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## Habitat effects on morphological plasticity of Saw-belly (*Hemiculter leucisculus*) in the Zarrineh River (Urmia Lake basin, Iran)

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### ABSTRACT

Saw-belly (*Hemiculter leucisculus*) is a non-native species, which unfortunately spreads in many aquatic ecosystems of Iran. This species has probably introduced to Zarrineh River with commercially cyprinid species from the Caspian Sea basin. The adverse effects of this species on native species can lead to ecological hazards. Therefore, the present study was conducted to investigate the effect of environmental factors on the shape variations of Saw-belly (*H. leucisculus*) in the Urmia Lake. Hence, a total of 120 Saw-belly specimens were caught from the Zarrineh River. Specimens were photographed using a digital camera and the landmarks put on two-dimensional images using TpsDig2 software. General Procrustes Analysis (GPA) was used to remove the effects related to the figure (including size, position and rotation). The significant relationships were found between environmental factors and shape data using Procrustes regression. The results showed that environmental factors including temperature, altitude and velocity had a significant effect on the body shape of Saw-belly. The results can be considered by conservationists and environmentalists to monitoring of this newly introduced fish.

**Key words:** Saw-belly, body shape, geometric morphometrics, environmental factors, Urmia Lake

## Introduction

Morphological traits are used to identify differences among fish populations (Cheng et al., 2005; Yakubu & Okunsebor, 2011). In addition, these characters can also be applied in stock and hybrid identification, analysis of life history and ecomorphological studies (Klingenberg et al., 2003; Letcher, 2003; Andersson et al., 2006; Liasko et al., 2012). Therefore, morphological traits are still one of the main research approaches used in ichthyology and fishery sciences (Petrtyl et al., 2014).

Body shape variation between fish stocks can provide a basis about stock structure (Begg et al., 1999). Strong effects of environmental factors seems to be common in body shape changes of fish (Staszny et al., 2013). Many studies, for example (Currens et al., 1989) and (Marcil et al., 2006) have stated that body shape in fishes is labile to environmental conditions, including water velocity, discharge, water depth, temperature, type of food and etc. Thus, differences in abiotic and biotic factors of the habitats of fish species lead to morphological differences. In other words, differences in morphology characteristics among fish populations result from a combination of adaptive plastic responses to different environmental conditions (Robinson & Parsons, 2002; Keeley et al., 2007). Plastic responses are the ability of a fish

to express different phenotypes in response to environmental conditions (Dudley & Schmitt, 1996; Andersson et al., 2006; Karlsson et al., 2010).

The statistical analysis of body changes is one of the important factors for the proper management of a species (Manjurul Alam et al., 2014). Today, the analysis of distant features obtained by image processing techniques is used for fish identification (Mohaddasi et al., 2013). Also, body shape changes of fish can be easily obtained by two-dimensional image analysis (Petrtyl et al., 2014; Mojekwu & Anumudu, 2015). Geometric morphometric is a powerful and beneficial tool which can complete other approaches to body shape changes of fish using imaging techniques (Cadrin & Friedland, 1999). This method has several advantages compared to other morphometric methods for the distinction and analysis of body shape (Kerschbaumer & Sturmbauer, 2011).

Urmia Lake is an endorheic basin (closed drainage basin) in Iran. This basin is considered as an important ecosystem in northwestern Iran (Ghasemi et al., 2015). Urmia Lake basin is fed by 13 permanent rivers. Eimanifar & Mohebbi (2007) and Stevens et al. (2012) reported that the Zarrineh and Simineh Rivers provide nearly half of its modern inflow.

The Zarrineh River passes the cities of Shahin-Dezh, Kashavar and Miandoab and into Urmia Lake (Karimi et al.,

