sex	Fishing location	n	Total length (cm)	Standard length (cm)	Fish weight (g)	C <sub>F</sub>
Female	Oder River	128	36.05 $\square$ 3.37 <sup>a</sup> 29.0–44.5	30.58 $\pm$ 2.98 <sup>a</sup> 24.58–38.5	456.27 $\pm$ 133.62 <sup>a</sup> 200.2–825.8	0.88 $\pm$ 0.11 <sup>a</sup> 0.70–1.19
	Lake Dąbie	129	34.57 $\square$ 3.22 <sup>b</sup> 18.5–48.9	28.57 $\pm$ 3.67 <sup>b</sup> 18.0–38.2	386.78 $\pm$ 168.92 <sup>b</sup> 44.6–1014.0	0.89–0.13 <sup>a</sup> 0.49–1.23
Male	Oder River	81	34.75 $\pm$ 3.56 <sup>c</sup> 27.4–41.4	28.66 $\pm$ 2.84 <sup>c</sup> 22.8–35.5	376.92 $\pm$ 125.57 <sup>b</sup> 175.5–852.6	0.87 $\pm$ 0.10 <sup>a</sup> 0.70–1.20
	Lake Dąbie	97	31.70 $\pm$ 3.71 <sup>c</sup> 25.3–44.6	26.48 $\pm$ 3.25 <sup>c</sup> 21.2–38.2	285.85 $\pm$ 122.14 <sup>c</sup> 119.5–800.3	0.84–0.10 <sup>a</sup> 0.64–1.16

**Table 5.** The GSI value changes of the blue bream (*Ballerus ballerus* L.) in stages of gonad maturity, mean  $\pm$  SD and range

Stage Sex	I	II	III	IV	V	VI
♀	-	0.86 $\pm$ 0.42 0.44–1.82	2.39 $\pm$ 1.39 1.19–5.34	11.52 $\pm$ 2.62 5.39–16.86	17.17 $\pm$ 0.94 15.45–19.12	1.97 $\pm$ 1.22 0.07–6.24
♂	0.11 $\pm$ 0.05 0.06–0.19	0.34 $\pm$ 0.36 0.10–1.45	1.74 $\pm$ 0.21 1.53–2.10	1.92 $\pm$ 0.43 1.17–2.72	0.98 $\pm$ 0.26 0.46–1.25	0.36 $\pm$ 0.21 0.19–0.78

## Reproductive Cycle of Female Oder River

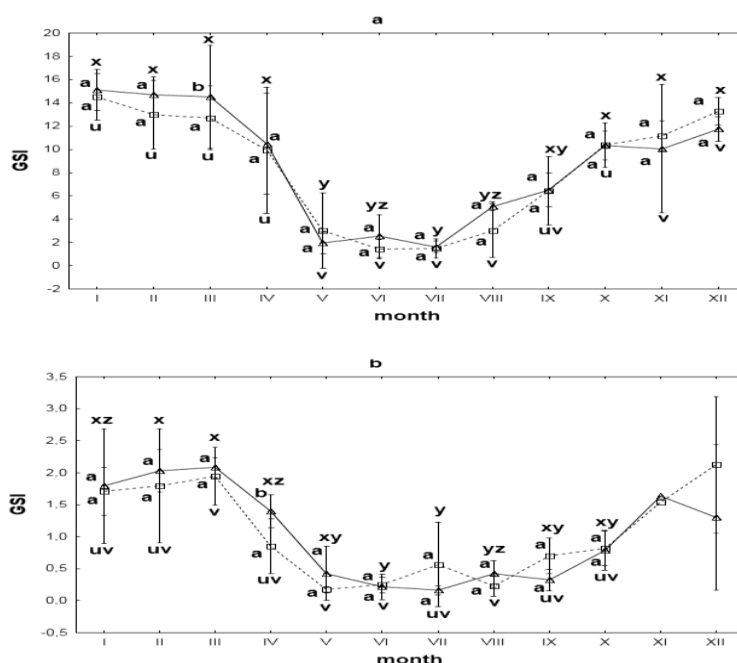
In June and July, all the blue bream females caught in the Oder River had gonads at stage III with oocytes at early stages of vitellogenesis and oocytes in previtellogenesis. In the gonads of the fish caught in July, there were numerous (more than 50% of gonad cross section area) degenerating oocytes resorbed after spawning (Figure 3a) Those oocytes were still observed in gonads in August (Figure 3b). At the end of that month, some females had gonads in advanced vitellogenesis (stage IV of gonad development). This stage was reached by all individuals in September and lasted until the first half of April (Figure 4a). In the second half of April, the females had gonads filled with oocytes with the nucleus at cell pole, i.e. spawning gonads (stage V). In early April, one female (30.1 cm in length) had gonads at stage III with degenerating oocytes (Figure 3c). However, the blue bream females caught in early and mid-May already had post-spawning gonads with post-ovulatory follicles and oocytes at early stages of vitellogenesis, (Figure 3d). All females caught in the second half of May had gonads at stage III. The smallest individuals with post-vitellogenic oocytes caught at that site were 24.5 cm in length.

