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SEX DIFFERENCES AND SEX IDENTIFICATION IN THE SMALL-SCALED SCORPIONFISH, *SCORPAENA PORCUS* (SCORPAENIDAE, SCORPAENIFORMES)

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Sex Differences and Sex Identification in the Small-Scaled Scorpionfish, *Scorpaena porcus* (Scorpaenidae, Scorpaeniformes). Peskov, V. N., Manilo, L. G. — Sex differences and the ability to determine the sex of *Scorpaena porcus* (Linnaeus, 1758) on morphological characters were studied in the population inhabiting coastal waters of the Black Sea near the southern coast of Crimea. These differences were revealed in size (females are larger than males) and in proportions of the body. It is found that variation of the absolute and relative values in plastic characters is higher in females compared with males. It is shown that in 92 % of individuals belonging to *S. porcus* we can determine the sex using morphological characters with a probability of 99.9–100 %.

Key words: sex, sex differences, plastic and meristic characters, scorpionfish, *Scorpaena porcus*, the Black Sea.

Introduction

Small-scaled scorpionfish, *Scorpaena porcus* (Linnaeus, 1758) belongs to the family Scorpaenidae, which is represented in the world fauna by 26 genera and 216 species. Issues on taxonomy, biology and ecology of these fishes are studied with varying degrees of completeness (Froese, Pauly, 2012). Biology of the small-scaled scorpionfish in some areas of the Mediterranean and the Black Sea have been studied relatively well (Pashkov et al., 1999; Bilgin, Çelik, 2009; Turan et al., 2009; La Mesa et al., 2010; Scarcella et al., 2011, etc.). However, despite the often quoted description of the morphology and information on some issues of morphological variability of the small-scaled scorpionfish (Svetovidov, 1964; Smirnov, 1986; Vasil'eva, 2007; Ferry et al., 2010, etc.), the relevance of the study of its intraspecific variability, including the sex difference, is obvious.

According to the literature data, males and females of *S. porcus* differ in the rate of growth, sexual maturation, life span, linear dimensions and proportions of the body. Sex differences in body size and proportions in the scorpionfish appear from the age of three (Smirnov, 1986). According to this author, at almost the same standard body length of sexually mature individuals of both sexes (SL_M = 12.27 cm and SL_F = 12.94 cm; t = 1.63 if P > 0.05), females in average have bigger relative size of the maximum body height (H/SL), preanal (PA/SL) and preventral (PV/SL) distances, height of spiny part of the dorsal fin (hD_1/SL), the height of head (hc/c), the largest head width (ic/c) and the height of the skin outgrowth above the eye (hs/c). The males in average have relatively bigger length of the head (c/SL) and the soft part of the dorsal fin (lD_2). All these differences are statistically highly significant (0.01 < P <0.001).

Sex differences on plastic and meristic characters in *S. porcus* were studied in more detail by Ferry et al. (2010) in the population of the Adriatic Sea. According to these authors, the average values of the relative sizes of the fins (LA/SL, LP/SL, LV/SL, and LC/SL) were significantly greater in males compared with females, which may indicate their higher motor activity. In females, on average, relative values of the standard body length (SL/TL), preventral (PV/SL) and postorbital (OLO/CL) lengths are significantly higher.

Considering all the above, we can conclude that the study of sex differences and the possibility of sex identification in scorpionfish on morphological characters seem very relevant, especially given the large amount of scientific material in museum collections. To date, the sex of *S. porcus* can only be determined by the gonads at the autopsy, which is prohibited by the rules of storage of museum collections. The aim of this paper is to study sex differences and the possibility of sex identification of *S. porcus* on plastic and meristic characters in the population inhabiting the coastal waters of the Black Sea near the southern coast of Crimea.

Material and methods

The basis of our work is the data on morphology of *S. porcus*, obtained in the processing of ichthyological collection in the National Museum of Natural History (NMNH) NAS of Ukraine. The material is represented by samples from the coastal waters of the Crimea. Eighty seven specimens of the scorpionfish were processed, including 14 males and 17 females, the sex of which is determined under the study of gonads, and 56 specimens without sex identification.

