Psammophilic bivalves of the Azov Sea. *Donax trunculus* (Linnaeus, 1758)

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Abstract. Typical members of the psammophilic fauna of bivalve mollusks inhabiting the coastal zone of the Kerch Strait (the waters of the Sea of Azov) are species of the family Donacidae (Linnaeus, 1758). It is noted that in recent years there has been a restoration of the populations of Donax trunculus (Linnaeus, 1758) and Donax semistriatus (Poli, 1795). The paper presents the results of studies of the length-weight composition, features of the reproductive cycle, technochemical composition and nutritional value of D. trunculus as the dominant species in 2023-2024. The length of the mollusk shells varied within the range of 14.0–37.0 mm, height -10.5-23.0 mm, average weight (4.99 ± 0.04) g, flesh yield -13.4%, the valves account 65.7% of the total weight of the mollusk. Correlations were found between the length and weight, the length and weight of soft tissues, and between the mollusk weight and the weight of soft tissues; the correlation coefficients (R) were 0.99, 0.98, and 0.99, respectively. The results of the studies of the chemical composition and energy value showed that D. trunculus flesh is low in calories, containing 74.8% of absolute value of protein substances, with valine and histidine being the limiting essential amino acids. The mollusk flesh has a high iodine content of 0.042% (RDI: 130-200 µg/day).

1 Introduction

The psammophilic bivalve community includes species that inhabit the sand layer in the coastal zone. The richness of the fauna is determined by the size of the sand particles (medium- and fine-grained ones are the richest), the amount of organic matter (the degree of saprobity) and the activity of the surf.

Two species of burrowing clams of the family *Donacidae* (Linnaeus, 1758): *Donax trunculus* (Linnaeus, 1758) and *Donax semistriatus* (Poli, 1795), inhabiting the Azov-Black Sea basin, are typical members of psammophilic fauna. These species are popular seafood delicacies for the population of a number of coastal countries, valuable sources of

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biologically active substances, protein and polyunsaturated fatty acids. They are edible, have good taste qualities, and are promising objects for fishing and aquaculture.

The total global catch of *Donax* genus mollusks according to FAO (2024) [1] in recent decades has reached more than 1000 tons (Figure 1). *D. trunculus* and *D. semistriatus* mollusks are widespread on the Mediterranean and Atlantic coasts of Western Europe (Spain, Portugal, and Italy are the leaders in production), as well as in the Mediterranean, Aegean, Red, Marmara, and Black Seas, where they are in high demand in the seafood market. [2].

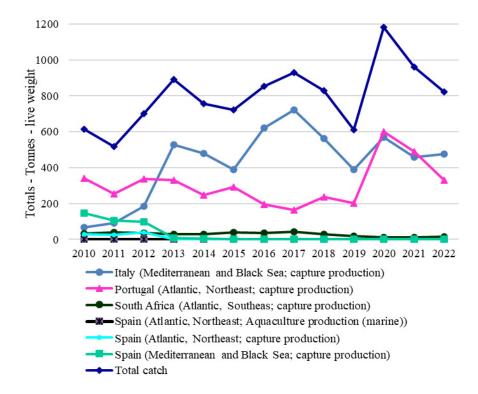


Fig. 1. World catch of clams of *Donax* genus (FAO, 2024).

Donax are usually inhabitants of open beaches, preferring to live at a depth of 0-2 m near the coast, subject to intense wave action. In such conditions, mollusks of the genus *Donax* form a dominant group, finding here shelter from predators, reduced competition and easy access to food supply [3].

Differences in the abundance and distribution of mollusks are largely related to the bottom characteristics. The lowest abundance of *Donax* is observed on beaches consisting of finer shell rock (sand). The highest was recorded on beaches composed of medium and large shell rock. A decrease in the abundance of *Donax* is noted in recreational areas during resort recreation and the active tourism season.

Previously, in the Black Sea, *Donax trunculus* and *Donax semistriatus* were found everywhere at depths of up to 10-15 m in the surf zone with intense hydrodynamics and high oxygen content [4]. In the Kerch Strait, *Donax* also predominate in the surf zones, and are characterized by resistance to changes in salinity and temperature [2].

