

CHARACTERISTICS OF REPRODUCTIVE SYSTEM OF *Abramis brama* (L. 1758) SEXUALLY MATURE FEMALES IN VARIOUS TYPES OF AQUATIC BASINS OF DNIESTER RIVER

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Abstract. The characteristics of *Abramis brama* sexually mature females reproductive system, inhabiting various types of Dniester river water basins is given. In pre-spawning period in some individuals from Vatra-Ghidighici reservoir degeneration affect the proper envelope of all oocytes of older generation. In individuals from the lower portion of the river these changes affect 90% of the oocytes of younger generation, which form the second portion of roe. In Some individuals of Dubăsari reservoir the resorption of sexual cells in the final phases of trophoplasmatic growth is observed, which leads to omission of spawning season.

Keywords: oogenesis, oocyte, gonads, *Abramis brama*, vitellogenesis.

Rezumat. Caracterizarea sistemului reproducător al femelelor adulte de *Abramis brama* (L. 1758) în diverse tipuri de ecosisteme acvatice ale bazinului fluviului Nistru. Este dată caracterizarea sistemului reproducător al femelelor adulte de *Abramis brama*, care populează diverse tipuri de habitate în bazinul fluviului Nistru. La unele femele din lacul de acumulare Vatra-Ghidighici în perioada prereproductivă membrana tuturor oocitelor generației mature este afectată de degenerări. La indivizii din sectorul inferior al fluviului aceste modificări afectează 90% din oocitele generației tinere, care formează a doua porție de icre. La unii reproducători din lacul de acumulare Dubăsari a fost observată resorbția celulelor sexuale în fazele finale de creștere trofoplasmatică, ceea ce duce la ratarea sezonului de depunere a icrelor.

Cuvinte cheie: oogeneză, oocit, gonade, *Abramis brama*, vitelogeneză.

INTRODUCTION

Bream is a widespread species in the rivers, aquatic reservoirs and ponds of Moldova. In Dniester River, before the regulation of its flow, the bream population was divided into resident and semi-anadromous forms. Currently, as a result of hydraulic engineering, several local intrapopulation groups, fairly isolated from each other were formed, which differ after their growth rate, linear sizes, number of age groups, terms of spawning and character of gametogenesis phases occurrence (CEPURNOVA, 1991). In the reconstructed water reservoirs of Moldova the bream showed high ecological plasticity and a wide range of structural and functional adaptation toward changing environmental conditions (BODAREU et al., 1986; CEPURNOVA, 1972; SHATUNOVSKII et al., 2009).

Research features of development of the reproductive system in representatives of cyprinid fishes in Ichthyologic literature were always in scientists' attention. However, the analysis of the development of sexual cells in the bream in the changed conditions of their habitats is relatively poor studied (KOSHELIOV, 1984; STATOVA, 1985; CEPURNOVA, 1991).

The peculiarities of oocyte development in bream from different water bodies is reflected in changes in the duration of the maturity stages during pre-vitellogenesis that affect the age of sexual maturity of fish, and during the reproductive cycle the duration of oocytes passage of vitellogenesis process affects the length of IV-th stage of gonad maturity. Furthermore, in different water basins in the gonads of bream females develop different number of fish roe portions. As evidenced by our study (FULGA & USATHI, 2008), the bream in Prut river and Dubăsari reservoir is at a time spawning fish, whereas in Kuchurgan and Costești-Stânca reservoirs live females with at a time spawning, as well as with portions spawning (STATOVA et al., 1988; FULGA et al., 2011).

In the present work the morphological and functional characteristics of the reproductive system of bream females from Dniester river is presented.

MATERIAL AND METHODS

For histological examination of gonads sexually mature females were used collected from net catches in the lower Dniester, and reservoirs Dubăsari and Vatra-Ghidighici during the period 2007-2010 in the number of 180 individuals. Samples of gonads were fixed in Bouen's fluid with subsequent treatment according to the standard technique. Stages of gonad maturity were determined according to the recommendations of Sakun, Butskaia, (SAKUN & BUTSKAIA, 1963), and the degree of oocytes development according to the classification of Kazanskii, (KAZANSKII, 1949). Slices were 7 microns thick and were stained after Mallory method (ROSCHIN & LIBENSON, 1957). Gonado-somatic index (GSI) was determined by the relative weight of the gonads to the weight of the carcass. The dimension of yolk oocytes was determined by ocular micrometer. All digital data were processed statistically (LAKIN, 1980). Photomicrographs were made with a microscope "Lomo, Mikmed-2" with a video camera, using the ocular increase of 10x; objective 15x.