## Lake Dąbie

Female gonads from June to August were at stage III and contained degenerating oocytes not released during spawning (observed in 50% of females in July). In September, 75% of females had gonads at stage IV with oocytes in advanced vitellogenesis and few small oocytes at stage IV, while the remaining females still had gonads at stage III (Figure 3e). Between October and March, all the blue bream females had gonads filled with oocytes at stage IV. In April, females had gonads ready for spawning (stage IV) with post-ovulatory follicles or spawning gonads (stage V). In the second half of April, post-spawning females (stage VI) could already be observed. In March and April, (Figure 3f), two females were caught (one per month, SL 24.0 and 25.5 cm, respectively) with gonads at stage II. Almost all females caught in May had post-spawning gonads and only single individuals from the second half of the month had gonads at stage V, ready to spawn. Therefore, most females from that site started spawning in the first half of April (Figure 4b). The smallest individuals with maturing gonads caught at that site were 18 cm long.

## Appearance and Size of Oocytes

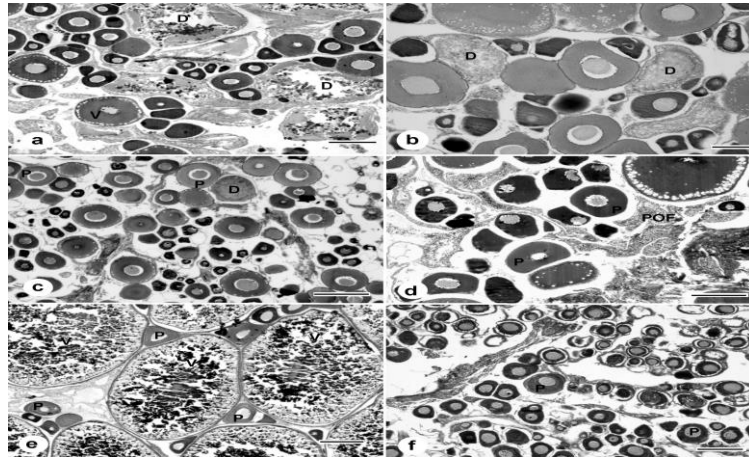
The histological image of gonads, as well as the appearance of oocytes in previtellogenesis and at the various stages of vitellogenesis in the blue bream females from the Oder River, Lake Dąbie and the



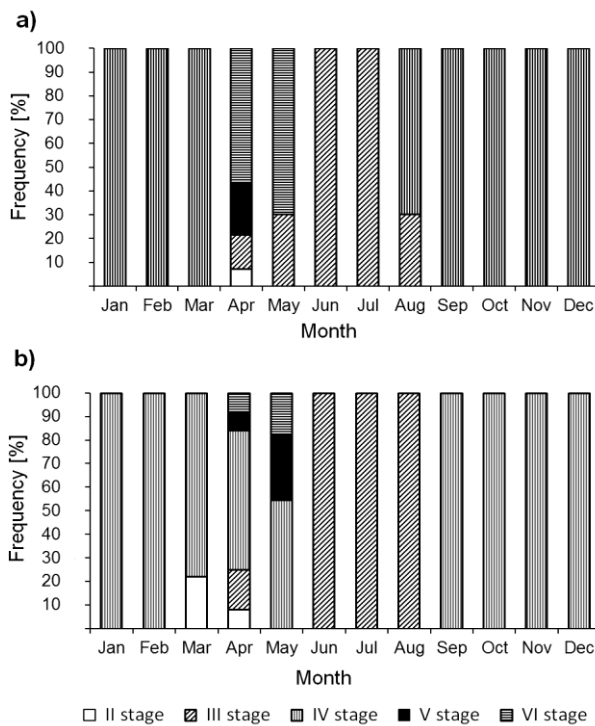
**Figure 2.** Monthly distribution of the blue bream (*Ballerus ballerus* L.) female (a) and male (b) gonadosomatic index GSI from the study areas:  $\Delta$  Oder River;  $\square$  Dąbie Lake.

