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Research Article

A comparative study on the body shape of *Alosa caspia* (Teleostei, Clupeidae) populations in the southern Caspian Sea basin using geometric morphometric analysis

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Abstract: Geometric morphometric method was used to examine body shape variations among the three populations of the Caspian shad, *Alosa caspia*, in the southern Caspian Sea coasts of Iran. A total of 289 *A. caspia* specimens were caught from three localities, from the west to the east including Guilan (Anzali), Mazandaran (Sari) and Golestan (Miankale), respectively. Shape data was extracted using 15 landmark points on 2-D pictures of specimens. The PCA, DFA, CVA/MANOVA analyses and cluster analysis (CA) based on Euclidean square distances were used to examine shape differences among the three populations. Significant differences were found among the populations of *A. caspia* in terms of body shape. Cluster analysis showed separation of Mazandaran populations from the other provinces. Results revealed that the studied populations are divided into two categories; a group with large body and head depth and short and wide caudal peduncle and the other one with shallow body, small head and longer caudal peduncle. Observed differences in the head shape of Mazandaran population of *A. caspia* could be indirectly related to the feeding resources. Changes in head and mouth shapes can be considered as reflective of differences in selection of food items and direction of feeding. The obtained body shape properties can provide a shape-based identification key for *Alosa caspia*, which is useful for fisheries and stock management or conservation programs.

Keywords: Shape variation, Discriminant, Clupeiformes, Landmark, Stock, Shad.

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Introduction

The study of morphological characters with the aim of defining or characterizing fish stock units has a great importance in ichthyology (Bektas & Belduz 2009). The morphometric characters are particularly important where the differences are mostly attributed to the environmental parameters rather than genetic differentiation (Bektas & Beldu 2009). Geographical isolation of populations and interbreeding can lead to morphometric variations among populations, and this morphometric variation can provide essential basis for population differentiation (Bookstein 1991). For species like the Caspian shad, investigation on morphometric variations is essential as this species is widely distributed in the Caspian Sea. The family Clupeidae is found in warmer marine waters with some anadromous or permanent freshwater residents. This family has about 200 species in 56 genera worldwide (Eschmeyer & Fong 2011; Coad 2017), with eight reported species in the Caspian Sea

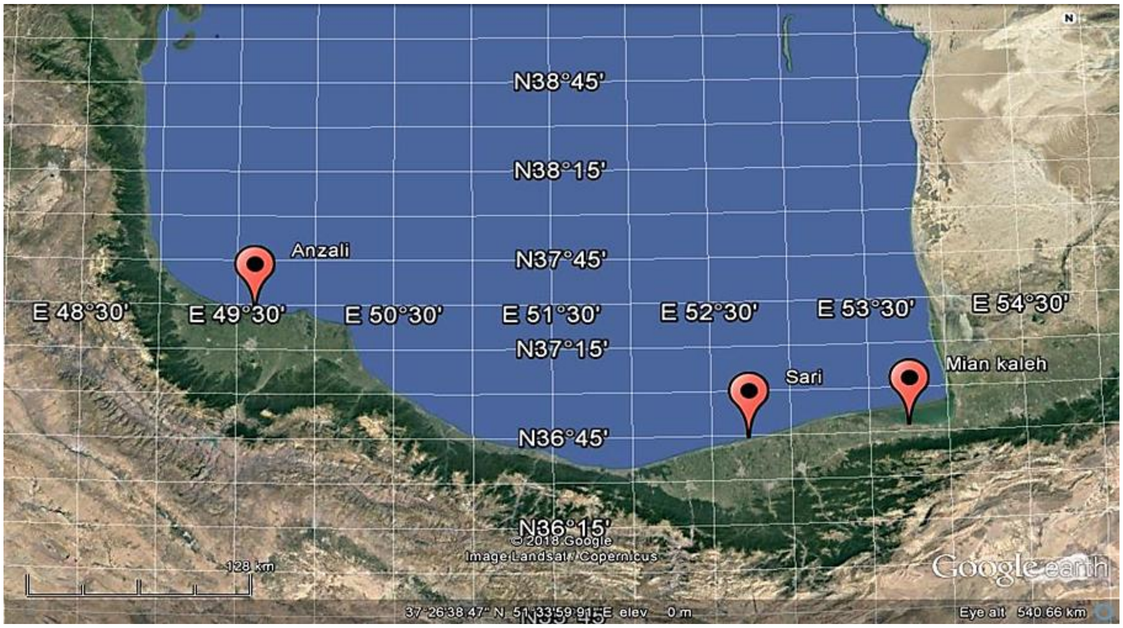

Fig.1. Sampling stations in the south Caspian Sea (north of Iran).