Morphological analysis was performed according to the methodic adopted for members of the family Scorpaenidae (Pravdin, 1966) under the scheme with minor changes: 1) TL — the total length of the body; 2) SL — standard body length; 3) LD — the length of the dorsal fin base; 4) LA — the length of the anal fin base; 5) LP — the length of the pectoral fin; 6) LV — the length of the pelvic fin; 7) LC — the length of the caudal fin; 8) pD — predorsal (antedorsal) length; 9) pD — postdorsal length (from the end of dorsal fin base to the beginning of the base of caudal fin rays); 10) l_{caud} — the length of caudal peduncle (from the end of the anal fin base before the base of caudal fin rays); 11) PA — preanal (anteanal) length; 12) PV — preventral (anteventral) length; 13) PP — prepectoral (antepectoral) length; 14) H — the maximum height of the body (on vertical of the third spiny dorsal fin); 15) h — height of the caudal peduncle; 16) CL — the length of the head from the start of the upper lip to the top edge of the gill cover; 17) O — the diameter of the eye; 18) PO — preorbital length; 19) IO — interorbital length; 20) OLO — postorbital length (from the posterior edge of the eye to the end of the horizontal spine on the gill cover); 21) mx — the length of the upper lip. The following meristic characters were also counted: D — the number of spiny rays in dorsal fin; d — the number of soft rays in dorsal fin; A — the number of spiny rays in anal fin; a — the number of soft rays in anal fin; P — the number of rays in pectoral fin; V — the number of spiny rays in ventral fin; v — the number of soft rays in ventral fin; sp. br. — the number of gill rakers on the first gill arch; squ — the number of bored scales in the lateral line; l.l. — the number of scales along the lateral side of the body.

In the study of body proportions were calculated relative values for 20 plastic characters in % of the standard body length — 15 (for all measurements except those for the head), in % of the head length — 5 (for the measurements of the head) and 1 — the ratio of the caudal peduncle height to the maximum height of the body.

The sex in scorpionfish was determined by the aggregate absolute values of plastic characters using a linear discriminant analysis, which is often used in the study of sex differences in vertebrates (Dzeverin, Lashkova, 2006). The Wilks' Lambda criterion was used for the statistical evaluation of the quality of models. A brief description of the algorithm and discriminant analysis of obtained results is shown below in the Results and discussion. Biological age of males was determined on the basis of study of their differentiation in terms of absolute values of 21 plastic characters (Peskov et al., 2013) using the hierarchical cluster analysis; Euclidean distance (DE) is calculated as the difference metric.

For each sample, standard statistical parameters in a number of variations are calculated: minimum (min), maximum (max) and the arithmetic mean (M) of character value, the error of the arithmetic mean (m), standard deviation (Sx) and the coefficient of variation (CV, %). Males and females were compared on an average amount of plastic and meristic characters using the Student's t-test. Trends of variability (CV series) in males and females were compared using the rank correlation coefficient for Kendal (τ). Student's t-test (Lakin, 1990) was used to compare the variability of characters (CV). Differences at the 5 % significance level were considered as statistically significant. All calculations were performed using the STATISTICA, v. 6.

Results and discussion

S e x i d e n t i f i c a t i o n i n *S. porcus* on absolute values of plastic characters was performed using the linear discriminant analysis, which is often used in the delineation of morphologically similar fish taxa (see, e. g., Manilo, Peskov, 2012 a, b).

On the first stage discriminant analysis was performed on absolute values of 21 plastic characters. Thus the whole sample (n = 87) was divided into three groups: 14 males, 17 females and 56 individuals whose sex was not determined. According to results of the first stage (Wilks' Lambda: 0.349; F = 4.35; $p < 0.0026$), all fourteen males (100 %) with known sex were defined as males with a probability of 55.4–99.8 %. Within the 17 females, 15 (88.2 %) of individuals were identified as female with a probability of 66.6–100 %, and 2 females (with a probability of 92.3 and 93.8 %) — as males. The probability of classifying 56 specimens whose sex has not been known to the male/female sample is ranged from 51.8 % to 100 %. Of the total sample (n = 87), only 14 specimens were identified as females, and 31 — almost uniquely the males (with a probability of 99.9–100 %).

On the second stage of the discriminant analysis, "training" samples of females (n = 14) and males (n = 31) have been formed from these specimens, the remaining 42 individuals were included in the analysis without the sex identification. The discriminant analysis