Values marked with different letters (a,b) show significant differences between sites in individual months ( $P < 0.05$ ; Mann – Whitney U test); average  $\pm$ SD.

Values marked with different letters (x,y,z - Oder River; u,v - Dąbie Lake) show significant differences in GSI between months ( $P < 0.05$ ; ANOVA Kruskal–Wallis test). Due to the low number of males caught in November and December, the statistic was not performed for that months; average  $\pm$ SD.



**Figure 3.** A blue bream (*Ballerus ballerus* L.) ovary with oocytes in the early vitellogenesis (V) and degenerated oocytes (D), July, bar 200 μm (a); August bar 100 μm (b); with oocytes in previtellogenesis with numerous degenerated oocytes (D) April, bar 200 μm (c); after spawning with post-ovulatory follicles (POF) and the oocytes at the beginning of the next generation of previtellogenesis (P), May, bar 100 μm (d); with oocytes in an advanced vitellogenesis (V) and early previtellogenesis (P) September bar 200 μm (e); in late development in previtellogenesis (P), not mature in the current season, March, bar 100 μm (f).



**Figure 4.** Percentage contribution of the number of the blue bream (*Ballerus ballerus* L.) female with gonad in particular stage of maturity in individual months of the calendar year from two aquatic environment a) Oder River, b) Dąbie Lake.

Warm Canal, was similar. The oocyte size at the end of protoplasmic growth and at the onset of trophoplasmic growth in the females from Lake Dąbie was significantly smaller than in those from the Oder River (Mann–Whitney U test,  $Z=2.83$ ,  $P<0.05$ ) (Figure 5; Table 6). The size of oocytes found in the gonads of the fish from the Warm Canal (March, April, August) did not differ significantly from the oocytes in the fish caught at the other sites (Mann–Whitney U test,  $Z=2.47$ ,  $P>0.05$ ).

### Reproductive Cycle of Male Males from the Oder River

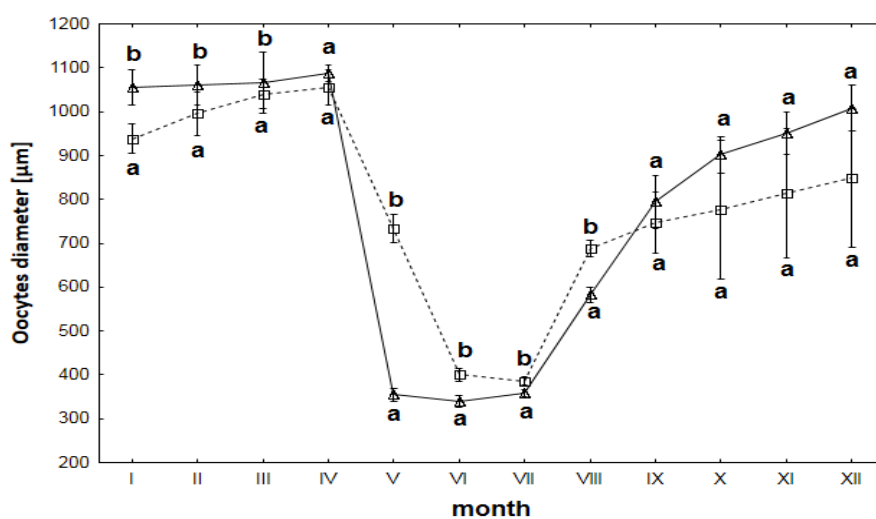
The blue bream males are ready for reproduction in early spring, when the proper environmental conditions occur, since their gonads reach maturity stage IV already in the winter. During winter, the gonads are filled with both numerous spermatozoa and cells at earlier stages of spermatogenesis located