(Esmaeili et al. 2014) and 11 species in the inland waters of Iran (Esmaeili et al. 2017). The Caspian shad, *Alosa caspia,* found in the Caspian and Black seas and the subspecies *Alosa caspia caspia,* Eichwald 1838, is found mostly in the western half of the Caspian Sea basin but is the most widely distributed subspecies, found throughout almost the whole sea (Coad 2017). This subspecies was the most important subspecies in the herring family in the Caspian Sea (Coad 2017).

The body shape differences of populations are considered as essential steps in process of speciation (Balon 1993; Margurran 1998). Geometric morphometric is a modern approach to analyze the shape of body (Bookstein 1991). Application of this method in fish body shape has been reported (Loy et al. 1999) and has also been considered as a useful tool to evaluate fish populations. This method describes organisms' body shape in terms of x and y (and also z) coordinates, obtained from a set of landmark points (Loy et al. 1999). Landmark points are defined as homologous points that bear information on the geometry of biological forms (Bookstein 1991). This method quantifies changes in shape, and patterns of morphometric variations within and between groups if each individual deviates from an average shape, i.e. the consensus configuration (Cadrin & Friedland 1999). Morphological studies are strong and applicable for determining discreteness of the similar species and extensively used to identify differences between fish populations (Mousavi-Sabet & Anvarifar 2013). Geometric morphometrics (GM), a quantitative approach to analysis shape, is widely applied to compare and determine shape variations of biological structures (Sansom 2009). One important advantage of the GM approach in relation to traditional ones is that GM does not need to decide a priori which measurements are likely to display differences (Rohlf & Marcus 1993). Despite traditional approaches in GM, data is obtained from the coordinates of landmark points (Rohlf & Marcus 1993; Adams et al. 2004), which are morphological points of specimens that are of biological interest (Richtsmeier et al. 2002). GM has been used in various studies in fish population biology such as stock identification and discrimination (Cadrin & Friedland 1999). This method, which allows the study of shape and size, offers powerful analytical and graphical tools for the quantification and visualization of morphological variation within and among organisms (Slice 2007).

Body shape is a major component of an

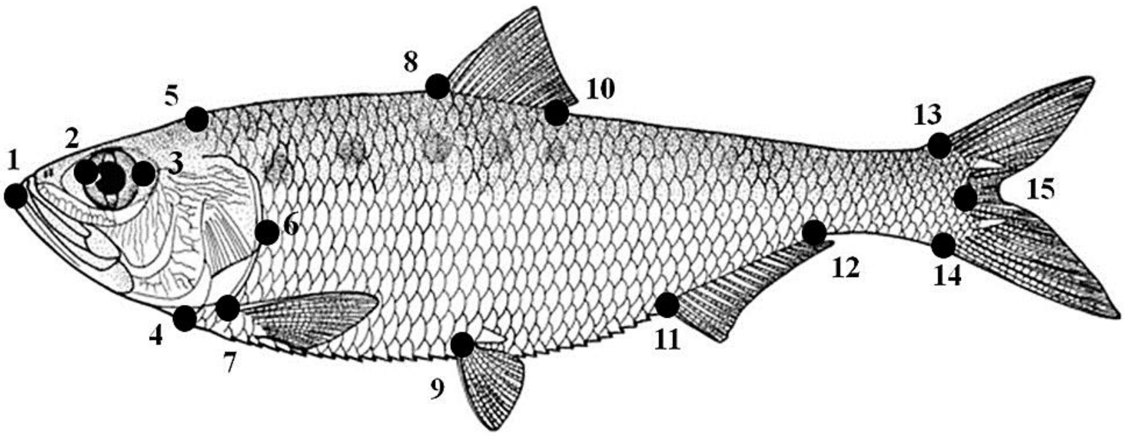

Fig.2. Defined landmark points to extract body shape. 1. Anterior tip of the premaxilla; 2. Front of the eye; 3. End of the eye; 4. Beginning of the scales at the dorsal side; 5. The lower beginning of operculum; 6. End of operculum; 7. Base of the pectoral fin; 8. Base of the pelvic fin; 9 & 10. Anterior and posterior insertion of the dorsal fin; 11 & 12. Anterior and posterior insertion of the anal fin; 13. Upper margin of caudal peduncle; 14. Lower margin of caudal peduncle; 15. End of the medial region of caudal peduncle.