By 2010, *D. trunculus* and *D. semistriatus* had significantly reduced their range in the Kerch Strait and the pre-strait area of the Black Sea, which is probably due to siltation of

the bottom and eutrophication of the sea, water constructions, pollution of port areas and other anthropogenic factors [2, 5].

In 2019, shells (paired valves) of both species of *Donax* were found in wave wash-outs in the area of Cape Maly, with 22 individuals, 13 to 20 mm long, belonging to the species *D. semistriatus* and only 5 individuals, 7 to 25 mm long, belonging to *D. trunculus*.

In the autumn of 2024, on almost all sandy beaches of the Crimean part of the Kerch Strait, valves of these species of mollusks were noted in the washes-out, with a significant predominance of *D. trunculus* (Figure 2). Thus, in the area of the village of Naberezhnoye, more than 100 valves were collected. The presence of psammophilic mollusks in the washes-out should probably be considered as evidence of the process of restoration of their populations in the Kerch Strait in their former habitats, where these species were previously widespread. This was also confirmed by the finds of live specimens of Donax and fairly large settlements of the rare Black Sea clam species *Donacilla cornea* (Poli, 1791) [6].



Fig. 2. Appearance of clams: *Donax semistriatus* (Poli, 1795) (1), *Donax trunculus* (Linnaeus, 1758) (2) (the Kerch Strait, October, 2024).

Due to the increase in the number of *D. trunculus* mollusks in the region, the study of the psammophilic community of this species is of great practical interest. The purpose of the research is to study the biology, length-weight characteristics and nutritional value of *D. trunculus* of the Kerch Strait in autumn.

2 Materials and methods

The material for the study was samples of the bivalve mollusks *Donax trunculus* (Linnaeus, 1758), collected in the Kerch Strait (Sea of Azov) in September 2019 and August-October 2023 and October 2024. Samples were processed in laboratory conditions according to the generally accepted method [7].

Studies of the chemical composition were carried out using standard methods adopted in a comprehensive chemical analysis, namely: the total content of nitrogenous substances – according to the Kjeldahl method using a FOSS auto-nitrogen analyzer; mineral substances – gravimetrically, after burning at a temperature of 600-700 °C, the composition of macro- and microelements – by capillary electrophoresis.

The protein-water coefficient of mollusk tissues and the lipid-protein coefficient, which characterize the tenderness of the flesh consistency, were determined by means of calculation methods. The ratio of the total content of lipids, protein, and carbohydrates to the water content determines the food saturation coefficient of mollusk flesh [8].

Assessment of the biological value of the proteins of mollusks was carried out according to the method of H. H. Mitchell & R. J. Block [9], in accordance with which the index of amino acid scores is calculated.

The experimental data were processed by parametric methods using MS Excel software packages. The results of the studies were statistically processed using Student's t-test (p = 0.95), the spread of data is presented by standard deviation.

3 Results and Discussion

Average weight of the mollusk is (4.99 ± 0.04) g $(\overline{x} \pm \Delta x)$, with p = 0.95 and n = 55), flesh yield – 13.4%, the valves account for 65.7% of the total weight of the mollusk (Table 1): the sample weights – 720 and 580 g, respectively.

Weight with	Shell length,	Flesh	Shell	Intervalvular		
a shell, g	a shell, g mm		weight, g	liquor, g		
n = 30, September, 2023						
5.10 ± 0.02	28.6 ± 1.1	0.70 ± 0.01	3.32 ± 0.04	1.09 ± 0.04		
n = 25, October, 2023						
4.87 ± 0.02	26.0 ± 1.5	0.70 ± 0.02	3.30 ± 0.04	1.04 ± 0.04		

Table 1. Averaged data on the length and weight composition *Donax trunculus* $(\overline{x} \pm S_x)$.

