

Shape variation of the Indian Ocean slender mudskipper, *Scartelaos tenuis* (Day, 1876) from the Persian Gulf and Gulf of Makran (Gobioidei: Gobiidae)

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Abstract

The shape diversity among populations of *Scartelaos tenuis* (Day 1876) from five stations (194 specimens) in the Persian Gulf, Strait of Hormuz (Qeshm Island), and the Gulf of Makran were investigated. The shape variation was assessed by geometric morphometrics, using 14 morphological landmarks. The two sexes were not distinguishable by the shape. Fish samples were classified into two distinct groups, the fish from the Persian Gulf (Heleh and Asaluyeh), and the specimens of Qeshm Island and the Gulf of Makran (Chahoo, Dargahan and Jask). The Persian Gulf and the Gulf of Makran are characterized by specific environmental parameters of surface waters, including temperature and salinity. The shape discrepancies of *Scartelaos tenuis* specimens from different stations are probably because of ecological parameters of the Persian Gulf and Gulf of Makran. Our results suggest that the observed pattern of morphological variation is almost some kind of polyphenism induced by distinct environmental factors.

Keywords: Geometric morphometrics, Environmental factors, Polyphenism,
Scartelaos tenuis, Persian Gulf, Gulf of Makran

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Introduction

Three species of mudskippers are distributed in the Persian Gulf and Gulf of Makran, including *Boleophthalmus dussumieri*, *Periophthalmus waltoni*, and *Scartelaos tenuis* (Ghanbarifardi and Malek, 2007). Based on recent molecular phylogenetic trees for mudskippers, the Indian Ocean slender mudskipper, *Scartelaos tenuis* (Day, 1876) is included in the “*Periophthalmus* lineage” of gobionelline-like gobies (Gobiidae) and sister taxa for *Boleophthalmus* genus; moreover, ecomorphological adaptations of three Iranian mudskippers have been examined by geometric morphometric method (Ghanbarifardi *et al.*, 2016; Polgar *et al.*, 2017). *S. tenuis* specimens from Chabahar and Qeshm Island have been compared using meristic and morphometric characteristics; this study proves some differences between fish from two stations regarding length-weight relationship and condition factor (Hashemi *et al.*, 2015). Geometric morphometric and traditional morphometric methods have been employed to investigate the morphologic diversity among various populations of *P. waltoni* (Ghanbarifardi *et al.*, 2014a,b). These two studies prove that *P. waltoni* populations from the Persian Gulf differ from those of the Gulf of Makran regarding the morphological characteristics. Age, growth (Abdoli *et al.*, 2010) and feeding habits (Abdoli *et al.*, 2012) of *Scartelaos tenuis* have been surveyed. However, there is no study to compare different populations of this species in a wide geographical range. Geometric morphometrics (GM) is a method to compare the shape of organisms (Rohlf and Marcus, 1993). Two- or three-

dimensional coordinates of biologically definable landmarks are used for Landmark-based geometric morphometric methods. Superimposition approaches remove non-shape variation (variation in position, orientation, and scale of the specimens) in configurations of landmarks by overlaying them and employing some optimization criteria. Generalized Procrustes Analysis (GPA) is usually used to superimpose landmark configurations and remove the non-shape variation. After the non-shape diversity has been eliminated, the shape variables could be compared via multivariate statistics, and graphical representations of shape (thin-plate spline transformation grids) are generated for comparison. GM presents shape diversification, and it is employed as a substitute to the linear measurements, counts, and ratios used in conventional morphometrics (Adams *et al.*, 2004). GM has been used to study fish ecology, population biology and stock discrimination (Silva, 2003; Costa and Cataudella, 2007; Mostafa *et al.*, 2010). It has also been established that several environmental elements have transiently or permanently affected the morphological traits of fish. Temperature (Loy *et al.*, 1996a), fish body condition (Fitzgerald *et al.*, 2002), adaptations to specific habitat types (Langerhans *et al.*, 2003), predation risk (Eklov and Svanback, 2006), and salinity (Loy *et al.*, 1996b) are among the most important ecological factors.

The Persian Gulf is a limited waterbody, linked to the Gulf of Makran via the narrow Strait of Hormuz (Fig. 1); therefore, its water transmission with the Gulf of Makran is restricted. The average

depth of the Persian Gulf is only ~35 m. The surface water of the Persian Gulf experiences broad seasonal temperature alterations and superior salinity readings because of the low rainfall and high evaporation values (Yao, 2008). On the other part, the Gulf of Makran is widely connected to the Indian Ocean and much deeper than the Persian Gulf (~1000 m). Enough circulation, direct connection with the Indian Ocean and confined extension of shallow regions are three characters of the Gulf of Makran that make ecological conditions of this waterbody steadier than those of the Persian Gulf (Reynolds, 1993).