organism's phenotype, and it bears important traits of its biological characters such as locomotors performance, feeding efficiency, vulnerability to predators, and reproductive success (Gill et al. 2003). Fish body shape can be the result of evolutionary adaptations to environmental pressures (Gatz 1979; Winemiller 1991), particularly, food collection and hydrodynamic conditions (Matthews 1998) making feasible more efficient utilization of available resources and improving fitness and performance (Pianka 1994). Therefore, the body shape differences of populations are considered as essential steps in the process of speciation (Balon 1993; Margurran 1998). Most members of the genus *Alosa* Linck, 1790 in Iran exhibit relatively different body form. Hence, this study was conducted to assess intraspecific body shape among three *Alosa caspia* (Eichwald, 1838) populations with visualization techniques afforded by the GM approach.

Materials and Methods

A total of 289 specimens of *A. caspia* were randomly collected by beach seine from three fishing regions along the southern Caspian Sea coasts, including provinces of Golestan, Mian kaleh (36°54′10.89″N, 53°48′48.33″E; 122 individuals), Mazandaran, Sari (36°48′04.63″N, 53°02′07.50″E; 86 individuals) and Guilan, Anzali (37°29′29.86″N, 49°27′39.59″E; 81 individuals), in the fishing season during the years 2015-2016 (Fig. 1). The specimens were kept on freezer and transported to the laboratory for further examinations. After thawing, each individual was photographed for digital analysis.

Laboratory works: The left side of the specimens (with dorsal and anal fins were held erected using pins) were photographed using digital camera (Canon Power Shot SX 30 IS). Fifteen homologous landmark-points were defined and digitized on 2-D images using tpsDig2 software version 2.16 (Rohlf 2004). The landmark-points were selected to the best represent the external shape of the body (Fig. 2). The landmark-points were chosen at the specific points, in which a proper model of fish body shape was extracted (Bookstein 1991).

Data analysis: The extracted landmark-points (body shape data) were submitted to a generalized procrustes analysis (GPA) to remove non-shape data in PAST software. These morphological differences may be solely related to body shape variation and not to size effects which were successfully accounted for by allometric transformation. On the other hand, size related characteristics play a predominant role in morphometric analysis and the results may be erroneous if not adjusted for statistical analyses of data (Tzeng 2004). In the present study, the size effect was removed successfully by procrustes action

Table 1. Mahalanobis distance analysis for the three populations of *Alosa caspia*.