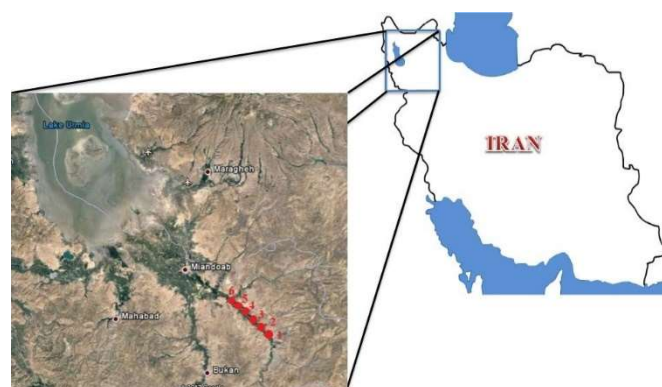
2014). According to previous studies, there is a vast number of fish that live in this river. Sharpbelly or Saw-belly (*Hemiculter leucisculus* Basilewsky, 1855) is tropical freshwater and brackish water fish, belonging to the cyprinidae family, which has been found in the Zarrineh River (Radkhah & Eagderi, 2015; Froese & Pauly, 2016). It usually found in rivers, shallow waters over sandy bottom, mountain streams and pools (Serov *et al.*, 2006). *H. leucisculus* is an exotic fish in the Urmia Lake basin. This species might be accidentally transferred to interior basins of Iran mixed in with Chinese carp (Bauer, 1991).

Introductions of invasive species into new ecosystems are a serious threat to other living species (Carlton, 2001). These species have a negative effect on a local ecosystem and native species through genetic deterioration (hybridization), competition, gene pollution and spread of disease or parasites (Esmacili *et al.*, 2014; Radkhah *et al.*, 2016). Therefore, Saw-belly may also have undesirable effects on endemic fish species (Pazooki *et al.*, 2011). On the other hand, detailed information on impacts of this species in non-native ecosystems (including Urmia Lake) is lacking. Therefore, it is essential to investigate the biological and ecological characteristics of *H. leucisculus* in Urmia Lake basin.

Hence, the present study applied a geometric morphometric approach to investigate the shape variation of Saw-belly (*H. leucisculus*) in the Zarrineh River (Urmia Lake basin) to better understand the biological and ecological aspects of this species in this important basin of Iran.

## Materials and Methods

For the present study, a total of 120 adult specimens of Saw-belly were caught on September 2015 from six separate stations of Zarrineh River, Urmia Lake basin (see Figure 1). The specimens after anesthesia with clove powder fixed in 10% formaldehyde solution and transferred to the laboratory for detail examination. Then, the lateral surface of their left side was photographed using digital 6 megapixel camera



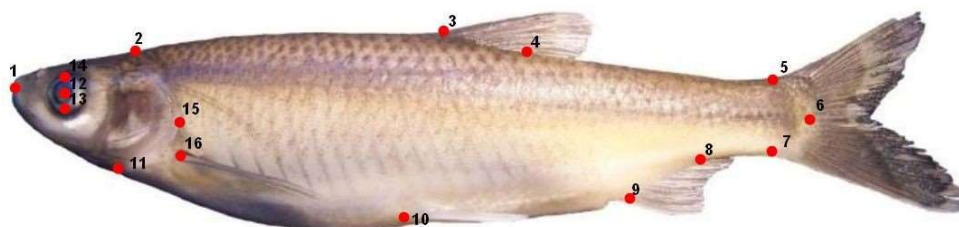
**Figure 1.** Map showing locations of the sampling sites in Zarrineh River from the Urmia Lake basin, Iran.

(Kodak) and sixteen landmark points were digitized on two-dimensional images using TpsDig2 software (Figure 2) to extract body shape data in (GM).

The landmark data submitted into R software space using the Geomorph package. General Procrustes Analysis (GPA) was used to remove the effects of size, rotation and positions of landmark-points from data. In addition, the environmental factors, including water temperature, altitude, discharge, flow velocity and water depth were measured immediately after sampling (Table 1) and then submitted as a matrix to analyze in R software space. The significant relationship between the environmental parameters and body shape of fish was studied (with 999 iteration) using Procrustes regression.

## Results

In the present study, body shape changes of *H. leucisculus* in Zarrineh River were investigated. Generalized Procrustes Analysis (GPA) of Saw-belly (*H. leucisculus*) is shown in Figure 3. GPA mathematically removed nonshape variation due to scale, size and position. In addition, GPA reduced induced covariance among landmarks. The results of the geometric morphometric analysis revealed that fish have undergone the various morphological changes during



**Figure 2.** The 16 landmark points identified on *H. leucisculus* for the geometric morphometric analysis: (1) anterior tip of snout at upper jaw; (2) upper tip of the operculum, (3) anterior base of the dorsal fin; (4) posterior base of the dorsal fin; (5) dorsal base of the caudal fin; (6) distal tip of the hypural bones; (7) ventral base of the caudal fin; (8) posterior base of the anal fin; (9) anterior base of the anal fin; (10) insertion of pelvic fin; (11) posterior most point of maxillary; (12) eye center; (13) lower point of the eye socket; (14) upper point of the eye socket; (15) posterior tip of the operculum; (16) insertion of pectoral fin.

