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STEROID HORMONES, SELENIUM, AND ZINC IN THE GONADS – GAMETES – LARVAE BIOLOGICAL SYSTEM OF THE MUSSEL *MYTILUS GALLOPROVINCIALIS* LAM.

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A. O. Kovalevsky Institute of Biology of the Southern Seas of RAS, Sevastopol, Russian Federation
E-mail: *lar_sa1980@mail.ru*

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Assessment of the interaction of marine farms with the environment in the industrial cultivation of the mussel *Mytilus galloprovincialis* is very important. In the mussel farm – environment system, biotic fluxes of chemical compounds through gonads, gametes (sperm and eggs), and larvae make a considerable contribution to this interaction. Since gonads play a key role in the mussel reproduction, it is interesting to study the budget of materials, that are directly involved in this process. Out of these materials, testosterone, estradiol, fatty acids, and some trace minerals, such as Se and Zn, are known to affect spawning, growth, and development. The molluscs absorb these materials from food and water. These materials are partly metabolically assimilated by mussels and partly excreted into the environment with gametes. The aim of this study was to estimate the components of the budget of steroid hormones, fatty acids, and two essential trace elements (Zn and Se) in mussel gonads, gametes, and larvae. The total testosterone and estradiol in gonads and gametes were quantified by enzyme-linked immunosorbent assay. The contents of the trace elements were found using inductively coupled plasma mass spectrometry. The fatty acid composition was determined by means of gas chromatography–mass spectrometry. The contents of Se and Zn in mussel gonads and gametes were found to depend on the stage of the reproductive cycle. In female gonads, Se and Zn concentrations were higher than in male ones. The highest concentration of Se was recorded in eggs: (14.7 ± 2.9) $\mu g \cdot g^{-1}$ dry weight (d. w.). In sperm, it was (14.4 ± 1.8) $\mu g \cdot g^{-1}$ d. w. Zn content in gonads before spawning was higher than in gametes. In male gonads and in sperm, its values were (27.5 ± 3.7) and (19.3 ± 6.4) $\mu g \cdot g^{-1}$ d. w., respectively. In female gonads and eggs, the contents of zinc were (53.6 ± 10.9) and (49.3 ± 8.2) $\mu g \cdot g^{-1}$ d. w., respectively. In spring, the mean values of Se and Zn assimilation degree (q) in gonads of the mussel were within 0.1–0.6. The limit values of the alimentary accumulation coefficient (K_{lim}) of Se and Zn ranged 0.6 to 1.4. While spawning, mussels excrete polyunsaturated fatty acids (PUFA), which are probably used by other marine organisms. Up to 56.2 % of PUFA are excreted with sperm, and 48.1 %, with eggs, whereas in larvae this fraction does not exceed 10.2 %. The data obtained indicate that the molluscs assimilate sex hormones, fatty acids, selenium, and zinc to maintain vital processes: prostaglandins are synthesized from PUFA in the body, and testosterone esters are formed from testosterone. Se and Zn, when coupled with proteins, play a key role in the reproduction and formation of larval shells.

Keywords: mussel *Mytilus galloprovincialis*, gametes, larvae, selenium, zinc, testosterone, estradiol, fatty acids, Black Sea

Testosterone, estradiol, polyunsaturated fatty acids (hereinafter PUFA), and trace elements possess high biological activity (Kapranova et al., 2019 ; Nikonova et al., 2017). Selenium protects an organism, as well as sperm vitality, against reactive oxygen species. Its absence during spermatogenesis affects

the sperm quality and fertility of animals (Ahsan et al., 2014). Zinc is known to be used to maintain the functions of the reproductive system, as well as to activate enzymes, DNA synthesis, and proteins in the body. Therefore, the studies that allow assessing the content of steroids, fatty acids (hereinafter FA), Se, and Zn in the reproductive system of hydrobionts are of particular relevance.