Station	Golestan	Guilan
Guilan	2.52	
Mazandaran	2.88	1.47

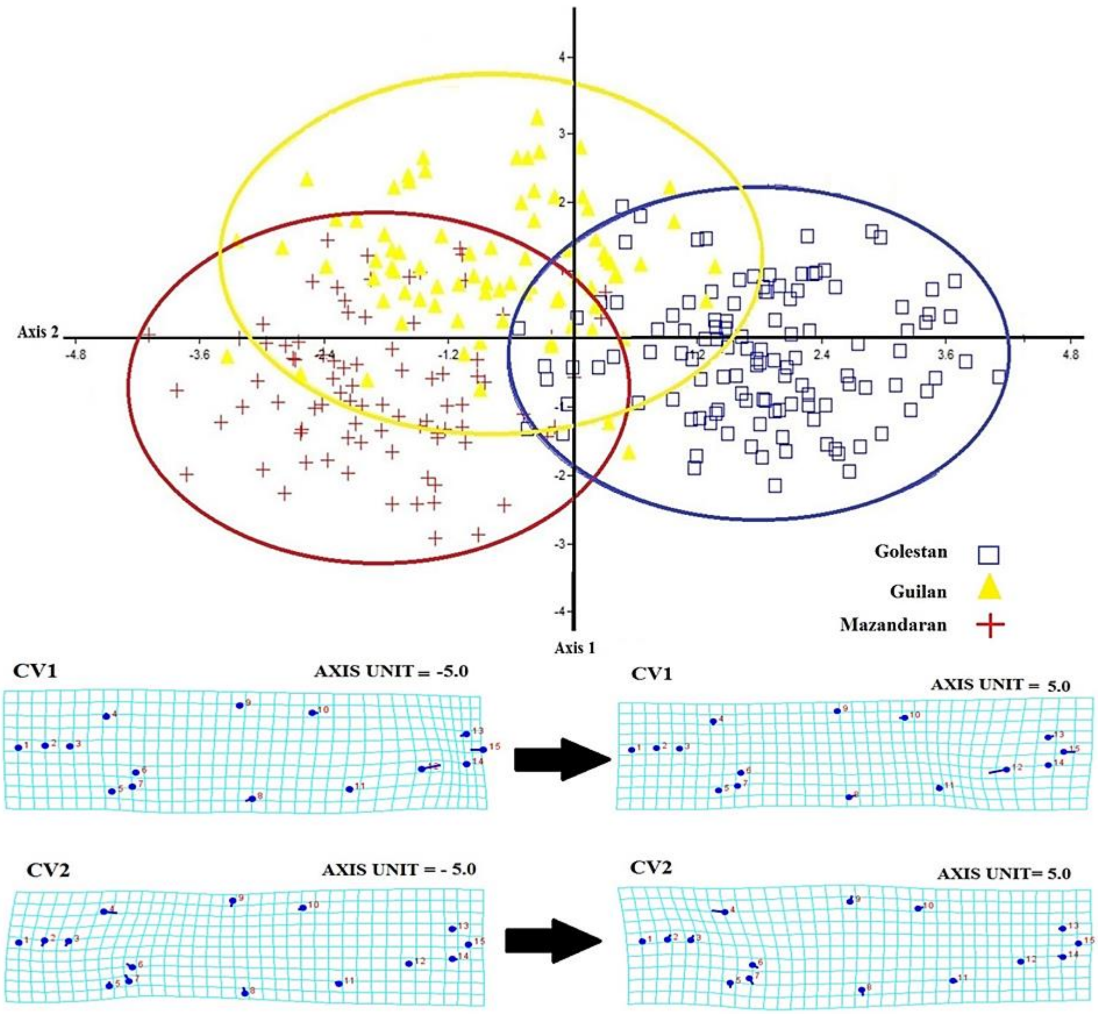

Fig.3. The results of Canonical variant analysis (CVA) of three populations of the species *Alosa caspia* body shape with respect to the first two canonical variables.

in PAST software.

Principal component analysis (PCA) was performed to summarize the variation among the specimens as few dimensions as possible. Canonical variant analysis CVA/MANOVA was accomplished to investigate power of distinction among different species by PAST statistical and MorphoJ software (Klingenberg 1998). As a complement to discriminant analysis, morphometric distances among the three localities were inferred to cluster analysis by adopting the Euclidean square distance as a measure of dissimilarity method as the clustering algorithm (Sneath & Sokal 1973). The patterns of taxon's body shape were illustrated in transformation grids in relation to consensus configuration of all specimens presented, depicting relative shape differences among the populations.

Results

The PCA analysis for all specimens explained 80.51% of shape variations by the first two PC axes extracted from the variance-covariance matrix