STUDY RESULTS AND DISCUSSIONS

Vatra-Ghidighici water reservoir, Lower Dniester. The analysis by age and sex structure of population of bream in the reservoir Vatra Ghidighici showed that if in 1990 in the population 9 age groups of fishes were observed (DOLGHII, 1993) currently the number fell to seven.

For histological studies 4 and 5 years old females from Vatra-Ghidighici reservoir and 4, 5 and 6 years old females from lower Dniester were used.

In all studied specimens from the reservoir is observed asynchronous type of oocyte development. Number of eggs of older generation who have completed the accumulation of yolk granules in the cytoplasm varies from 70-76% of all oocytes of trophoplasmatic growth. The ovules of younger generations are in the phases of complete vacuolization of the D₃ cytoplasm and the beginning of D₄ vitellogenesis. This composition of sexual cells is characteristic to the females with portion type of spawning. Biological analysis of bream females from the reservoir Vatra Ghidighici showed that their body length and weight are higher than that of individuals of the same age from the Lower Dniester ($P > 0.999$; $P > 0.999$), respectively (Table 1).

Table 1. Biological characteristics of bream females from different populations.

Age, years	Vatra Ghidighici reservoir			Lower Dniester		
	Body length, cm	Body weight, g	Gonad weight, g	Body length, cm	Body weight, g	Gonad weight, g
4	35.5±0.57	948±44.08	150.0±14.86	29.3±0.43	587±25.58	70±10.70
5	39.5±0.70	1286±63.74	221.5±38.31	30.5±0.54	645±22.64	99.3±4.02
6	-	-	-	35.4±2.14	968±35.42	201.0±21.00

The high index of growth rate of four and five-year-old bream females in the reservoir indicates a higher level of food supply, providing favourable conditions for foraging animals. As a result, there is an increase in mass of produced sexual products. Their value is significantly higher $P > 0.95$, than the Dniester bream of the same age (Table 1).

The values of GSI and size of oocytes in the phase of completed vitellogenesis in four and five-year-old females from the Lower Dniester are somewhat lower than in specimens from the reservoir, which is associated with more favourable feeding conditions in the last ones. But the differences of these parameters are not significant ($P < 0.95$) (Table 2).

Table 2. Indexes of individual reproductive capacity in breams from different populations.

Age, years	Vatra-Ghidighici reservoir			Lower Dniester		
	GSI, %	Oocyte dimension in phase "E", μm	FC (after Clark)	GSI, %	Oocyte dimension in phase "E", μm	FC (after Clark)
4	19.92±1.38	837±4.99	1.67±0.04	14.63±2.39	812±5.28	1.93±0.10
5	22.30±2.89	856±5.44	1.60±0.08	20.44±1.75	842±7.60	1.77±0.09
6	-	-	-	28.2±1.20	933±7.52	1.61±0.5

According to our research, the spawning period in bream from the reservoir Vatra-Ghidighici begins in late April - early May. In females, caught on May 2, there is an asynchronous maturation of oocytes and their ovulation in small portions. In the gonads, in this period, there are sporadic empty follicular membranes, oocytes at different stages of maturation and completed vitellogenesis with the nucleus in the centre, as well as oocytes from younger generation in phases D₃, D₄. It should be noted that in some females their own oocyte membrane of older generation are affected by degenerative changes. This is manifested in its swelling and the disappearance of striation (Fig. 1). Such oocytes ovulate, but their capacity of fecundation decreases.

For bream females in the Lower Dniester the asynchronous type of gametogenesis is typical and before the spawning two generations of oocytes form, which is consistent with the data of Chepurnova (CEPURNOVA, 1972). The author points out that the second generation of oocytes doesn't spawn, and is subjected to resorption in the phase of completed vitellogenesis. However, the data of Statova (STATOVA, 1985) indicate the presence of females in the Lower Dniester, which spawn two portion of roe. Our study showed that in the gonads of bream mature females in the prespawning period there are two generating of oocytes: the eldest are in the phase of completed vitellogenesis (E), while the younger cells are at the stage of complete vacuolization (D₃) and beginning of vitellogenesis (D₄).

The first portion of roe constitutes an average of 72% of all oocytes of tropho-plasmatic growth. A large number of oocytes from younger generation (90%), forming the second portion of roe are affected by resorption process, expressed in oocyte membrane swelling, disappearance of nuclei and merge of vacuoles content in a homogeneous mass (Fig. 2).