Marine mussel farms are an important component of coastal marine ecosystems. In the mussel farm – environment interaction, a key role is played by biotic fluxes of chemical compounds and energy in the matter → gonads → gametes (sperm and eggs) → larvae system. The balance approach allows estimating the fluxes of the compounds through these components. The energy balance of the Black Sea mussel settlements in natural populations has been studied in most detail (Finenko et al., 1990). For marine farms, a similar approach was implemented in the investigation of the flux of carotenoids in the environment – mussel (*Mytilus galloprovincialis*) – mussel biodeposition system determining the qualitative and quantitative composition of carotenoids in various organs of *M. galloprovincialis*, depending on the season and assessment of the pigment assimilation by the mollusc (Pospelova & Nekhoroshev, 2003). As a result of the study of individual carotenoids' transformation in the process of metabolism and quantitative assessment of their assimilation, the number of carotenoids assimilated and excreted by molluscs was determined.

However, the data on the element balance of other biologically active substances involved in the metabolism, such as steroid hormones, FA, and biogenic trace elements, in mollusc gonads and gametes are very limited. It determined the aim of this work.

MATERIAL AND METHODS

The material for the research was the bivalve *Mytilus galloprovincialis* Lamarck, 1819, grown in the mollusc farms of Sevastopol: in Laspi Bay (44°24.56′N, 33°42.19′E) and Karantinnaya Bay (44.61°N, 33.49°E). Mussels were collected by diving from a depth of 2–3 m in April and May 2020. The water temperature range was +7…+21 °C. A total of about 600 uniform-size mussels with a shell length of 50–60 mm was processed. Before the experiment, the molluscs were kept in filtered seawater for 3–6 hours to clear the digestive tracts. Mussel sex and sexual maturation stage were determined on gonadal smears under a microscope, basing on the analysis of histological preparations of the gonads (Pirkova et al., 2019). Mussel sperm and eggs were obtained by the method previously described by Nikonova *et al.* (2017). The dry weight (hereinafter d. w.) of gametes was determined after drying 1 mL of the suspension of homogenized sperm and eggs at +105 °C. At the same temperature, the dry weight of gonads was determined.

The total concentration of testosterone and estradiol in gonads and gametes was quantified by enzyme-linked immunosorbent assay (Nikonova et al., 2017). The trace element concentration was determined by inductively coupled plasma mass spectrometry using multielement standard IV-ICPMS-71A (Inorganic Ventures, the USA). The relative composition of FA was determined according to the method developed by Kapranova *et al.* (2019). The larvae were obtained and their FA composition was determined using the method described in (Kapranova et al., 2020). Measurements of trace elements and FA composition were carried out at the "Spectrometry and Chromatography" core facility at IBSS.

RESULTS AND DISCUSSION

The results of the analysis of steroid hormones, Se, and Zn concentration in gonads, eggs, and sperm of the mussel *M. galloprovincialis* at different ripening stages are given in Table 1.

Table 1. Concentration of steroid hormones, selenium, and zinc in gonads, eggs, and sperm of the mussel *M. galloprovincialis*

Gonadal ripening stages (gametes)	Concentration of steroid hormones, $\times 10^{-6}$ μg·g^{-1} d. w.				Concentrations of trace elements, μg·g^{-1} d. w.			
	Total testosterone		Estradiol		Selenium		Zinc	
	♂	♀	♂	♀	♂	♀	♂	♀
1	7757.8 ± 2315.2	2154.5 ± 643.1	90.1 ± 28.6	512.5 ± 33.1	n. d.	n. d.	n. d.	n. d.
2	2453.1 ± 1409.8	592.1 ± 112.8	120.9 ± 27.8	623.0 ± 40.8	12.0 ± 1.5	n. d.	33.3 ± 6.6	n. d.
3	781.1 ± 60.1	210.3 ± 30.0	104.7 ± 30.1	747.0 ± 30.0	8.7 ± 0.7	10.8 ± 1.7	24.6 ± 3.6	35.6 ± 11.8
4	979.0 ± 83.9	352.0 ± 192.0	119.5 ± 26.3	636.7 ± 22.0	9.3 ± 1.9	9.0 ± 2.5	16.3 ± 1.9	45.5 ± 31.2
5A	975.1 ± 464.3	859.0 ± 116.1	132.2 ± 34.3	529.0 ± 26.1	10.7 ± 2.8	9.7 ± 3.3	22.9 ± 5.7	56.3 ± 17.9
5B	692.2 ± 115.4	144.2 ± 14.4	110.6 ± 20.4	501.8 ± 34.4	8.8 ± 1.4	13.7 ± 1.9	27.5 ± 3.7	53.6 ± 10.9
Eggs	n. f.	10.1 ± 4.8	n. f.	539.5 ± 122.8	n. f.	14.7 ± 2.9	n. f.	49.3 ± 8.2
Sperm	14 284.8 ± 259.2	n. f.	194.4 ± 59.2	n. f.	14.4 ± 1.8	n. f.	19.3 ± 6.4	n. f.