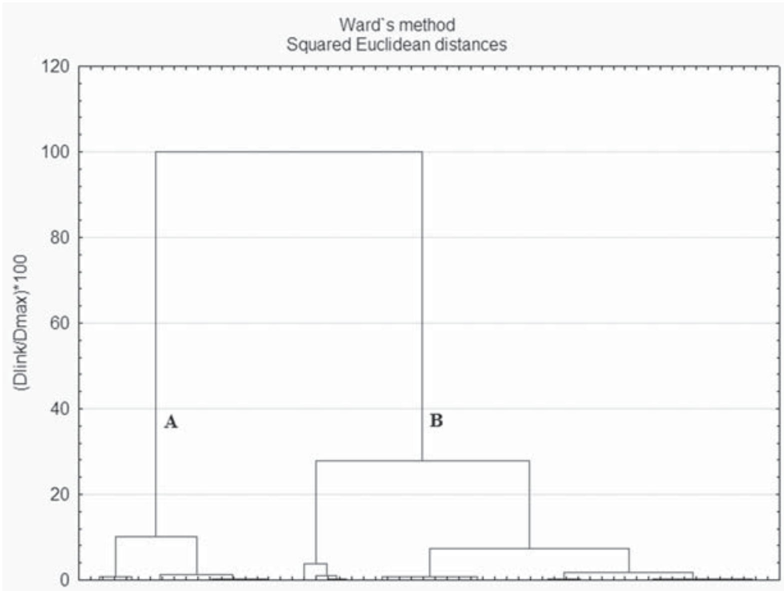

Fig. 1. Differentiation of males of the small-scaled scorpionfish on absolute values of 21 plastic characters.

was performed on the absolute values of 9 characters (LD, LA, LP, LC, PD, PA, PV, O and mx), selected in the first phase as a result of stepwise discriminant analysis procedure (inclusion method).

According to the results of the second phase (Wilks' Lambda: 0.3054; F = 9.35; $p < 0.0000$), 21 specimen was classified as female, 59 — as male with a probability of 99.9–100 %. These data allow us to say that in 80 (92.0 %) specimens of *S. porcus* sex was determined with almost 100 % probability. Probability of classifying 7 specimens (8 %) in the sample of males and females varies from 67 to 97 %, so we did not use them for further analysis.

Size and age composition of the samples. In a sample of females (n = 21) the total body length (TL) is ranged from 131.0 to 261.0 mm, in males (n = 58) — from 86.0 to 205.0 mm. According to the literature (Smirnov, 1986; Bilgin, Çelik, 2009; Ferry et al., 2010), all females may be considered as mature, whereas males, presented in the sample, are clearly immature. On this basis, it was decided to use the hierarchical cluster analysis, comparing the 58 individuals to each other by the absolute values of 21 plastic characters using the Euclidean metric (DE).

As seen in fig. 1, the sample of males is quite clearly divided into several subsamples. As a part of the first of them (subsample A) 18 individuals with a body length of 86 to 126 mm were differentiated from others; specimens with such sizes, according to the literature (Smirnov, 1986), can be seen as young (1–2 years) immature males. The second subsample (B, n = 40), appear to be composed of adult (3 years or older) adult males, body length of which varies from 131 to 205 mm. Adult males of this sample were used in comparison with adult females.

Sex differences in *S. porcus* were based on the average absolute (table 1) and relative values (table 2) of plastic characters using the Student's t-test. The following table shows the data only on characters, whose average values on sex differences are statistically significant.

The results of comparison of adult males and females in terms of absolute values of 21 plastic characters (table 1) show that females are larger than males, but the sex differences were statistically significant for the average values of only 11 characters (TL, SL, LD, PD, pD, PA, PP, H, h, CL and OLO). Females compared with males are more variable

Table 1. Comparison of males and females of the small-scaled scorpionfish from the Black Sea population in absolute values of plastic characters

Character	Females (n = 21)				Males (n = 40)				t
	Min	Max	M	Sx	Min	Max	M	Sx	df = 59
TL	131.0	261.0	170.95	35.29	131.0	205.0	153.79	16.76	2.11*
SL	106.0	204.0	133.86	27.60	102.0	159.0	118.43	12.95	2.43*
LD	65.0	118.8	84.00	16.14	62.3	101.9	74.69	8.54	2.47*
PD	34.8	67.9	46.10	8.32	35.0	54.2	40.52	4.40	2.87**
pD	7.8	19.3	11.82	3.16	7.2	14.8	10.25	1.70	2.12*
PA	69.4	151.1	91.90	22.27	69.2	116.7	80.68	10.51	2.18*
PP	41.2	86.4	52.73	11.68	40.4	68.8	47.22	6.14	2.02*
H	37.6	77.0	49.93	11.28	35.4	60.9	44.09	5.49	2.24*
h	10.8	21.5	14.09	3.00	10.8	16.7	12.53	1.42	2.25*
CL	44.8	88.2	57.83	11.88	43.0	71.5	51.55	6.35	2.26*
OLO	20.5	43.1	26.70	6.26	20.0	30.5	23.60	2.62	2.17*

Note. In tables 1–3: Min, M and Max — minimum, average and maximum values of the character; Sx — standard deviation (variance); t — Student's t-test; df — degrees of freedom. Statistically significant values of the t-test are marked with an asterisk (*$P < 0.05$; **$P < 0.01$; ***$P < 0.001$).

for 20 characters (fig. 2), and only diameter of eyes varies almost identical ($CV_M = 13.35$ % and $CV_F = 13.76$ %).