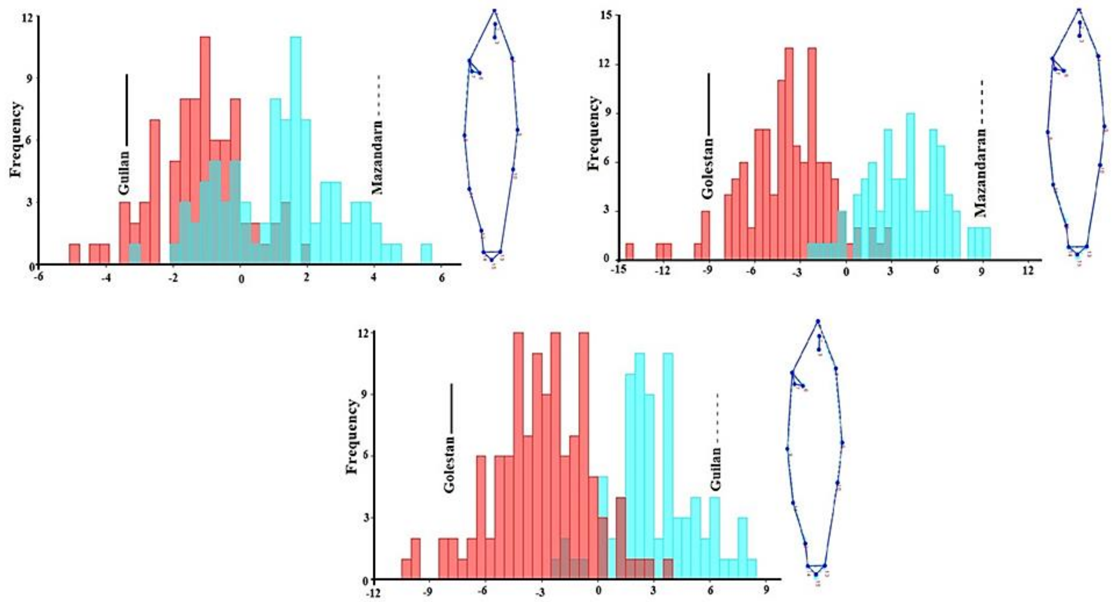

Fig.4. Histogram of discriminate analysis (DA) functions of three populations of the species *Alosa caspia* body shapes in different locations with respect to the first two canonical variables.

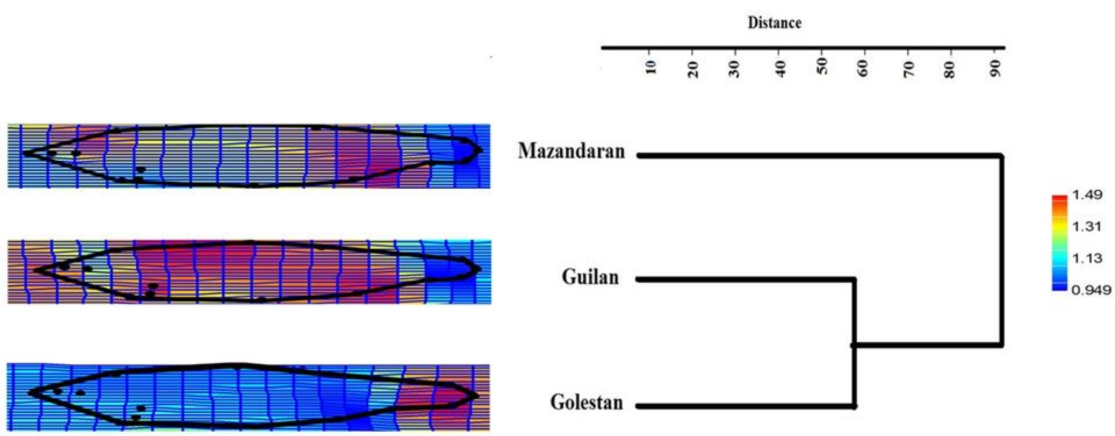

Fig.5. Dendrogram derived from cluster analysis of morphometric variables on the basis of Euclidean distance of the three populations of *Alosa caspia* in Iran. Mean shape of species in relation to consensus shape of the three populations of *Alosa caspia* are represented.

(PC1=57.46% and PC2=23.05%). The CVA/ MANOVA revealed significant different in body shape among the populations (P<0.05). The CVA plot revealed the clear separation between Golestan population from Mazandaran and Guilan populations (Fig. 3). The Mahalanobis distances among the groups are represented in Table 1.

There was a high degree of separation in body shape characters between Golestan and the other two provinces, with a slight degree of separation between Guilan and Mazandaran populations (Fig. 4). Discriminant analysis (DA) on the relative warps classified 84.1% in origin data and 81.3% in cross validation of specimen into the correct groups (Table 2). The dendrogram derived from cluster analysis of Euclidean square distances showed that the three populations of *A. caspia* were distinct from each other in terms of morphometric characters (Fig. 5).

The grids of Figure 5 indicate elongated head and snout, short and deep caudal peduncle and large body depth in midsection for Mazandaran *A. caspia* population, but shallow body depth, small head, short

Table 2. Classification matrix showing the number and percentage of individuals that were correctly classified.

	Mazandaran	Golestan	Guilan	Total
Original (%)				
Mazandaran	84.9	2.30	12.8	100.0
Golestan	2.50	88.5	9.00	100.0
Guilan	13.6	9.90	76.5	100.0
Cross-validate (%)				
Mazandaran	82.6	2.30	15.1	100.0
Golestan	3.30	87.7	9.00	100.0
Guilan	18.5	11.1	70.4	100.0

snout and longer caudal peduncle for Guilan *A. caspia* population. Shallow body depth, small head, short snout and longer anal fin base were observed in Golestan *A. caspia*. Body shape of *A. caspia* in Golestan and Guilan showed very similar body shape.