In the samples dated October 2024, *D. trunculus* was predominant, the length of the mollusk shells varied within 14.0-37.0 mm (average (25.7 ± 0.4) mm), height -10.5-23.0 mm (average (15.0 ± 0.2) mm). In terms of length, the population was mainly represented by size groups from 22 to 30 mm (Figure 3, a), and in height -12-18 mm (Figure 3, b). A close correlation was found between the length and height of the shells, which is best described by a power-law dependence law (Figure 4).

The maximum weight of *D. trunculus* reached 4.2 g, which is lower than the values for the same period in 2023 (Table 1) and can be explained by the different physiological state of the mollusks.

Correlation links were found between length and weight, length and weight of soft tissues, and between the weight of the mollusk and the weight of soft tissues (Figure 5, a-c).

It is known that the weight of *Donax* changes during the year depending on the state of the reproductive system [10-12]. After spawning in the warm season, its weight begins to decrease, then in the autumn and winter periods, in the process of preparing for gametogenesis, the body weight can increase (in the Black Sea species – from November to February [10]). As a rule, after winter growth, the weight stabilizes until July, when the eggs begin to mature. After spawning, the weight decreases and does not change until November.

The reproductive cycles of *D. trunculus* and *D. semistriatus* are close; in the Black Sea they have one spawning peak per year, with more than 90% of individuals entering the dormant stage in September-October [4]. During the winter period, their gonads are in a state of rest, starting in November, nutrients gradually accumulate in the eggs, the content of protein, lipids, carbohydrates increases. Histological analysis showed a gradual growth of oocytes, which goes into an active phase with increasing temperatures in spring and summer [10].

In the Black Sea, *Donax* spawn once a year, while in the Mediterranean Sea, off the coast of Egypt and Algeria, *Donax* have two spawning peaks [11-12], and in Australia, *D. (Plebidonax) deltoides* (Lamarck, 1818) spawns throughout the year [13].

Environmental conditions affect the gonadosomatic index and meat yield, which are lower in less productive waters. The gonads occupy most of the interior of the shell, as well as a small part of the leg [14].

The lifespan of *Donax* is 4-6 years [5]. In *D. semistriatus*, a slight predominance of males was noted in the population, while in *D. trunculus*, the differences were expressed to a lesser extent. It has been shown [12, 14] that the sex ratio can change depending on the state of the habitat. With significant pollution, the number of males in the population increases. These mollusks are a kind of indicator of the state of the environment.

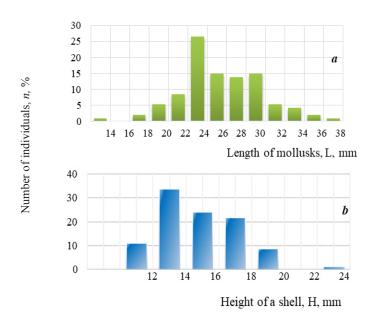


Fig. 3. Size composition of *Donax trunculus* in the Kerch Strait (October, 2024).

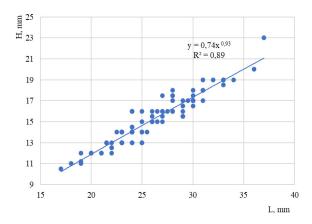


Fig. 4. Dependence of the height (H) of the shell of *Donax trunculus* on the length (L) (n=92).

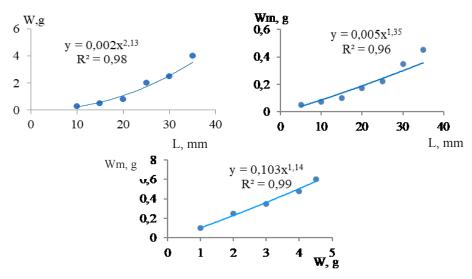


Fig. 5. Dependence of the total weight (W) and the weight of soft tissues (Wm) on the length (a, b) and the weight of soft tissues (Wm) on the total weight (W) (c) for *Donax trunculus*.