Trends of variability for 21 plastic characters on the coefficient of variation (CV) in males and females are insignificantly similar ($\tau = 0.37$; $P < 0.05$). The head length varies stronger (CV = 20.54 %) as compared to the eye diameter (CV = 13.76 %), while the differences in males in the variation of these characters (12.32 and 13.35 %, respectively) are absent.

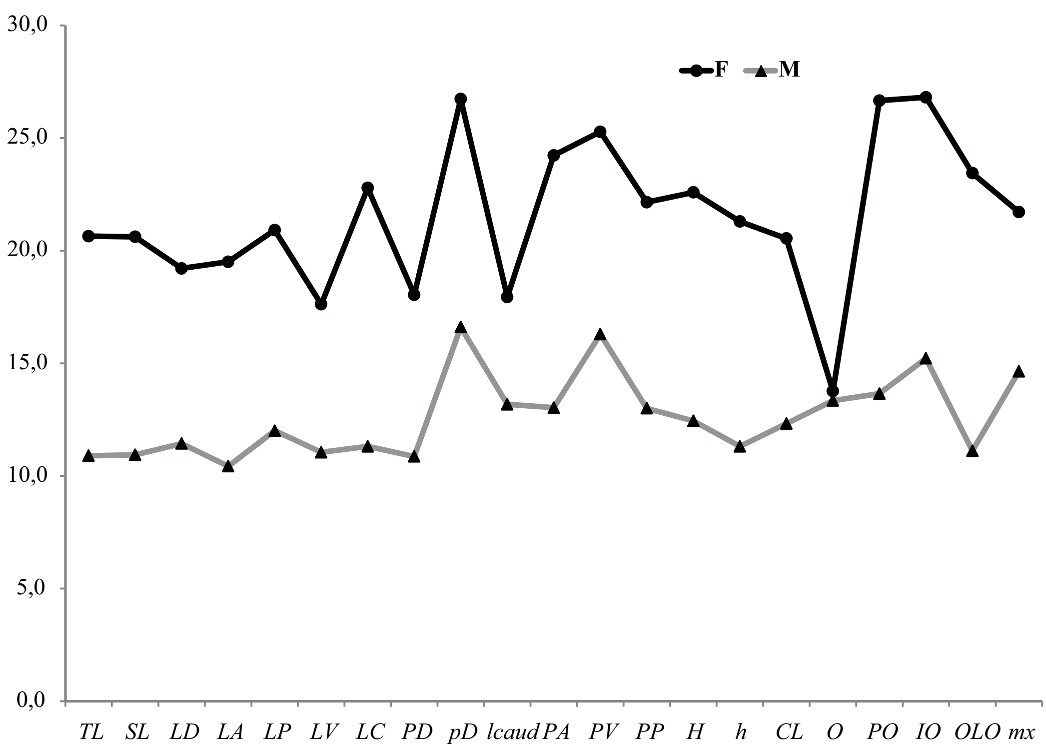

Fig. 2. Coefficient of variation (CV, %) for 21 plastic characters in adult males (M) and females (F).

Table 2. Comparison of males and females of the small-scaled scorpionfish from the Black Sea population in relative values of plastic characters

Character	Females (n = 21)				Males (n = 40)				t
	Min	Max	M	Sx	Min	Max	M	Sx	df = 59
SL/TL	76.2	84.0	78.46	1.955	75.2	78.5	76.80	0.840	3.73***
LA/SL	14.4	20.2	17.29	1.387	16.4	20.8	18.61	1.068	3.80***
LP/SL	24.8	29.3	27.42	1.400	25.7	32.9	28.70	1.588	3.24**
LV/SL	20.9	28.1	25.25	1.776	23.3	30.8	26.72	1.627	3.16**
LC/SL	19.1	30.6	27.41	2.935	27.4	33.0	30.23	1.425	4.15***
O/CL	18.5	26.3	22.13	2.111	19.3	26.5	23.30	1.785	2.15*
mx/CL	46.5	50.9	48.46	1.253	46.1	55.0	49.35	1.836	2.22*

Sex differences in body proportions are shown in table 2. The relative value of the standard body length (SL/TL) in average is significantly higher in females. Males have significantly greater relative length of the pectoral (LP/SL), abdominal (LV/SL) and caudal (LC/SL) fins, as well as anal fin base (LA/SL). In addition, they have in average greater relative diameter of the eye (O/CL) and the length of the upper jaw (mx/CL).