Discussion

Food is the major source of all the nutrients a host needs. Nutrients are chemical substances needed for Among several geometric approaches to morphometrics, the landmark based is one the most widespread and the best understood in its mathematical and statistical properties (Bookstein 1996; Small 1996; Dryden & Mardia 1998). The present study on the three populations of *A. caspia* species shape variation demonstrated significant variation among the populations. Differences among the studied species can be divided into two categories; (1) large body and head depth and short caudal peduncle in *A. caspia* population of Mazandaran province and (2) shallow body depth and longer caudal peduncle in the other populations. The causes of morphological differences among populations are often quite difficult to explain (Bookstein 1991). It has been suggested that the morphological characteristics of fishes are determined by genetic, environment and the interaction between them (Kohestan-Eskandari et al. 2014). Morphological divergence in shape among conspecific populations may be caused by a phenotypic plastic response to local environmental differences or be an adaption resulting from differences in resource use (Langerhans et al. 2003). The environmental factors prevailing during the early development stages, when the individual's phenotype is more amenable to environmental influence (Eschmeyer & Fong 2011). The phenotypic variability may not necessarily reflect population differentiation at the molecular level (Bookstein 1991). Apparently, different environmental conditions can lead to an enhancement of pre-existing genetic differences, providing a high interpopulation structuring (Eschmeyer & Fong 2011; Mousavi-Sabet & Anvarifar 2013).

Results revealed an association between body depth and short and deep caudal peduncle among the populations. This result is consistent with the previous studies on other fishes such as darters (Page & Swofford 1984) in which the situation was explained as an adaptation for inhabiting in stream riffles. In contrast, the relatively shallow bodies may be useful for benthic pool fishes as pointed out by Wood & Bain (1995). Adult morphology may be determined by a diversifying selection and ecological adaptation. Observed differences in head shape of Mazandaran population of *A. caspia* with other *A. caspia* populations can be indirectly related to the feeding resources (Andersson et al. 2005). Changes in head and mouth shapes can be considered as reflective of differences in selection of food items and direction of feeding (Langerhans et al. 2003). Costa and Cataudella (2007) found that shape differences were related to trophic ecology for several species of the family Sparidae, thus indicating local adaptation and possibly ecological radiations (Schluter & McPhail 1992; Langerhans et al. 2003). If shape is related to either environmental influences on larval development (Cadrin & Silva 2005) or diversifying selection and ecological adaptation at a trophic level (Costa & Cataudella 2007), then spatially or latitudinally different environmental factors (e.g., temperature and resource availability) may explain the variations in body shape among the studied species. The importance of geographical isolation on producing morphological differentiation in fish species is well-known (Yamamoto et al. 2006). According to Cadrin & Silva (2005), the geographic variation in adult morphology for yellowtail flounder *Limanda ferruginea* may be explained by differences in ontogenetic rates among local populations if morphology is a product of ontogenetic history. Different prey resources for larvae and juveniles may lead to different adult body shapes, suggesting that there is a phenotypic plastic response to resource availability (Wimberger 1990). This might be a case in *A. caspia* population of Mazandaran province. Our results showed significant body shape differences among the populations of *Alosa caspia* in Iranian Caspian Sea waters. Hence, it can be interpreted as differences at the species level suggesting possibility of well-differentiation species based on their body shape. These results provide an identification key based on body shape to the population of the species *Alosa caspia*, which can be useful for further fisheries and stock managements or conservation programs in the region.

Acknowledgments

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References

Adams, D.C.; Rohlf, F.J. & Slice, D.E. 2004. Geometric morphometrics: ten years of progress following the "revolution." Italian Journal of Zoology 71: 5-16.

Andersson, C.B.; Hughes, S.O.; Fisher, J.O. & Nicklas, T.A. 2005. Cross-cultural equivalence of feeding beliefs and practices: The psychometric properties of the child feeding questionnaire among Blacks and Hispanics. Preventative Medicine 41: 521-531.

Balon, E.K. 1993. Dynamics of biodiversity and mechanisms of change: a plea for balanced attention to form creation and extinction. Biological Conservation 66: 5-16.

Bektas, Y. & Belduz A.O. 2009. Morphological variation among Atlantic horse mackerel, *Trachurus trachurus* populations from Turkish coastal waters. Journal of Animal and Veterinary Advances 8: 511-7.