This survey explores shape variation among populations of *S. tenuis* from five stations in the Persian Gulf, Strait of Hormuz (Qeshm Island) and Gulf of Makran, utilizing GM.

Materials and methods

One hundred and ninety four specimens of *S. tenuis* (standard length > 70 mm) were collected from five stations (Heleh 39,

Asaluyeh 34, Chahoo 38, Dargahan 39, Jask 44) along the Iranian coasts of the Persian Gulf and Gulf of Makran, in February 2013 (Fig. 1, Table 1). The specimens are transferred to the Zoological Museum of Ferdowsi University of Mashhad (ZMFUM, Table 1). Specimens were caught by hand net during low tide and preserved in 5% formalin. The urogenital papilla is pointed in males and spatulate in females, and this character is used to sex determination of all fish (Miller, 1986; Ghanbarifardi *et al.*, 2014a). To evaluate the trustworthiness of this method, 20 accidentally chosen specimens were also sex determined by dissection. The left side of each sample was photographed using a Canon SX150 digital camera mounted on a bench-top camera stand. Fish images were converted into TPS files with tpsUtil v. 1.38 (Rohlf, 2006), and 14 definable homologous landmarks (Fig. 2) were digitized from each image using tpsDig v. 2.17 (Rohlf, 2013).

Table 1: Sampling coordinates, area, number and call numbers of *Scartelaos tenuis* specimens collected at five stations.

Station	Coordinates	Area	Specimens	ZMFUM call numbers
Heleh	29° 14.93' N 50° 40.72' E	Persian Gulf	13 females, 26 males	104hs-142hs
Asaluyeh	27°26'N 52°40'E	Persian Gulf	15 females, 19 males	3as-36as
Chahoo	26° 41.47' N 55° 28.75' E	Qeshm Island (Strait of Hormuz)	18 females, 20 males	143qws-149qws 18qws-49qws
Dargahan	26° 58.39' N 56° 4.85' E	Qeshm Island (Strait of Hormuz)	19 females, 20 males	50qes-88qes
Jask	25° 33.85' N 58° 10.61'E	Gulf of Makran	21 females, 23 males	100js-118js 74js-99js



Figure 1: Sampling stations. 1: Heleh, 2: Asaluyeh, 3: Chahoo, 4: Dargahan, 5: Jask.

Landmark data from each specimen was aligned using Generalized Procrustes Analysis (GPA) for superimposition (Rohlf, 1999) with MorphoJ v. 1.04a (Klingenberg, 2011). The GPA method translates, rotates, and scales all landmarks to eliminate the non-shape variation and size factor (Zelditch *et al.*, 2004). After Procrustes analysis, discriminant function analysis (DFA) was carried out with MorphoJ v. 1.04a to assess any shape difference between sexes. Canonical variate analysis (CVA) of landmark data was performed to evaluate any significant dissimilarity in shape among fish samples. CVA was conducted using SPSS 20.0 (© IBM); for CVA, 34 specimens of each station were used. To illustrate body shape variation, thin-plate spline transformation grids and lollipop plot (vectors showing transpositions from mean landmark positions) are generated with MorphoJ v. 1.04a. In this method, the shape changes from mean landmark positions are depicted as functions of their position along CV axes (e.g. CV1).

Results

DFA method could not distinguish significant difference in shape between females and males of *S. tenuis* (Table 2). Therefore, all specimens of each population were considered as a whole and CVA was conducted. The thin-plate spline (transformation grids) and lollipop plot along CV1 were used to depict the deformation in shape. The shape of the snout (Landmark 1), the eye situation (landmarks 3 and 4) and the position of the first and second dorsal fins and anal fin (landmarks 5, 6, 7, 8, 10 and 11) are disfiguring for the most (Fig. 3). The initial three canonical variates (CVs) achieved 66.0%, 19.1% and 10.6% of total variance, correspondingly. CV1 obviously detaches Chahoo, Dargahan (Qeshm Island) and Jask (Gulf of Makran) from Heleh and Asaluyeh (Persian Gulf) (Fig. 4). Totally, 91.8% of the grouped specimens of *S. tenuis* from different stations were properly categorized by CVA (Table 3).

Table 2: DFA of male and female specimens of *Scartelaos tenuis* from different sampling stations, based on GM data.