Note: n. f. denotes not found; n. d., no data; 5A and 5B, gonads before and after spawning.

Selenium concentrations in gonads and gametes of female and male mussels (Table 1, Fig. 1) change similarly to the concentrations of steroid hormones (Nikonova et al., 2017) and depend on the mollusc reproductive cycle (Goede et al., 1993). Se concentration in male gonads is positively correlated with testosterone concentration. The Pearson correlation coefficient is 0.89 ($p = 0.045$).

The maximum concentration of selenium is determined in the gametes: in eggs, (14.7 ± 2.9) μg·g^{-1} d. w., and in sperm, (14.4 ± 1.8) μg·g^{-1} d. w. The decrease in Se content in mussel gonads after spawning is due to the beginning of the post-spawning restructuring phase.

Selenium is an essential element for mussels. The percentages of its entry into the body are 90 % with food and 10 % with water (Ahsan et al., 2014). All this Se is in a bivalent organic form, with selenocysteine (SeCys) predominating in animal products and selenomethionine (SeMet) predominating in plant products. Selenium enters the animal body mainly in the form of selenomethionine. Se is transported and deposited by selenoproteins containing selenocysteine. Selenoproteins are selenocysteine-containing redox proteins involved in the antioxidant reaction. Se is an integral part of selenoproteins, which protect sperm during maturation against oxidative damage and also serve as structural components of mature sperm. Thus, selenium and selenoproteins provide sperm viability, as well as protection against reactive oxygen species. Genetic studies of selenoproteins have shown that their absence during spermatogenesis results in abnormal sperm development, which in turn affects sperm quality, fertility, and libido (Ahsan et al., 2014).

In the study of selenoprotein from the pearl mussel *Cristaria plicata*, it was possible to express messenger ribonucleic acid (mRNA) from tissues of the mantle, gills, hemocytes, muscles, and hepatopancreas. The highest expression occurred from hepatopancreas tissues (Hu et al., 2014 ; Kopp et al., 2018).

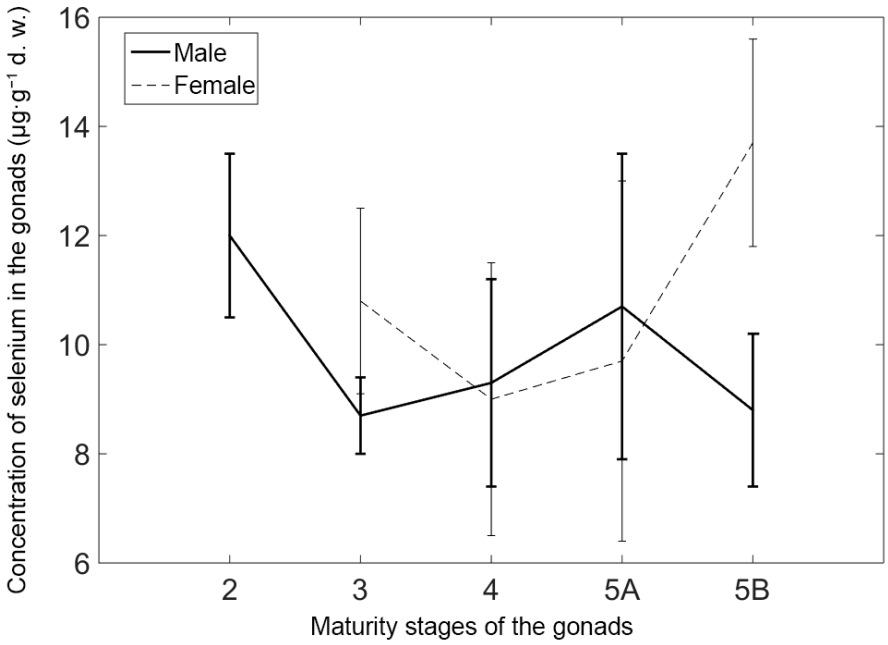

Fig. 1. Concentration of selenium in gonads of the mussel *M. galloprovincialis*: 5A and 5B denote gonads before and after spawning