Similar changes in the development of eggs from younger generations in the prespawning period are not mentioned by any of the above mentioned authors.

Actually, in bream females in the prespawning period minor variations in the coefficient of fatness were revealed by age from 1.61 to 1.91. According to Statova (STATOVA, 1985) materials, this indicator has an average of 2.12, which, at its turn, determines the best environmental and trophic conditions for bream in that period.

Gonado-somatic index (GSI) in the six-year-old females before spawning reaches 28.9%, whereas in four and five years old individuals the values of this parameter differ significantly: $P < 0.95$ (Table 2).



Figure 1. Resorption of oocytes in the phase of vitellogenesis beginning (original).



Figure 2. Swelling of membrane of oocytes of the second generation in the phase of completed vitellogenesis (original).

According to our data given in Tables 1 and 2, indicating slight differences in the values of the GSI, of the length and weight at four and five-year-old females $P < 0.95$, it can be assumed that they are for the first time maturing individuals.

In the past years (ZELENIN et al., 1990), bream females became sexually mature at four years, with an average body mass of 605 g, and the body length of 31.6 cm. But there is evidence that only 21% of the bream population from Dniester reach sexual maturity at four year, and the rest – at the age of five year old (TOMNATIC & BATIR, 1970).

Analyzing the above presented data we can say that with age in females, especially the repeatedly maturing ones, there is an increase in the GSI and in the size of definitive oocytes, but coefficient of fatness decreases. This is the result of more intensive generative metabolism, when the organism resources are spent on body growth and development of eggs.

The bream spawning is currently happening in the same calendar period as in previous years (STATOVA, 1985) in late April - early May. The study has revealed the asynchronous maturation and ovulation of mature eggs in small portions. In histological preparations single empty follicular membranes can be observed, oocytes at different stages of maturation, early vitellogenesis and complete vacuolization, most of which are affected by degenerative changes.

Dubăsari Water reservoir. After the spawning season, in the second half of June, the ovaries of bream pass into II-III stages of maturity. In the gonads, together with the protoplasmic growth period oocytes there are present oocytes of cytoplasm vacuolization beginning (Fig. 3). Along with the development of a new generation cells the resorption of empty follicular membranes and of not spawned yolk oocytes.

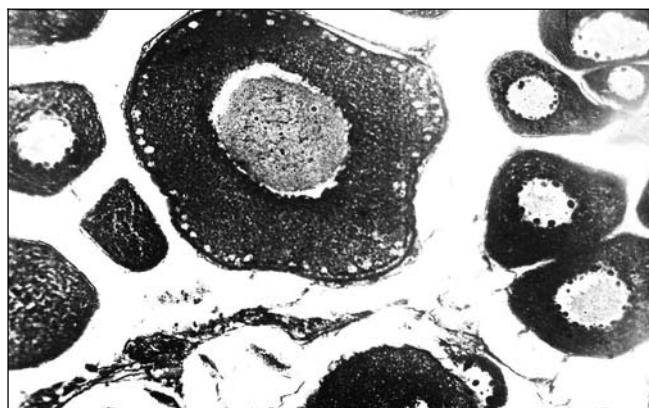


Figure 3. Ovary after spawning bream on the II-III stage of maturity (original).

The gonad-somatic index of spawned females decreases to $2.42 \pm 0.05\%$. The lack in mature bream females from Dubasari Reservoir of II stage of gonad maturity was observed also in the first years after its formation (ZELENIN, 1960).

Further development of oocytes is asynchronous. In August, the gonads of repeatedly maturing females at III maturity stage contain oocytes of 400-480 microns, which are with fully vacuolated cytoplasm (D_3) and oocytes with diameter of 234-332 microns with one or two rows of vacuoles (D_1 - D_2).

Asynchrony in the development of sexual cells continues in the autumn. In the process of accumulation of yolk granules, together with the oocytes in phases of intense vitellogenesis (D_5 - D_6), whose diameter is 700-784 microns, are present oocytes with diameter of 530-590 microns in the initial phase of yolk accumulation in the cytoplasm (D_4). This composition of the oocytes describes the IV incomplete maturation stage of the gonads, which begins in late October. GSI by this time reach $15.59 \pm 1.20\%$.