Heleh		Predicted group membership		
P-value: 0.1628		female	male	Total
Count	female	8	5	13
	male	8	18	26

Asaluyeh		Predicted group membership		
P-value: 0.2801		female	male	Total
Count	female	7	8	15
	male	9	10	19

Chahoo		Predicted group membership		
P-value: 0.0913		female	male	Total
Counts	female	9	9	18
	male	10	10	20

Dargahan		Predicted group membership		
P-value: 0.8694		female	male	Total
Counts	female	7	12	19
	male	11	9	20

Jask		Predicted group membership		
P-value: 0.6968		female	male	Total
Counts	female	8	13	21
	male	12	11	23

Table 3: CVA of *Scartelaos tenuis* from different populations. The percentage of specimens correctly classified to each station is written in bold. Overall, 94.3% of the specimens from different stations were accurately classified.

	Heleh	Asaluyeh	Chahoo	Dargahan	Jask
Heleh	85.3	11.8	2.9	0.0	0.0
Asaluyeh	14.7	85.3	0.0	0.0	0.0
Chahoo	0.0	0.0	97.1	2.9	0.0
Dargahan	0.0	0.0	11.8	79.4	8.8
Jask	0.0	0.0	0.0	11.8	85.3

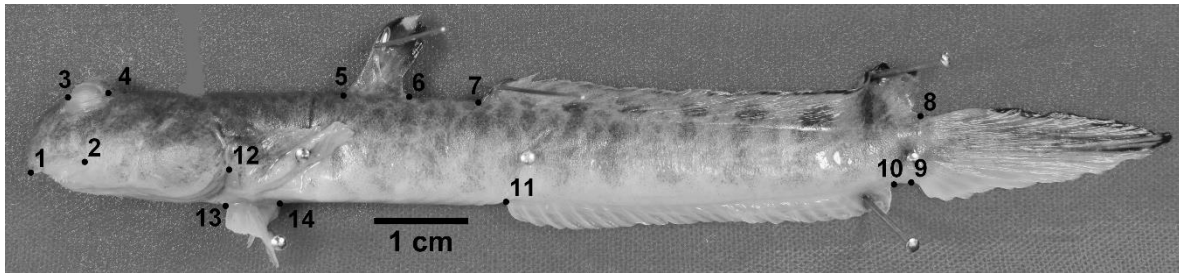


Figure 2: 14 morphological landmarks shown on *Scartelaos tenuis*. 1: anterior tip of the snout on the upper jaw. 2: rictus. 3 and 4: eye's anterior and posterior edges, correspondingly. 5 and 6: anterior and posterior points of insertion of the first dorsal fin, respectively. 7 and 8: anterior and posterior points of insertion of the second dorsal fin, sequentially. 9: ventral point of insertion of the caudal fin. 10 and 11: posterior and anterior points of insertion of the anal fin, respectively. 12: Ventral point of attachment of the left branchiostegal membrane. 13 and 14: anterior and posterior points of insertion of the left pelvic fin (landmarks are named after Ghanbarifardi *et al.*, 2014a).

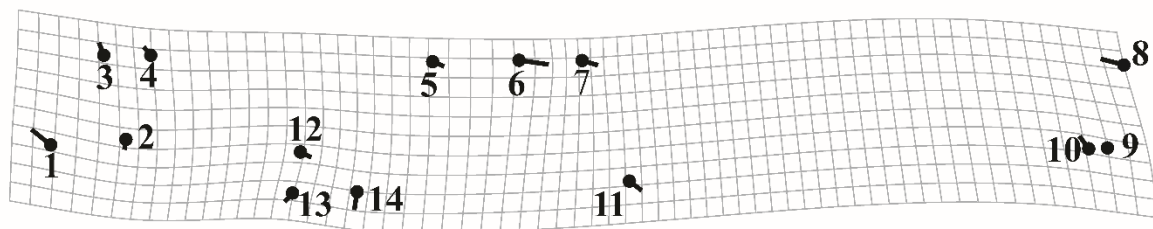


Figure 3: Transformation grids and lollipop plot of all specimens along CV1 axis, accounting for 66.0% of total morphometric variance.