Zinc is used by the animal body to activate enzymes, synthesize DNA and proteins, and maintain the functions of the reproductive system. It also acts as antioxidant inhibiting the ability of free radicals to damage cell tissue and genetic material. Zn excretion from the soft tissues of mussels seems to occur through eggs during spawning (Lowe & Moore, 1979). In *M. galloprovincialis* collected in several seaports of Western Algeria, zinc concentration varied within 87.1–731.5 μg·g^{-1} d. w. (Hadj et al., 2012). In the Black Sea mussel sperm, Zn concentration was (30.4 ± 6.4) μg·g^{-1} d. w.; in eggs, it was (115.4 ± 24.2) μg·g^{-1} d. w. (Karavantseva et al., 2012). In the present work, the higher Zn content was recorded in gonads before spawning than in gametes, which indicates a partial excretion of zinc with sperm and eggs. In male gonads before spawning, Zn content was (27.5 ± 3.7) μg·g^{-1} d. w., and in sperm, it was (19.3 ± 6.4) μg·g^{-1} d. w. In female gonads, the concentration was (53.6 ± 10.9) μg·g^{-1} d. w., and in eggs, it was (49.3 ± 8.2) μg·g^{-1} d. w.

Zinc assimilated with food is related mainly to the mussel soft tissues, while Zn from seawater is deposited mostly in shells (Fisher et al., 1996). Changes in the mass of soft tissues and shells in *M. galloprovincialis* are likely to result from different levels of Zn accumulation. Specifically, the weight of zinc-contaminated mussel shells increased after a 51-day cleaning period (Soto et al., 2000).

In our studies, a negative correlation between zinc content and estradiol concentration in female gonads at different ripening stages was revealed. The Pearson correlation coefficient was –0.98 ($p = 0.024$). Estradiol is recognized as an important regulator of both food intake and energy consumption (Mauvais-Jarvis et al., 2013). Estradiol regulates body weight, contributing to its increase (Mauvais-Jarvis et al., 2013).

As shown in (Soto et al., 2000), Zn was accumulated in the soft tissues proportionally to its concentration in seawater, while its content in hemolymph was slightly higher than in the environment. Zinc assimilation was through intestines, mantle, and gills. Zn was transported to the kidneys from gills and intestines ($t_{½} \approx 8$ days) through the hemolymph either as a high-molecular complex or as granular amoebocytes. Most of zinc was in granular amoebocytes, which were found either in all body tissues or in intestines

and kidneys. Thus, in the mussel *Mytilus edulis*, the kidney is an organ for the storage of many trace elements; it contains 30 % (about 1000 μg·g^{-1} d. w.) of total zinc concentration in the body. In this case, Zn is localized in the form of insoluble granules in some intracellular organelles, which occupy about 20 % of the cell volume. Zinc is excreted by defecation, exocytosis of renal granules to the urine, and diapedesis of amoebocytes (George & Pirie, 1980).

To assess the assimilation degree of Se and Zn with food by hydrobionts, a mathematical model was applied based on the equation of G. G. Polikarpov and V. N. Egorov (1986):

$$\frac{dC_{\text{mussels}}}{dt} = R(C_f q - C_{\text{mussels}} q_f) - C_{\text{mussels}}\, p\,, \qquad (1)$$

where $C_{mussels}$ and C_f are concentrations of a chemical element in the hydrobiont and its food, μg·g^{-1};

R is the relative food intake rate, day^{-1};

q is the degree of an element assimilation from food;

q_f is the degree of food assimilation for growth (= K_2);

p is the element exchange rate of the hydrobiont, day^{-1}.

The equation proposed by G. G. Polikarpov and V. N. Egorov (1986) for describing the kinetics of trace element exchange in hydrobionts during element assimilation with food can be applied for mussels if the concentration of trace elements in their gonads and gametes is measured given the known coefficient K_2. The degree of trace element assimilation from food is estimated from the coefficient q, which may be an important characteristic determining the need of marine organisms for the trace elements in question.