Bookstein, F.L. 1991. Morphometric tools for landmark data. Cambridge University Press, Cambridge, UK. 435 p.

Bookstein, F.L. 1996. Biometrics, biomathematics and the morphometric synthesis. Bulletin of Mathematical Biology 58: 313-365.

Cadrin, S.X. & Friedland, K.D. 1999. The utility of image processing techniques identification. Fisheries Research 43: 129-139.

Cadrin, S.X. & Silva, V.M. 2005. Morphometric variation of yellowtail flounder. ICES Journal of Marine Science 62: 683-694.

Coad, B.W. 2017. Freshwater fishes of Iran. Available at: http://www.briancoad.com (accessed on 17 June 2017).

Costa, C. & Cataudella, S. 2007. Relationship between shape and trophic ecology of selected species of Sparids of the Caprolace coastal lagoon (central Tyrrhenian Sea). Environmental Biology of Fishes 78: 115-123.

Dryden, I.L. & Mardia, K.V. 1998. Statistical Shape Analysis. New York: John Wiley and Sons.

Eschmeyer, W.N. & Fong J.D. 2011. Animal biodiversity: An outline of higher level classification and survey of taxonomic richness. Zootaxa 3148: 26-38.

Esmaeili, H.R.; Coad, B.W.; Mehraban, H.R.; Masoudi, M.; Khaefi, R.; Abbasi, K.; Mostafavi, H. & Vatandoust, S. 2014. An updated checklist of fishes of the Caspian Sea basin of Iran with a note on their zoogeography. Iranian Journal of Ichthyology 1: 152-184.

Esmaeili, H.R.; Mehraban, H.; Abbasi, K.; Keivany, Y. &

Coad, B. 2017. Review and updated checklist of freshwater fishes of Iran: Taxonomy, distribution and conservation status. Iranian Journal of Ichthyology 4(Suppl. 1): 1-114

Gatz, A.J., Jr. 1979. Community organization in fishes as indicated by morphological features. Ecology 711-718.

Guill, J.M.; Hood, C.S. & Heins, D.C. 2003. Body shape variation within and among three species of darters (Perciformes: Percidae). Ecology of Freshwater Fish 12: 134-140.

Klingenberg, C.P. 1998. Heterochrony and allometry: the analysis of evolutionary change in ontogeny. Biological Reviews 73: 79-123.

Kohestan-Eskandari, S.; Anvarifar, H.; Mousavi-Sabet, H.; Yousefi, M. & Khanzade, M.; 2014. A morphology-based hypothesis for homeward migration success and population differentiation in the anadromous *Kutum Rutilus kutum* (Pisces: Cyprinidae) along the southern Caspian Sea, Iran. Folia Zoologica 63(3): 151-160.

Langerhans, B.R.; Laydon, C.A.; Langerhans, A.K. & Dewitt, T.J. 2003. Habitat-associated morphological divergence in two Neotropical fish species. Biological Journal of the Linnaean Society 80: 689-698.

Loy, A.; Boglione, C. & Cataudella, S. 1999. Geometric morphometrics and morpho-anatomy: a combined tool in the study of sea bream (*Sparus aurata*) shape. Journal of Applied Ichthyiology 15: 104-110.

Magurran, A.E. 1998. Population differentiation without speciation. Philosophical Transactions of the Royal Society of London. Series B: Biological Sciences 353: 275-286.

Matthews, W.J. 1998. Patterns in Freshwater Fish Ecology. New York, Chapman and Hall. 757 p.

Mousavi-Sabet, H. & AnvariFar, H. 2013. Landmark-based morphometric variation between *Cobitis keyvani* and *Cobitis faridpaki* (Pisces: Cobitidae), with new habitat for *C. faridpaki* in the southern Caspian Sea basin. Folia Zoologica 62: 167-175.

Page, L.M. & Swofford, D.L. 1984. Morphological correlates of ecological specialization in darters. Environmental Biology of Fishes 11: 139-159.

Pianka, E.R. 1994. Evolutionary ecology. New York, Harper Collins. 397 p.

Richtsmeier, J.T.; Deleon, V.B. & Lele, S.R. 2002. The promise of geometric morphometrics. The Yearbook of Physical Anthropology 45: 63-91.

Rohlf, F.J. & Marcus, L.F. 1993. “A revolution in morphometrics,”Trends in Ecology and Evolution 8(4): 129-132.