The proportions of the body are also more variable in females compared to males (fig. 3), although the sex difference in this case is much less than on linear dimensions. It is important to emphasize that in adult males and females of *S. porcus* body proportions are less variable in comparison with its linear dimensions (see figs 2 and 3). These data confirm the well-known fact that in the late ontogenesis in most of fishes body proportions are less variable than its linear dimensions. Therefore, in the comparative morphological studies of fish preference is usually given to the relative sizes of plastic characters compared with the absolute ones (Pravdin, 1966).

It is important to note that by comparing the males and females of the Black Sea small-scaled scorpionfish using the discriminant analysis and Student's t-test statistically significant sex differences were established both on individual characters and in their entirety. The data we obtained are in good agreement with the results of comparison of

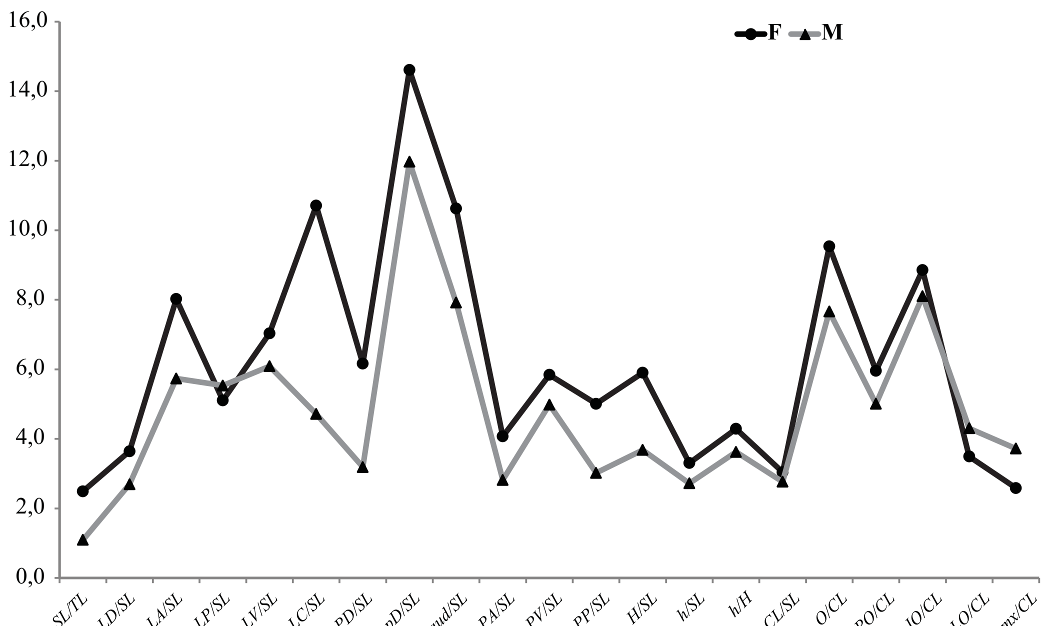

Fig. 3. Coefficient of variation (CV, %) for relative values of plastic features in adult males (M) and females (F).

Table 3. Comparison of males and females of the small-scaled scorpionfish from the Black Sea population in average values of meristic characters

Character	Females (n = 21)				Males (n = 40)				t
	Min	Max	M	Sx	Min	Max	M	Sx	df = 59
D	11	12	11.90	0.337	11	13	12.00	0.309	–1.08
d	10	11	10.24	0.439	9	11	10.14	0.467	0.79
A	3	3	3.00	0.000	3	3	3.00	0.000	—
a	5	7	6.05	0.464	5	7	6.02	0.344	0.21
P	15	17	16.33	0.621	16	18	16.36	0.570	–0.15
V	1	1	1.00	0.000	1	1	1.00	0.000	—
v	5	5	5.00	0.000	5	5	5.00	0.000	—
sp.br.	12	14	13.05	0.690	12	14	13.40	0.580	–2.03*
squ	24	28	25.71	1.224	23	28	25.40	1.364	0.90
l.l.	59	65	62.67	1.835	58	65	61.14	2.199	2.87**

male and female of *S. porcus* in population of the Adriatic Sea (Ferry et al., 2010). It is shown, first of all, the success of the use of linear discriminant analysis in determining the sex of *S. porcus* on plastic characters, and secondly, the high level of similarity in the manifestation of sex differences in two geographically isolated populations of this species.