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**Table 1.** Geographic location and environmental factors of sampling stations in the Zarrineh River from Urmia Lake basin, Iran.

| Stations  | Latitude and longitude | Temperature | Altitude | Discharge | Velocity | Depth |
|-----------|------------------------|-------------|----------|-----------|----------|-------|
| Station 1 | 36 39 57 / 58 N        | 15          | 1348     | 44        | 1.54     | 101   |
|           | 46 32 45 / 88 E        |             |          |           |          |       |
| Station 2 | 36 39 32 / 93 N        | 15          | 1347     | 44        | 0.84     | 55    |
|           | 46 33 5 / 88 E         |             |          |           |          |       |
| Station 3 | 36 38 13 / 36 N        | 16          | 1350     | 44        | 1.26     | 40    |
|           | 46 33 20 / 90 E        |             |          |           |          |       |
| Station 4 | 36 35 33 N             | 16          | 1349     | 44        | 1.12     | 55    |
|           | 46 33 4 / 59 E         |             |          |           |          |       |
| Station 5 | 36 35 28 / 46 N        | 17          | 1348     | 46        | 1.05     | 63    |
|           | 46 33 22 E             |             |          |           |          |       |
| Station 6 | 36 42 46 / 7 N         | 19          | 1349     | 44        | 1.20     | 103   |
|           | 46 9 48 / 68 E         |             |          |           |          |       |

**Table 2.** The results of procrustes regression between the environmental factors and body shape data of *H. leucisculus* in the Zarrineh River, Urmia Lake basin.

| Parameter   | df | SS       | MS        | Rsqr     | F      | Z      | P. value | Sig. |
|-------------|----|----------|-----------|----------|--------|--------|----------|------|
| Temperature | 1  | 0.004003 | 0.0040031 | 0.070108 | 4.0964 | 3.3119 | 0.01     | c    |
| Altitude    | 1  | 0.003528 | 0.0035280 | 0.061788 | 3.6103 | 2.6996 | 0.01     | c    |
| Discharge   | 1  | 0.002747 | 0.0027468 | 0.048106 | 2.8108 | 1.9310 | 0.06     | NS   |
| Velocity    | 1  | 0.002356 | 0.0023557 | 0.041257 | 2.4106 | 1.7129 | 0.04     | c    |
| Depth       | 1  | 0.001467 | 0.0014672 | 0.025697 | 1.5014 | 1.2240 | 0.16     | NS   |
| Residuals   | 44 | 0.042998 | 0.0009772 |          |        |        |          |      |
| Total       | 49 | 0.057099 |           |          |        |        |          |      |

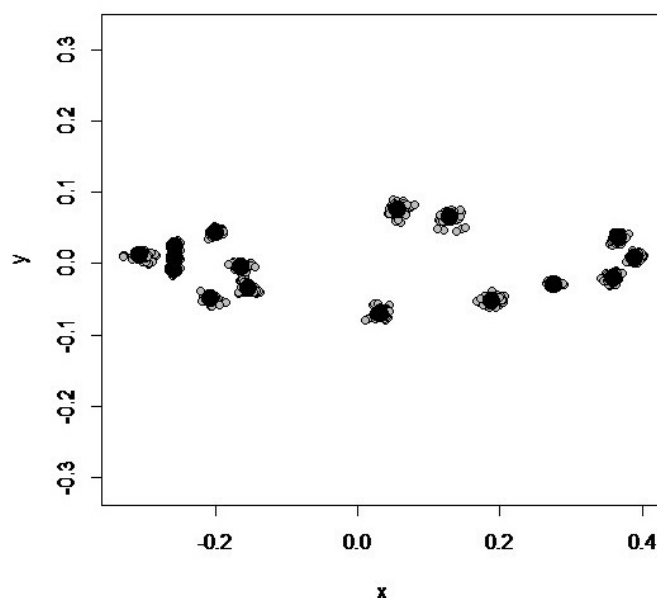
**Legend:** c – significant at 5% level ( $P < 0.05$ ); Note: NS – not significant ( $P > 0.05$ ).

adaptation to ecological conditions. These morphological changes include shallow-bodies, shallow caudal peduncles and elongated bodies.