Due to the growing demand for shellfish flesh and the need to increase their catch volumes, the study of the biological characteristics of individual species and the development of methods for their artificial breeding are becoming a priority. Thus, in Australia, the reduction in the catch of the bivalve mollusk D. deltoides, which lives on the beaches of south-eastern Australia, prompted researchers to begin producing its juveniles in artificial conditions [13]. For this purpose, adult mature individuals of the mollusk with a shell length of 35-65 mm were collected in the intertidal zone in October/November 2008 and kept in pools of the system without a sand substrate. The water temperature was maintained at a recirculation level of 23 °C, and a mixture of Isochrysis sp., Diacronema (=Pavlova) lutheri and Chaetoceros muelleri microalgae was used as feed. The mollusks adapted well to the conditions of the recirculation systems, despite the lack of a substrate in which they could burrow. Most adults extended their legs and siphons and easily filtered the algal diet (faeces appeared within 12 hours). However, attempts to initiate spawning by temperature shocks (increasing the temperature by 4-5 °C) were unsuccessful. Therefore, gametes were obtained through incisions on the surface of the gonads, from which eggs and sperm were collected by means a pipette.

Two batches of zygotes were obtained, and after 48 hours, D-veliger larvae were collected and transferred to fresh seawater. A total of 1.0×10^5 larvae were obtained. The larvae were fed a mixture of microalgae. This study confirmed that it is possible to obtain *Donax* eggs and larvae in artificial conditions, but further research is needed to improve the methods of spawning, fertilization, and larval rearing.

In contrast to the *Donax* culture method used in Australia, two types of devices were tested for *Donax* culture in North Carolina (USA) [15]: an exact replica of a standard clam channel and a modified version of an "upweller". The channels were made from standard PVC gutters 1 m long. The upwellers were made from 50 cm long, 10 cm diameter PVC pipes with attached bottoms. Water was supplied to the bottoms of the upweller cylinders via centrally located 50 cm long, 3 cm diameter PVC pipes. The results showed that the accumulation of sediments and feces of *Donax spp.* occurred quite rapidly, requiring cleaning of the channels and upwellers every two weeks.

The authors carried out parallel studies over a period of 21 months. They compared the growth and survival of mollusks in the mentioned cultivation systems according to the

following parameters: farming method, water flow rate, mollusks density per unit area, and the ratio of raw and filtered seawater [15].

The highest survival rate of *Donax spp.* (81%) maintained in the channel method was lower than the lowest survival rate of mussels in upwellers (94%), which indicates a more efficient method of farming in upwellers. In addition, it was found that the addition of a mixture of live algae at 20 °C caused more efficient spawning – up to 100,000 fertilized eggs. Gonad analysis showed that in March and November, almost 100% of *Donax spp.* have mature gonads and are capable of spawning.

Thus, the results of the studies confirm the prospects of farming *Donax* spp. and producing aquatic foods in a commercial scope.

During the experiment on keeping D. trunculus in artificial conditions during the autumn period, it was found that the mollusk tolerates captivity conditions (in an aquarium) well with recirculation of sea water with a salinity of 18-20‰. With a gradual decrease in temperature from 24 to 21-22°C, the increase in length and weight was, respectively, +0.4 mm and -0.10 g. The weight of the mollusk decreased in accordance with the change in the state of the reproductive system and the transition to a state of rest.

Table 2 presents the results of the nutritional value studies *D. trunculus*. The mollusk flesh is hydrated (more than 80%), the content (% a.d.m.) of protein substances is 74.8, carbohydrates – up to 12.2. The protein-water coefficient (PWC) of *Donax* flesh is 13.4%. For comparison: PWC of the flesh of commercial clam other species is on average for *Chamelea gallina* (Linnaeus, 1758) – 19.9%, *Anadara kagoshimensis* (Tokunaga, 1906) – 17.4%. The lipid-protein coefficient (LPC) is an indicator of the consistencies of the flesh of aquatic organisms: low values correspond to a denser and drier consistency of muscle tissue; for the flesh of *D. trunculus*, the LPC is 0.07, while the flesh of the gastropod *R. venoza* is tougher, namely 0.02.