in cysts. The finalization of spermatogenesis in some males (20%, stage V) occurred in the second half of April. In the following month, half of the males had spawning gonads at stage V and the first post-spawning males (30%, stage VI–II) were observed, while in the remaining males were at substage II<sub>E</sub>. Multiplying type B spermatogonia were already observed in gonads at stages V and VI. All post-spawning gonads found in the blue brems in that period were at stage VI–II. In June, some males still had gonads at stage V (20%), while most males had gonads at stage II, containing a varying number of closed cysts with type B spermatogonia. In July, neither spawning nor post-spawning gonads were observed. In that period, the process of multiplication of type A (stage I, Figure 6a) and type B (stage II, Figure 6b) spermatogonia continued in the cysts. The occurrence of spermatocytes and few spermatozoa in the gonads was reported in November (stage III, Figure 6c). Numerous spermatozoa in the lumen of the seminal tubules and numerous cysts with cells at earlier stages of spermatogenesis adjacent to gonad walls were observed in December at stage IV (Figure 6d). The gonads remained at that stage of

development until spring. Cysts with maturing cells disappeared in the first individuals in April, and the tubules were filled only with spermatozoa (stage V, Figure 6e). The first post-spawning individuals were observed in May (stage VI–II, Figure 6f), while some males had gonads filled with large amounts of sperm until the first half of July (stage V). Detailed percentage of males with gonads at each maturity stage is shown in Figure 7a.

**Males from Lake Dąbie**

The sexual cycle of the blue bream males from Lake Dąbie was similar to that of the males from the Oder River. In the winter, the gonads of the blue brems were at maturity stage IV. Males in that environment reached stage V, finalizing their spermatogenesis slightly sooner than the males from the Oder River. Only one male with delayed maturation was at maturity stage III in April. In May, a higher percentage of post-spawning males (45%) and a lower percentage of spawning males (20%) was reported at that site. The last male with a greater amount of sperm in the gonads (stage V) was found in



**Figure 5.** Monthly distribution of the blue bream (*Ballerus ballerus* L.) diameter of the most developed oocytes from the study areas: Δ Oder River; □ Dąbie Lake. Values marked with different letters (a,b) shows significant differences between the features in study area in particular month (P<0.05; Mann – Whitney U test); average ±SD.

**Table 6.** Sizes of oocytes at different stages of vitellogenesis in the blue bream (*Ballerus ballerus* L.) from the Oder River and Lake Dąbie, mean ± SD and range. Values marked with different letters (a,b) show significance of the differences in characteristics (P<0.05; Mann Whitney U - test)

Fishing location	Oocytes size (µm)			
	Previtellogenesis	Beginning of vacuolisation	First occurrence of the yolk	Completed vitellogenesis
Oder River	230.58±16.61 <sup>b</sup>	292.85±22.75 <sup>b</sup>	601.82±57.10 <sup>a</sup>	1008.13±40.42 <sup>a</sup>
Lake Dąbie	205.73±23.95 <sup>a</sup>	270.32±20.76 <sup>a</sup>	582.35±42.68 <sup>a</sup>	1039.77±85.72 <sup>a</sup>
	163.51-249.17	226.02-291.45	509.87-697.04	897.75-1327.16

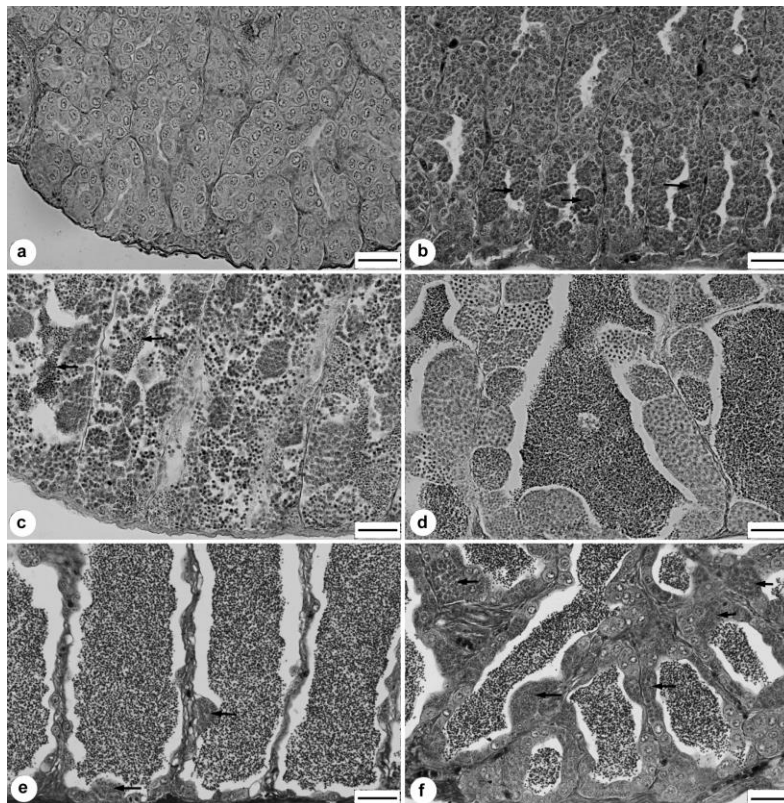
June. From July to October, few groups of degenerating cells were observed. In October, several males still had residual spermatozoa from the previous cycle in the seminal tubules. In November, similarly as in the individuals from the Oder River, the reproductive cells initiated meiotic divisions and the gonads reached stage III. In December, the gonads already had visible spermatozoa (stage III and IV). The gonads remained at maturity stage IV in the winter until the subsequent reproductive season. Detailed percentage of males with gonads at each maturity stage is shown in Figure 7b.