In addition, the results of Procrustes regression between the environmental parameters and shape data of *H. leucisculus* in the Zarrineh River are shown in Table 2. These results showed that there is a significant relationship between temperature, altitude and velocity with shape data of fish ( $P < 0.05$ ). But other environmental factors (discharge and water depth) had no significant relationship with morphological features ( $P > 0.05$ ).

## Discussion

Generalized Procrustes analysis (GPA) is a common statistical tool that can be used to compare the body shape of fish. It is a multivariate technique widely used in shape analysis to find the optimal superimposition of two or multiple configurations (Gower, 1975; Goodall, 1991). In the present study, GPA showed factors influencing on the body shape of Saw-belly (*H. leucisculus*) in the Zarrineh River. The results of the geometric morphometric analysis revealed that body shape significantly is affected by environmental conditions. Findings of the present study showed that among the recorded factors, temperature, altitude and velocity effects the body shape of Saw-belly. Temperature as a major abiotic factor had a considerable effect on survival, growth and

**Figure 3.** Scatter plot of Generalized Procrustes Analysis (GPA) for shape data of *H. leucisculus*.

distribution of fish (Jobling, 1997; Rieman et al., 1997; Wilkes et al., 2001). Martinez-Palacios et al. (2002) and Ridha et al. (1998) stated that water temperature is one of the most important environmental parameters in the life cycle of fishes because of its direct effects on food intake, food

efficiency, oxygen consumption and reproduction. Water temperature is a driving force in the fish life because its effects are more than any other single factor (Pandit & Nakamura, 2010). The metabolic processes of Saw-belly are sensitive to changes in water temperature and a decrease in water temperatures to below the optimum results in reduced feed intake and growth. In addition, temperature also is known to be one of the important environmental parameters that strongly affect the body shape differentiation (Wimberger, 2008; Sfakianakis *et al.*, 2011). Previous studies have reported that temperature had a significant impact on the body shape of fish. For example, Sfakianakis *et al.* (2011) studied the impact of rearing temperature on body shape in Zebrafish (*Danio rerio*) juveniles. In this study, Zebrafish juveniles exhibit highly phenotypic plasticity induced by diverse thermal conditions during their early ontogenetic stages. The results suggest that water temperature effects on body shape of Zebrafish juveniles. In another study, Nasrolah-Pourmoghdam & Eagderi (2013) investigated the effect of temperature on the body shape of angelfish (*Pterophyllum scalare*) during early development using geometric morphometric analysis. The results showed that angelfish have lower caudal peduncle, higher body depth and fusiform body shape in higher temperature. This study indicated that temperature regulates the body shape of angelfish during early development. The results of these studies were similar to our research finding.

According to the results of this study, water flow is another very important environmental factor that can influence the body shape of Saw-Belly. A number of studies have shown that water velocity can be effective on morphological characteristics of fish species. Reyes (2015), who studied the effects of water flow on the morphology of *Astyanax mexicanus*, stated that body shape variation of this species was significantly associated with body size.

The causes of body changes among fish populations are usefully difficult to explain (Poulet *et al.*, 2004). Differences between the body shapes of fish populations can result from differences in habitat characteristics, genotypes or both of these (Solomon *et al.*, 2015). Body shape can show a high degree of plasticity in response to environmental variables such as temperature, turbidity, food availability, water depth and flow (Thompson, 1991; Wimberger, 2008; Mohaddasi *et al.*, 2013). Robinson & Parsons (2002) reported that phenotypic plasticity in fish can indicate the adaptation to environmental changes. Adaptation is physical and behavioral features that allow a living organism to better survive in its habitat. Therefore, body shape differences in fishes are often reflective of adaptation to ecological conditions and can be used as a diagnostic of a species (Park *et al.*, 2013; Kennedy, 2017).

The obtained results of the study showed the utility of geometric morphometric analysis in describing shape differences. In the present study, this method was very useful in describing body shape of *H. leucisculus* in the Zarinneh River. Also, the results indicated that Saw-belly (*H. leucisculus*) has made many adaptations in order to survive better in its habitat. This species has shallow caudal peduncle and elongated body in order to survive better in the Zarrineh River. Thus, this body shape of Saw-belly can help it adapt to its' environment but shouldn't forget that environmental parameters can also change the morphological features.

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