Rohlf, F.J. 2004. TpsDig–Thin Plate Spline Digitize, Version 1.4 [Computer software]. Stony Brook: Dept. of Ecology and Evolution, State Univ. of New York.

Sansom, R.S. 2009. Phylogeny, classification, and character polarity of the Osteostraci (Vertebrata). Journal of Systematic Paleontology 7: 95-115.

Schluter, D. & McPhail, J.D. 1992. Ecological character displacement and speciation in sticklebacks. American Naturalist 140: 85-108.

Slice, D.E. 2007. Geometric Morphometrics. Annual Review of Anthropology 36: 261-281.

Sneath, P.H. & Sokal, R.R. 1973. Numerical taxonomy. The principles and practice of numerical classification.

Small, C. 1996. The statistical theory of shape (p. 227). New York: Springer.

Tzeng, T.D. 2004. Morphological variation between populations of spotted Mackerel Scomberaus *tralasicus* of Taiwan. Fisheries Research 68: 45-55.

Wimberger, P. H.; 1990. Plasticity of fish body shape: the effects of diet, development, family, and age in two species of Geophagus (Pisces: Cichlidae). Biological Journal of the Linnean Society 45: 197-218.

Winemiller, K.O. 1991. Ecomorphological diversification in lowland freshwater fish as assemblages from five biotic regions. Ecological Monographs 343-365.

Wood, B.M. & Bain, M.B. 1995. Morphology and microhabitat use in stream fish. Canadian Journal of Fisheries and Aquatic Sciences 52: 1487-1498.

Yamamoto, S.; Maekawa, K.; Tamate, T.; Koizumi, I.; Hasegawa, K. & Kubota, H. 2006. Genetic evaluation of translocation in artificially isolated populations of white-spotted charr (*Salvelinus leucomaenis*). Fisheries Research 78: 352-358.

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مقاله پژوهشی

مطالعه مقایسه‌ای شکل بدن جمعیت‌های شگ‌ماهی خزری *Alosa caspia* (ماهیان استخوانی عالی: شگ ماهیان) در سواحل جنوبی دریای خزر با استفاده از ریخت‌سنجی هندسی

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چکیده: روش ریخت‌سنجی هندسی جهت بررسی تفاوت‌های شکل بدن ۳ جمعیت از شگ ماهی خزری (*Alosa caspia*) در سواحل جنوبی دریای خزر در ایران مورد استفاده قرار گرفت. در مجموع ۲۸۹ نمونه از شگ ماهی خزری از غرب به شرق قسمت جنوبی دریای خزر، گیلان (انزلی)، مازندران (ساری) و گلستان (میانکاله) جمع‌آوری شدند. داده‌های شکلی از نقاط ۱۵ لندمارک روی تصاویر دو بعدی نمونه‌ها استخراج شدند. آزمون‌های مولفه‌های اصلی PCA، تابع تشخیص DFA، CVA/MANOVA و آزمون خوشه ای CA بر اساس فواصل مربع اقلیدسی جهت بررسی تفاوت‌های شکلی میان ۳ جمعیت مورد استفاده قرار گرفتند. تفاوت‌های معنی‌داری در رابطه با شکل بدن بین جمعیت‌های مختلف شگ‌ماهی خزری مشاهده شد. آنالیز خوشه ای تمایز بین جمعیت مازندران از دیگر استان‌ها را نشان داد. نتایج نشان داد که جمعیت مورد مطالعه به دو دسته تقسیم می‌شوند: یک گروه با ارتفاع زیاد بدن و سر و ساقه دمی کوتاه و پهن و گروه دیگر با ارتفاع کم بدن، سر کوچک و ساقه دمی طویل. تفاوت‌های مشاهده شده در شکل سر جمعیت شگ ماهی خزری در مازندران با جمعیت‌های دیگر شگ ماهی خزری می تواند به‌طور غیر مستقیم به منبع غذایی بستگی داشته باشد. تغییرات شکل سر و دهان می تواند انعکاسی از تفاوت در انتخاب مواد غذایی و رژیم غذایی در نظر گرفته شود. مشخصات به‌دست آمده بر اساس شکل بدن شگ ماهی خزری می‌تواند یک کلید شناسایی فراهم آورد، که برای مدیریت ذخایر شیلاتی و برنامه های حفاظتی مفید است.

کلمات‌کلیدی: تغییرات شکلی، تمایزدهنده جمعیت، شگ ماهی شکلان، لندمارک، ذخیره، پوزانک.