Water		Caloric					
content, %	Protein	Ash	Lipids	Carbohydrates	value, kJ		
September, 2019							
85.2	11.00	1.00	0.80	1.80	244.1		
October, 2024							
84.6	12.10	1.30	0.61	1.50	250.7		

Table 2. Nutritional value of *D. trunculus*.

The coefficient of food saturation of D. trunculus meat is 0.17 conventional units, i.e. it belongs to low-saturated food raw materials, and in terms of energy value – to low-calorie ones.

Analysis of the amino acid composition of the proteins of the tissues of mollusks revealed the presence of all essential amino acids in *D. trunculus*, with valine and histidine being limiting (Figure 6, Table 3). It is possible to compensate for the content of limiting essential amino acids and increase the biological value of the finished product through the development of combined food products.

The tissues of all bivalve mollusks are characterized by a low lipid content, however, the fatty acid (FA) composition is dominated by essential polyunsaturated fatty acids (PUFA). Thus, gas chromatography of the total lipid content of *D. trunculus* showed a high percentage of n-3 polyunsaturated fatty acids (19.5-34.9% of the total FA) and a low level of total n-6 polyunsaturated fatty acids (5.6-9.7% of the total fatty acids); the PUFA/SFA ratio is 1.22-1.33 [16].

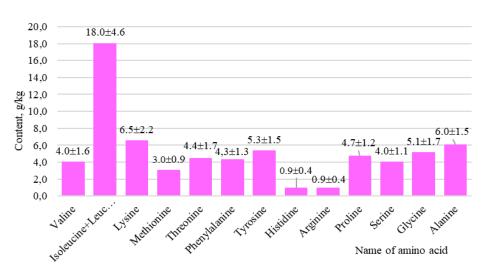


Fig.6. Amino acid protein composition of *D. trunculus* flesh (11.0 %).

Table 3. Amino acid score of proteins in *D. trunculus*.

Name of the	Score,	Amino acid score in the "ideal" protein, g/100 g	
amino acid	%	protein (FAO/WHO Recommendations, 2011)	
Val	91	4.0	
Ile + Leu	180	3.0 + 6.1	
Lys	123	4.8	
Met + Cys	119	2.3	
Phe + Tyr	213	4.1	
Thr	160	2.5	
Trp	_	0.66	
His	51	1.6	

Of the macroelements in the mineral composition of the mollusk meat, the following ones were determined (g/kg): potassium (4.0 \pm 0.8), calcium (6.0 \pm 1.3), magnesium (0.8 \pm 0.2), sodium (54.0 \pm 10.8) and phosphorus (0.13 \pm 0.01) %, with the recommended daily intake (RDI, mg/day): magnesium – 310-420, potassium – 4700, calcium – 1000-1200.

A high iodine content has been noted in the flesh of *D. trunculus* -0.042% (RDI: 130-200 mcg/day).

The values of the toxic element content in D. trunculus flesh did not exceed permissible levels (mg/kg): mercury - less than 0.0025, lead - less than 0.01, $arsenic - (1.31\pm0.33)$, $cadmium - (0.14\pm0.03)$.

4 Conclusion

The decline in commercial stocks of many valuable species of aquatic organisms in the world, including clams, due to intensive fishing or deterioration of environmental conditions, make work related to the study of their biology, nutritional value, and the possibility and conditions of cultivation increasingly in demand.

Of the two species of psammophilic mollusks, *D. trunculus* and *D. semistriatus*, typical inhabitants of the coastal zone of the Kerch Strait, *D. trunculus* predominates in 2023-2024. This species is considered by the authors as a more promising object for aquaculture and a source of valuable nutrients.

The obtained results of *D. trunculus* studies confirm the possibility of introducing shellfish meat into the composition of diets for dietary preventive nutrition. The biological value of the mollusk can be increased by farming clams in artificial conditions or by means of food combinatorics.

Continuation of work in this direction, as well as global experience in the cultivation of mollusks can serve as a scientific and practical basis for ensuring the consistent development of conchioculture of clam aquaculture in the Azov-Black Sea basin.

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