By transforming this equation (Pospelova et al., 2018), a formula was obtained for estimating the assimilation degree of selenium (zinc) q from the measurement data of element concentrations in mussel tissues and gametes:

$$q = \frac{C_{\text{gon.}} q_f}{C_{\text{gon.}} q_f + C_{\text{gametes}}(1 - q_f)}\,, \qquad (2)$$

where q is the degree of element assimilation from food;

$C_{gon.}$ is the concentration of a chemical element in gonads, μg·g^{-1};

$C_{gametes}$ is its concentration in mussel sperm or eggs, μg·g^{-1};

q_f is the degree of food assimilation for growth (= K_2).

It should be noted that this approach did not take into account Se and Zn fractions that could be excreted through biodeposition.

Previously, a study of the kinetics of trace element content in *M. galloprovincialis* showed as follows: the concentration and exchange of a trace element in mussels could be considered as an integral process in ontogenesis (Polikarpov & Egorov, 1986). In the first approximation, the mean annual value of the food assimilation for growth – q_f (K_2) (Finenko et al., 1990) – can be used to assess the element assimilation by molluscs. For q_f limits (0.14 and 0.42), q values were determined for Se (Zn) in spring (Table 2).

In mussel gonads in spring, the mean annual q values for selenium fluctuate in the range 0.1 to 0.4, which is lower than the degree of food assimilation for growth (K_2). It is known that if the efficiency of trace element assimilation from food is lower than the degree of food assimilation for growth, the transfer of matter along the trophic chain proceeds with a decrease in the rate of contamination in the subsequent link (Polikarpov & Egorov, 1986). Mean annual q values for zinc are 0.1–0.6.

Table 2. Assimilation of selenium and zinc with food (q) by the mussel *Mytilus galloprovincialis*

Gonadal ripening stages	q Se (q_f = 0.14)		q Zn (q_f = 0.14)	
	♂	♀	♂	♀
1	n. d.	n. d.	n. d.	n. d.
2	0.12 ± 0.03	n. d.	0.22 ± 0.08	n. d.
3	0.09 ± 0.03	0.11 ± 0.07	0.17 ± 0.01	0.11 ± 0.01
4	0.10 ± 0.04	0.09 ± 0.07	0.12 ± 0.05	0.13 ± 0.02
5A	0.11 ± 0.06	0.10 ± 0.08	0.16 ± 0.01	0.16 ± 0.01
5B	0.09 ± 0.04	0.13 ± 0.08	0.19 ± 0.01	0.15 ± 0.01
Gonadal ripening stages	q Se (q_f = 0.42)		q Zn (q_f = 0.42)	
	♂	♀	♂	♀
1	n. d.	n. d.	n. d.	n. d.
2	0.38 ± 0.09	n. d.	0.55 ± 0.07	n. d.
3	0.30 ± 0.06	0.35 ± 0.02	0.48 ± 0.01	0.34 ± 0.03
4	0.32 ± 0.01	0.31 ± 0.01	0.38 ± 0.08	0.40 ± 0.06
5A	0.35 ± 0.02	0.32 ± 0.02	0.46 ± 0.02	0.45 ± 0.03
5B	0.31 ± 0.11	0.40 ± 0.20	0.51 ± 0.07	0.44 ± 0.20

Note: n. d. denotes no data; 5A and 5B, gonads before and after spawning.

With known values of the degree of an element assimilation from food (q) and the degree of food assimilation for growth (q_f), it is possible to determine the limiting coefficient of food accumulation of a trace element according to the equation $K_{lim} = Rq / (Rq_f + p)$ (Polikarpov & Egorov, 1986). From this equation (at p = 0, where p is the element exchange rate in the hydrobiont, day^{-1}), $K_{lim} = q / q_f$. That is, K_{lim} is equal to the ratio of the degree of Se or Zn assimilation from food to the degree of food assimilation for growth (Table 3).