Trends of variability for relative values of 21 plastic characters (series of coefficients of variation) in males and females are similar enough ($\tau = 0.64$; $P < 0.001$). The main differences boil down to the fact that LA/SL in females varies significantly stronger than LP/SL, while the both these characters in males can vary almost the same. The relative length of the caudal fin (LC/SL) in females is more variable compared to the pelvic fins (LV/SL). In males, we can see a completely opposite situation (fig. 3).

Sex differences of eight meristic characters in the sample we studied were not statistically significant (table 3). At the same time, the number of spiny rays in the anal (A = 3) and ventral (the V = 1) fins, as well as soft rays (v = 5) in the ventral fin is identical in all studied specimens (n = 87). These data are in good agreement with the data of other authors (Smirnov, 1986; Bilgin, Çelik, 2009; Ferry et al., 2010).

Sex differences were found on 2 meristic characters in the studied sample. Males have significantly larger number of gill rakers on the first gill arch, the females — the number of scales in the lateral line (see table 3). In the first case, the sex differences are determined by the fact that the maximum number of gill rakers (14) on the first gill arch was observed in 45 % of males and only 24 % of females. The maximum number of scales in the lateral line (64 and 65) was observed in 48 % of females and only 15 % of males. However, given the relatively small amount of studied samples, marked differences must be considered as a preliminary result. Information about the existence of sex differences in meristic characters are absent in the available literature.

Conclusion

It is found in our study that in a population of *S. porcus*, inhabiting the coastal waters of the Black Sea near the Crimea, the absolute values of 21 plastic characters are in average higher in adults mature females compared with males of the same age. Sex differences were statistically significant for the average values of eleven plastic characters (TL, SL, LD, PD, pD, PA, PP, H, h, CL and OLO). The relative value of the standard body length (SL/TL) in average is significantly higher in females. Males have significantly greater relative length of the pectoral (LP/SL), ventral (LV/SL) and caudal (LC/SL) fins and anal fin base (LA/SL). In addition, they have an average greater relative eye diameter (O/CL) and the length of the upper jaw (mx/CL). Linear dimensions of the body in both males and females are more variable than its proportions. At the same time, the variability of absolute and relative char-

Table 4. The coefficients of classification functions for the sex identification in the small-scaled scorpionfish

Characters	Function 1, females	Function 2, males
TL	0.284	–0.199
LC	–0.174	0.967
LA	–0.010	0.895
LP	0.053	0.364
Constant	–23.261	–16.611

acters of females is significantly larger than those in males. Using the linear discriminant analysis, it is possible to determine the sex in 92 % of specimens belonging to *S. porcus* on morphological characters with a probability of 99.9–100 %. Using the hierarchical cluster analysis, immature (TL = 86–126 mm) and mature (TL = 131–205 mm) males were defined in the sample on absolute values of 21 plastic characters.

Sex identification in the scorpionfish. As a result of incremental discriminant analysis, 4 morphometric characters (TL, LC, LA and LP) were selected, and the reliability of sex determination for the small-scaled scorpionfish reaches 91.2 % (taking into account all above-mentioned characters).

To determine the sex of each specimen according to the values of morphometric characters we have developed two classification functions, which in general terms can be expressed in a linear regression equation:

$$y = a+b_1 x_1+ b_2 x_2 + b_3 x_3 + b_4 x_4,$$

where: y — the value of classification function, a — free term of the equation, or constant; b_1, b_2, b_3, b_4 — coefficients for classification functions, calculated from 4 characters; x_1, x_2, x_3 and x_4 — the values of these characteristics in a given individual.

The values of constants and coefficients for each function are shown in the table 4. Substituting these values and the values of characters for the specific individual to the appropriate classification functions, it is necessary to calculate the value of the two functions for the sex identification in this individual. At the same time, this specimen should be determined hereto sex, value of the classification function for which was higher.

Using the developed classification functions, seven individuals of the scorpionfish were diagnosed as males (sex of these specimens was not determined by the results of the second half stage of the discriminant analysis). Values of classification functions in seven other specimens (6 females and 1 male) were very close, so it is necessary to do an autopsy in 8.8 % of the individuals and study their gonads for the final sex determination.

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