After the winter, in early spring, the intense process of yolk formation in oocytes continue and at the beginning of the spawning season in repeatedly maturing females the majority of the gonad mass are constituted by the oocytes in the phase of the completed vitellogenesis, reaching in the first half of May the definitive size of 800-890 microns. GSI reaches its maximum values of $23.26 \pm 1.87\%$.

According to the researches of Zelenin (ZELENIN, 1960), the IVth stage of maturity in bream lasted 5 months, from December to May. At present, the period of accumulation of yolk granules in oocytes increased to 6 months, but the ovulation of mature oocytes, as in previous years, occurs in the II half of May to early June. A wide range of spawning temperatures allows females to start spawning at lower range of temperature optimum.

During the recent years, due to lack of environmental conditions that meet the regulatory requirements for the development of oocytes in the final phase of tropho-plasmatic growth, the resorption of yolk oocytes occurs and as a result, the females skip the spawning season (Fig. 4).

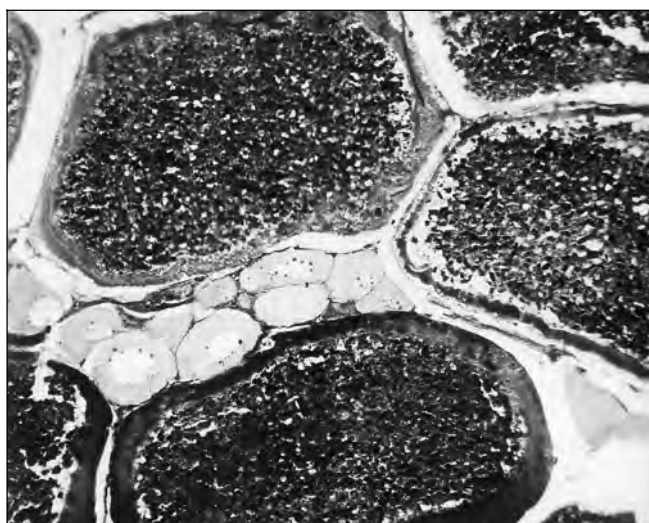


Figure 4. Resorption of yolk oocytes in the final phase of tropho-plasmatic growth (original).

In mid-June in 30% of studied females in the gonads were resorbed oocytes in phases of intensive and completed vitellogenesis, indicator of which is the absence of the nucleus and of cortical vacuoles, as well as the destruction of yolk granules and fusion of their contents in a homogeneous mass. We have also found females, in which there was a partial spawning of mature eggs, confirmation of this is the presence in the ovary of a small number of empty follicular envelopes and yolk oocytes in the initial stage of resorption. The reason for this was the influx of cold water from the middle part of the Dniester River in Dubăsari Reservoir, which has led to organic water pollution and the danger of the dissolved oxygen concentration lowering to critical values. In addition, the spring water releases from the Dnestrovsk reservoir, in Dubăsari Reservoir inflow more mineralized water with a predominance of sulphate ions, and in industrial sewage the predominance of mineral and organic components of the detergent was recorded (ZUBKOVA et al., 2004).

CONCLUSIONS

1. In the bream females inhabiting the Lower Dniester River and the reservoir Vatra Ghidighici two generations of oocytes form, the eldest of whom is numerous and constitute an average of 72% and 74% respectively.
2. In the prespawning period in some females of the reservoir Ghidighici the degeneration affect the proper membrane all oocytes of older generation. In females of the Lower Dniester these changes occur in 90% of oocytes of the younger generation, which for the second roe portion.
3. In females, inhabiting in different ecological conditions, along with the age, the relative weight of gonads and the size of eggs that had completed vitellogenesis increase, but the coefficient of fatness decreases, which indicates a higher level of generative metabolism in repeatedly maturing individuals.
4. As result of negative anthropogenic impact on the fish fauna of the lower Dniester River and Vatra - Ghidighici reservoir, the number of age groups in the bream population has reduced.
5. Currently, in certain bream individuals from Dubăsari Reservoir, due to the lack of optimal conditions for the completion by oocytes of trophoplasmatic growth, in the prespawning period their resorption is observed, which leads to the omitting by females of the spawning season.
6. Despite the increase in the duration of vitellogenesis in oocytes by one month, the spawning in bream females from Dubăsari reservoir occurs in the same calendar period as before the construction of the Dnestrovsk hydroelectric Power Station.

The study was performed under the fundamental project of Institute of Zoology 10. 08. 079 F.

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Received: March 20, 2013
Accepted: April 16, 2013