## Discussion

In the blue bream populations included in the study, males was outnumbered by females. In other cyprinid fish females dominated quantitatively over males as well (Balik *et al.*, 1999; Hamalosmanoğlu 2003; Okgerman *et al.*, 2011, 2012). Although other authors recorded more equal proportions of the sexes (Gürsoy, 2001; Altındağ, 2002; Yılmaz *et al.*, 2012) or domination of males (Koç *et al.*, 2006; Vatandoust *et al.*, 2014). The female to male ratio in the blue

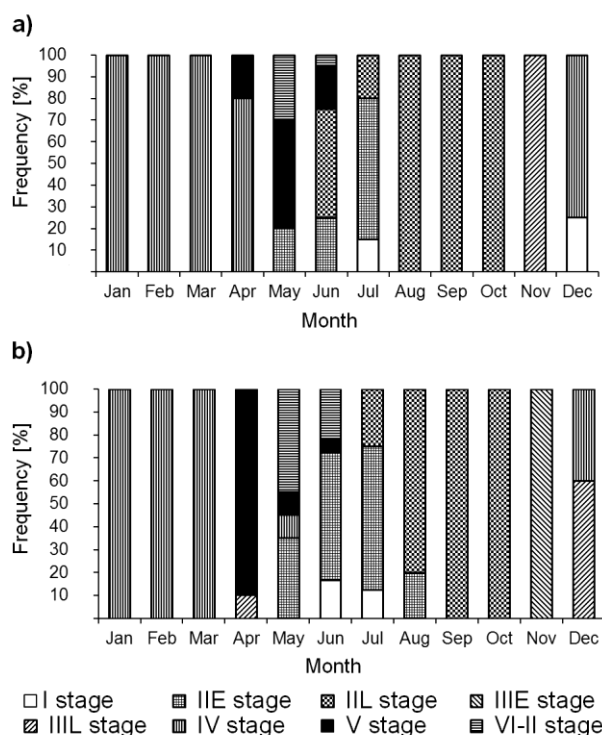
bream populations reported by other authors was of 1:0.25 (Krzykowski *et al.*, 1996) or nearly 1:1 (in the pre-spawning and spawning periods) (Karabanowicz and Kompowski, 1994; Kompowski and Blaszczyk, 1997).

The males caught in this study were smaller than the females which is confirmed by other authors (Karabanowicz and Kompowski, 1994; Krzykowski *et al.*, 1996). The differences in body sizes between the sexes were not always greater than 1 cm (Chašem, 1969; Karabanowicz and Kompowski, 1994). Body length and mass of the analysed fish were slightly greater than in the study by Kompowski (1991) in which the average length of the mature blue breams from Lake Dąbie was 25.0 cm (22 cm in the first spawning period), and in those from Szczecin Lagoon was 27 cm (20.3 cm in the first spawning period). The condition coefficient of the analysed fish was subject to seasonal variation. The highest value observed in the study was in the winter, while the lowest value occurred after spawning. In the study by Kompowski and Blaszczyk (1997), a similar fluctuation was noted. The condition coefficient in the females analysed by those authors decreased from 1.66 in the winter to