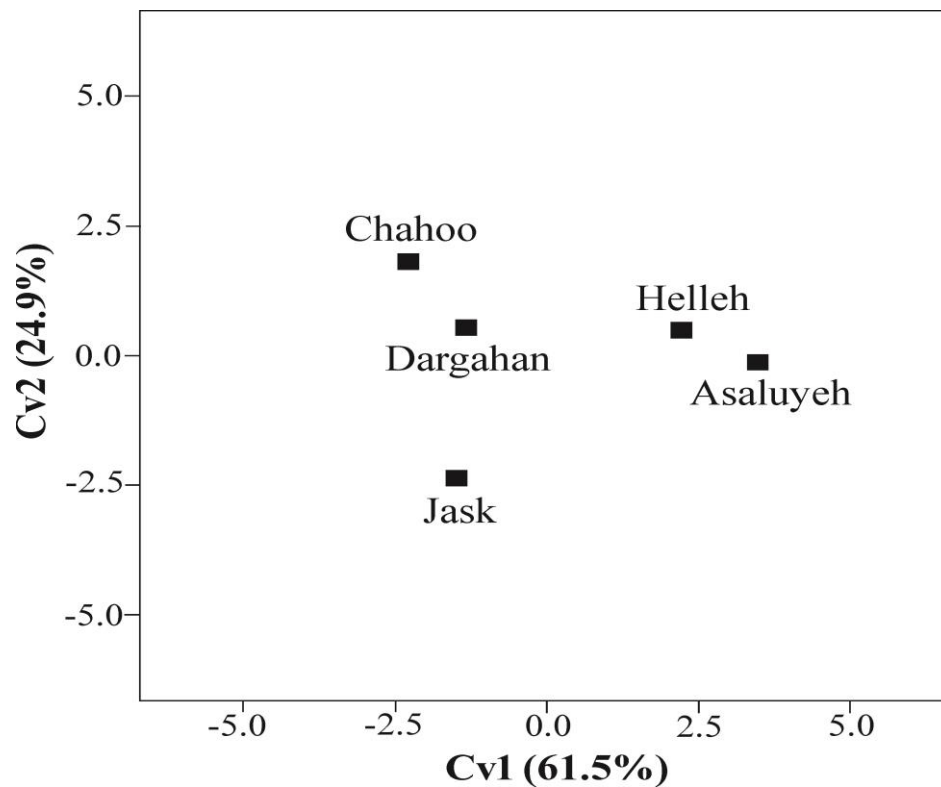


Figure 4: Scatter plot of canonical variate analysis for the group centroids of *Scartelaos tenuis* from the five stations.

Discussion

The shape diversities between *S. tenuis* specimens separate western Persian Gulf populations (Heleh and Asaluyeh) from the other three populations (Chahoo, Dargahan and Jask) (Fig. 4). *S. tenuis* specimens from Chabahar and Qeshm Island are distinguishable regarding morphometric and meristic characteristics (Hashemi *et al.*, 2015). These results are also coherent with recent morphological studies conducted on the other sympatric mudskipper *Periophthalmus waltoni* Koumans, 1941 (Ghanbarifardi *et al.*, 2014a,b). Moreover, investigations on *Lutjanus johnii* (Bloch, 1792) (otolith shape) and *Leptodius exaratus* (H. Milne Edwards, 1834) (morphometric characteristics) exhibit discrepancies between populations from the Persian Gulf and the Gulf of Makran (Sadighzadeh *et al.*, 2014; Afkhami *et al.*, 2016). *Aphanius dispar* populations from the Persian Gulf and the Gulf of Makran have been compared using morphometric and otolith characteristics (Teimori *et al.*, 2012a,b). The results from these studies proved that both coastal and landlocked populations of the Gulf of Makran are distinctive from those of the Persian Gulf. The mentioned studies assume otolith characters to be under genetic control; therefore, the phenotype diversities among populations of this species would be a result of geographical isolation and not related to environmental variations. Such morphological variation (Fig. 3) is almost some kinds of polyphenism induced by distinct environmental factors (e.g. temperature and salinity); notwithstanding populations of these intertidal species (*S. tenuis* and *P. waltoni*)

are genetically connected (West-Eberhard, 1989). Surface waters of the Persian Gulf (Heleh and Asaluyeh) have different physical conditions compare to the Gulf of Makran (Jask) (Thoppil and Hogan, 2010; Yao, 2008; Pous *et al.*, 2004; Swift and Bower, 2003; Reynolds, 1993). Chahoo and Dargahan are located in Qeshm Island (Strait of Hormuz); however, mudskipper populations collected from these two stations are probably influenced by the currents entering the Persian Gulf from the Gulf of Makran (Fig. 1). Therefore, the specimens from Qeshm Island are similar to those of Jask (Gulf of Makran) (Fig. 4). Such different conditions might induce intraspecific phenotypic plastic responses. In fact, temperature and salinity can both affect growth and shape (Meador and Kelso, 1990; Loy *et al.*, 1996a,b; Turan *et al.*, 2005).

Mudskippers can tolerate wide ranges of salinity and temperature; however, changing these environmental factors could alter some behavioral and physiological characteristics of these fish. Temperature and salinity affect oxygen consumption and Na^+ , K^+ , ATPase activity in mudskippers, respectively (Clayton, 1993). In specific, a study on *P. waltoni* in Iran showed that increasing salinity significantly induces elevation of RBC, Hct and Hb. (Soltanian *et al.*, 2016). These mentioned physiological responses could influence the shape of fish in different stations with distinct temperature and salinity readings. Future phylogeographic molecular studies on *S. tenuis* could determine if these shape differences bear genetic variation.

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