Table 3. Maximum coefficient of food accumulation of trace element (K_{lim}) by the mussel *M. galloprovincialis*

Gonadal ripening stages	K_{lim} Se (q_f = 0.14)		K_{lim} Zn (q_f = 0.14)	
	♂	♀	♂	♀
1	n. d.	n. d.	n. d.	n. d.
2	0.86 ± 0.21	n. d.	1.57 ± 0.59	n. d.
3	0.64 ± 0.23	0.79 ± 0.52	1.21 ± 0.07	0.79 ± 0.07
4	0.71 ± 0.31	0.64 ± 0.53	0.86 ± 0.35	0.93 ± 0.12
5A	0.79 ± 0.42	0.71 ± 0.62	1.14 ± 0.07	1.14 ± 0.07
5B	0.64 ± 0.74	0.93 ± 0.61	1.36 ± 0.07	1.07 ± 0.07
Gonadal ripening stages	K_{lim} Se (q_f = 0.42)		K_{lim} Zn (q_f = 0.42)	
	♂	♀	♂	♀
1	n. d.	n. d.	n. d.	n. d.
2	0.90 ± 0.20	n. d.	1.31 ± 0.2	n. d.
3	0.71 ± 0.12	0.83 ± 0.05	1.14 ± 0.02	0.81 ± 0.07
4	0.76 ± 0.02	0.74 ± 0.02	0.90 ± 0.23	0.95 ± 0.14
5A	0.83 ± 0.05	0.76 ± 0.05	1.10 ± 0.05	1.07 ± 0.07
5B	0.74 ± 0.23	0.95 ± 0.54	1.21 ± 0.22	1.05 ± 0.54

Note: n. d. denotes no data; 5A and 5B, gonads before and after spawning.

K_{lim} values for selenium and zinc in mussel gonads in spring are 0.64 to 1.36, and those are higher than the degree of assimilation and involvement in biochemical processes (q) of the considered trace elements. This fact shows that Se and Zn are excreted with gametes into the environment.

Since we used mussels grown on marine farms, we calculated the concentration of steroid hormones, selenium, and zinc in gametes of one ton of molluscs – 71,124 specimens with 51–60 mm in size (Kholodov et al., 2017) (Table 4).

Table 4. Concentration of testosterone, estradiol, selenium, and zinc *per* one ton of the mussel *M. galloprovincialis*

Stages	Content of steroid hormones, $\times 10^{-6}$ g·t^{-1} d. w.				Content of trace elements, g·t^{-1} d. w.			
	Total testosterone		Estradiol		Selenium		Zinc	
	♂	♀	♂	♀	♂	♀	♂	♀
5A	975.1 ± 464.3	859.0 ± 116.1	132.2 ± 34.3	529.0 ± 26.1	10.7 ± 2.8	9.7 ± 3.3	22.9 ± 5.7	56.3 ± 17.9
5B	692.2 ± 115.4	144.2 ± 14.4	110.6 ± 20.4	501.8 ± 34.4	8.8 ± 1.4	13.7 ± 1.9	27.5 ± 3.7	53.6 ± 10.9
Eggs	n. f.	10.1 ± 4.8	n. f.	539.5 ± 122.8	n. f.	14.7 ± 2.9	n. f.	49.3 ± 8.2
Sperm	14 284.8 ± 259.2	n. f.	194.4 ± 59.2	n. f.	14.4 ± 1.8	n. f.	19.3 ± 6.4	n. f.

Note: n. f. denotes not found; 5A and 5B, gonads before and after spawning.

According to Table 4 and literature data (Kapranova, 2020 ; Scott, 2018), steroids and trace elements are excreted by molluscs into the aquatic environment with gametes. In spring, during mass spawning, testosterone, estradiol, and selenium are largely transferred to gametes. Zinc is only partly excreted, which is most likely due to the portioned mussel spawning. Mass mussel spawning in the Black Sea is repeated twice a year: in spring and autumn. The maximum number of spawning mussels is recorded in mid-April. Mass spawning in autumn begins in September – October and continues in November – December (Kholodov et al., 2017). The peak of mussel spawning in the southeastern Crimean coast of the Black Sea is recorded in December – January, and the lesser one is registered in May – June. Thus, the seasonal duration of the mass spawning averages 4 months *per* year.

The work of A. V. Pirkova *et al.* (2019) shows the dynamics of gonad maturation and sex ratio of mussels depending on the season. The maximum number of males spawn in July, and the ratio of females to males in the sample (%) is 25.0 : 75.0.

The elements of FA balance in the mussel *M. galloprovincialis* can be represented in the *gonads – gametes – trochophores* conditional system as shown in Table 5.