**Figure 6.** A blue bream (*Ballerus ballerus* L.) testis in stage I of maturation, by the tubule wall spermatogonia A occurred, July (a). A gonad in late stage II<sub>L</sub>, the tubules contain numerous cyst with multiplying spermatogonia B (arrow), September (b). Stage III<sub>L</sub>, all type of spermatogenic cells are visible in the gonad, first spermatozoa are formed (arrow), December (c). The spawning stage IV, the tubule lumen is filled with spermatozoa, by the tubule wall a numerous cysts with maturing cell occurred, February (d); The spawning stage V-II, the gonad with finalized spermatogenesis, tubules filled with spermatozoa, the tubule wall contains only resting type A spermatogonia and multiplying type B spermatogonia (arrow) May (e). A spent gonad in stage VI-II. Shrunken tubules filled with unexpelled spermatozoa. The tubule wall contains type A spermatogonia and cysts with type B spermatogonia (arrow), May (f). Scale bar 50 µm.





**Figure 7.** Percentage contribution of the number of the blue bream (*Ballerus ballerus* L.) male with gonad in particular stage of maturity in individual months of the calendar year from two aquatic environment a) Oder River, b) Dąbie Lake.

1.38 after spawning, while in the males, the value decreased from 1.59 to 1.45. The authors also observed that the males had a higher condition coefficient than the females. The phenomenon can be explained by the higher consumption of energy needed for the growth of the reproductive organs in females (Kompowski and Blaszczyk, 1997).

To date, no studies describing the sexual cycle of the blue bream using histological techniques have been conducted. Gonad development has been described based on gonad macroscopic analysis and the spawning period has been described based on the fluctuations in the GSI value (Kompowski and Blaszczyk, 1997).

The spawning period of the studied blue bream, a single-portion spawner, from Lake Dąbie and the lower Oder River was short and occurred in April. In the same locations, the common bream had spawned from April to June (one portion, Domagala *et al.* 2015a), and the white bream spawned from May to June (2 or 3 portions, Domagala *et al.* 2015b). In the post-spawning gonads of the blue bream females caught at both sites in July, degenerating oocytes resorbed after spawning were observed, similarly as in the white breams from Lake Kortowskie (Kopiejewska and Kozłowski, 2007) or from the Danube River, Lake Balaton and the Meuse River (Rinchar and Kestemont, 1996; Lefler, 2010). In the fish analysed in this study, oocyte vacuolization occurred in May and egg yolk appeared in August, while in the blue breams from Rybinski Reservoir

(Volodin, 1980), vacuolization occurred in June and egg yolk, as in the Polish individuals, appeared in August.

Oocyte sizes in the previtellogenesis and beginning of vacuolisation were smaller in the fish from Lake Dąbie than in those from the Oder River. However, there are no literature data to enable a comparison of the oocyte sizes of the blue breams from other sites. As confirmed by other studies where the size of oocytes is positively correlated with the standard length (Rana, 1988; Reid and Chaput, 2012). In other studies, however, has been no such correlation (Linhares *et al.*, 2014). In this population the size of the oocytes can also be affected adverse habitat (Hainfellner *et al.*, 2012) and how many times they consisted of female eggs (in the first spawning are larger oocytes) (Kjesbu, 1989).

The dynamic of male gonad maturation in the studied blue bream differed from that in the white bream and the common bream. In December, the analysed blue bream males reached maturity stage IV and awaited the early spring spawning period. In the winter, the white bream and the common bream have gonads at earlier maturity stages (II or III) and spawning in these species takes place later (Sakun and Butskaya, 1968; Brylińska and Długosz, 1973; Domagala *et al.*, 2015b). The blue bream male finalized spermatogenesis in April, however, the cycle in Lake Dąbie was completed slightly earlier than in the Oder River. The gonad filled with a large amount of spermatozoa was observed during 3 months, a

period similar to that in the white bream, although only a few individuals in the studied species retained this stage in this period (Domagała *et al.*, 2015b). The GSI of the analysed males was 5–6 times lower than in the females just before spawning (2 and 12%, respectively). Similar proportions were also noted in the blue bream by Kompowski and Błaszczuk (1997).

## Conclusion

For the first time, the blue bream gonad maturation has been described in both sexes based on histological analysis. The spawning period of the blue bream, a single-portion spawner, inhabiting the waters of the lower Oder River is short and lasts approximately two weeks in April. The blue bream males pass the winter with gonads at a more advanced maturity compared to other cyprinid species occurring in this area. The blue bream avoids thermally polluted waters. Therefore, the expected climate change involving an increase in water temperature (Souchon and Tissot, 2012) may have an effect on the occurrence of this species in the future.

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