Changes in FA profile in mussel gonads and gametes depending on the maturation stage have already been discussed in (Kapranova et al., 2019). The content of SFA in mussel trochophores is approximately equal to the total content of SFA in eggs and sperm (Kapranova et al., 2020) (Table 5). This dependence can be explained by the fact that before the formation of organs and tissues, mussel trochophores are passively fed; SFA seem to perform mainly a protective function, forming the shells of cell membranes (Fokina et al., 2010). Moreover, at the initial maturation stages, FA are probably involved in the esterification of steroid hormones since this process involves predominantly C16 and C18 SFA (Scott, 2018).

Table 5. Fatty acid content (% of total lipids) in gonads, reproductive products, and trochophores of the mussel *M. galloprovincialis*

Gonadal ripening stages (gametes)	Saturated fatty acids (SFA)			Monounsaturated fatty acids (MUFA)			Polyunsaturated fatty acids (PUFA)		
	♂	♀	Larvae	♂	♀	Larvae	♂	♀	Larvae
1	100	100	58.2	n. f.	n. f.	31.6	n. f.	n. f.	10.2
2	48.2	100		15.8	n. f.		36.0	n. f.	
3	35.1	100		11.3	n. f.		53.6	n. f.	
4	42.5	81.1		35.9	3.7		21.6	15.2	
5	100	44.4		n. f.	22.1		n. f.	33.5	
Eggs	n. f.	46.7		n. f.	5.2		n. f.	48.1	
Sperm	34.0	n. f.		9.8	n. f.		56.2	n. f.	

Note: n. f. denotes not found.

MUFA content in both female and male gametes of mussels is almost two times lower than in gonads. This fact shows that both saturated and unsaturated FA enter the mussel body with food and water and then are used in processes maintaining their vital activity (Pospelova et al., 2018 ; Orban et al., 2002). Similar to MUFA content, PUFA content in larvae is an order of magnitude lower than the total PUFA content in female and male gametes since most of FA enter the mussel tissues from microalgae and are accumulated throughout the life cycle. PUFA are necessary for adaptation to environmental conditions (temperature, salinity, *etc.*) (Fokina et al., 2010 ; Orban et al., 2002). In addition, in molluscs, PUFA are probably predecessors of prostaglandins (Rowley et al., 2005). Total prostaglandin content in molluscs is low (Tadasi & Hiroshi, 1976); nevertheless, prostaglandins and related eicosanoids, being oxygen-containing metabolites of C20 PUFA, have a physiological effect on bivalve spawning (Stanley-Samuelson, 1994). Excessive amounts of PUFA are possibly released into the aquatic environment and then absorbed by other hydrobionts.

Conclusion. The data obtained indicate that the mussels are most likely unable to synthesize testosterone, estradiol, and fatty acids. Molluscs assimilate selenium, zinc, sex steroids, and fatty acids with food and water to maintain vital functions. In the process of biochemical transformations, prostaglandins are synthesized from PUFA in the body, and testosterone esters are formed from testosterone. Se and Zn play a key role in mollusc reproduction. Selenium content in male gonads is positively correlated with testosterone content. Zinc affects the weight of mussel gonads. Zn concentration in female gonads is negatively correlated with estradiol content. By the example of selenium and zinc in spring, the values of the assimilation degree of these elements by mussel gonads from food (q) were quantified along with the limiting coefficient of food accumulation of these trace elements (K_{lim}). For the considered trace elements, q values are lower than the degree of food assimilation for growth (K_2), which indicates an active interaction of mussels with the environment. Excessive amounts of free forms of testosterone, estradiol, fatty acids, Se, and Zn are excreted with gametes in order to maintain a balance between free and bound forms of these substances. Together with gametes, the necessary amounts of steroids and trace elements are transferred to the nascent larvae, which are endogenously fed during the first few days. During spawning, one ton of mussels is capable of excreting into the environment with sperm up to $14.28 \cdot 10^{-3}$ mg of testosterone, $0.19 \cdot 10^{-3}$ mg of estradiol, 14.4 g of zinc, and 19.3 g of selenium. With eggs of one ton of mussels, $0.54 \cdot 10^{-3}$ mg of estradiol, 14.7 g of Se, and 49.3 g of Zn are excreted. Mussels serve as a source of PUFA, which are probably used by other hydrobionts. With sperm, up to 56.2 % and with eggs, up to 48.1 % of PUFA is excreted; in larvae, this value does not exceed 10.2 %.

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СТЕРОИДНЫЕ ГОРМОНЫ, СЕЛЕН И ЦИНК В БИОЛОГИЧЕСКОЙ СИСТЕМЕ ГОНАДЫ — ПОЛОВЫЕ ПРОДУКТЫ — ЛИЧИНКИ МИДИИ *MYTILUS GALLOPROVINCIALIS* LAM.

Л. Л. Капранова, В. И. Рябушко, М. В. Нехорошев, С. В. Капранов

ФГБУН ФИЦ «Институт биологии южных морей имени А. О. Ковалевского РАН»,
Севастополь, Российская Федерация
E-mail: *lar_sa1980@mail.ru*

Оценка взаимодействия морских хозяйств с окружающей средой при промышленном выращивании мидии *Mytilus galloprovincialis* весьма актуальна. В системе мидийная ферма — среда важную роль играют биотические потоки веществ через гонады, половые продукты (сперма и яйцеклетки) и личинки. Поскольку гонады выполняют ключевую роль в размножении мидий, представляется интересным рассмотреть элементы баланса веществ, принимающих непосредственное участие в этом процессе. Тестостерон, эстрадиол, жирные кислоты, а также селен и цинк, потребляемые моллюсками вместе с пищей и водой, прежде всего необходимы им для осуществления нереста, а также для роста и развития. Часть потребляемых веществ задействуется организмом мидий в процессе метаболизма, а часть экскретируется в водную среду вместе с половыми продуктами. Цель данной статьи — провести количественное определение элементов баланса стероидных гормонов, жирных кислот и биогенных микроэлементов в гонадах, половых продуктах и личинках моллюсков, играющих важную роль в метаболизме их организма. Концентрацию общего тестостерона и эстрадиола в гонадах и половых продуктах определяли методом твёрдофазного иммуноферментного анализа. Содержание микроэлементов измеряли методом масс-спектрометрии с индуктивно-связанной плазмой. Относительный состав жирных кислот гонад, половых продуктов и личинок мидий определяли методом хромато-масс-спектрометрии. Концентрации селена и цинка в гонадах и половых продуктах мидий зависят от стадии репродуктивного цикла. В женских гонадах содержание селена и цинка выше, чем в мужских. Наибольшая концентрация селена обнаружена в яйцеклетках — (14,7 ± 2,9) мкг·г$^{-1}_{сух}$. Концентрация в сперматозоидах — (14,4 ± 1,8) мкг·г$^{-1}_{сух}$.

Содержание цинка в гонадах до нереста выше, чем в половых продуктах. В гонадах самцов до нереста концентрация цинка составляет (27,5 ± 3,7) мкг·г$^{-1}_{сух}$, в сперматозоидах — (19,3 ± 6,4) мкг·г$^{-1}_{сух}$. В гонадах самок — (53,6 ± 10,9) мкг·г$^{-1}_{сух}$, в яйцеклетках — (49,3 ± 8,2) мкг·г$^{-1}_{сух}$. В весенний период значения степени усвоения селена и цинка из пищи (q) в гонадах мидий колеблются в диапазоне от 0,1 до 0,6. Значения предельного коэффициента пищевого накопления селена и цинка ($К_п$) составляют от 0,6 до 1,4. Мидии служат источником полиненасыщенных жирных кислот (ПНЖК), которые, вероятно, используются другими гидробионтами. Со спермой выделяется до 56,2 % ПНЖК, с яйцеклетками — 48,1 %, тогда как в личинках этот показатель не превышает 10,2 %. Полученные данные свидетельствуют о том, что моллюски потребляют гормоны, жирные кислоты, селен и цинк из пищи и воды для поддержания жизненных процессов: из ПНЖК в организме образуются простагландины, из тестостерона — сложные эфиры тестостерона. Селен и цинк, соединяясь с белками, играют ключевую роль в размножении и формировании оболочек личинок.

Ключевые слова: мидия *Mytilus galloprovincialis*, половые продукты, личинки, селен, цинк, тестостерон, эстрадиол, жирные кислоты, Чёрное море