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RESEARCH PAPER

# Otolith Shape Analysis of Five Sillaginidae Species from the Persian Gulf and the Oman Sea

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## Abstract

The discrimination power of sagittal otolith shape analysis is evaluated for five closely related sillaginid species from the Persian Gulf and the Oman Sea; (*Sillago arabica, Sillago attenuata, Sillago indica, Sillago sihama* and *Sillaginopodys chondropus*) besides the description of morphological characters using scanning electron microscopy (SEM). The identification of sillaginids is difficult due to the limited reliable diagnostic morphological characters. The otolith shape was described by six shape indices (Roundness, Rectangularity, Circularity, Ellipticity, Form factor and Aspect ratio) and Elliptic Fourier Descriptors (EFD). For all shape indices, there was at least an overlap between the two species and no one was significantly different among the five species. Based on EFD analysis all five species were discriminated (91.7%) evidencing the high taxonomic value of sagittal otolith for the studied taxa. SEM images revealed sulcus acusticus as homosulcoid with an inframedian position in all five species. *S. arabica* and *S. sihama* are characterized by having a pseudo-ostiocaudal, while *S. indica*, *S. attenuata* and *Sillaginopodys chondropus* have ostial mode of opening. The ostium was tubular and slightly curved in all studied species, which was more strongly curved in *S. arabica*. These findings suggest that the sagittal otolith shape is a reliable tool to recognize inter-specific diagnosis for sillginid species in the Persian Gulf and the Oman Sea.

## Introduction

The family Sillaginidae (Smelt-whitings) is a group of demersal marine fishes that inhabit inshore waters throughout the Indo-West Pacific regions (McKay, 1992; Kaga et al., 2010; Nelson et al., 2016). The family contains about 40 species (Cheng et al., 2021; Xiao et al., 2021; Saha et al., 2022; Yu et al., 2022), many of which are exploited in commercial and recreational fisheries (McKay, 1992). The most important character primarily used to identify Sillaginid species is the swim bladder. The swim bladders of some species (such as two congeneric species, *S. sihama* and *S. indica*) are very similar and make the identification of these species difficult. Generally, the identification of species in Sillaginids is difficult and confusing due to the external morphological similarities and overlapping meristic characters are common (McKay, 1992; Gao et al., 2011; Xiao et al., 2016; Khandan-Barani et al., 2023). Five species of this family including *S. arabica*, *S. attenuata*, *S. indica*, *S. sihama* and *Sillaginopodys chondropus* inhabit the southern marine waters of Iran in the Persian Gulf and the Oman Sea (Alavi-Yeganeh et al., 2016; Khandan Barani et al., 2023). Due to distribution overlap

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in fishery catch, there is a need to use an inexpensive, reliable and easy method to discriminate between stocks within a species and between species. The morphological differences between these five species of Sillagindes are confusing. For example, the commonly used feature of first dorsal spine number overlaps between species of Sillaginides with 12-13 spines in *S. arabica*, 12-13 in *S. attenuata*, 10-9 in *S. indica*, 11 in *S. sihama* and 11-12 in *Sillaginopodys chondropus*. Sillaginids species in this area are similar in external morphology and their meristic characters have overlapping distributions (McKay, 1992; Alavi-Yeganeh et al., 2016; Panhwar et al., 2018). This study is the first attempt to evaluate the discrimination potential of sagittal otolith shape by using shape indices and elliptical Fourier analysis in five sympatric Sillaginid species in the Persian Gulf and the Oman Sea.

Currently, fish species discrimination by using quantitative tools, such as shape indices and geometric morphometrics, is widely used by fish taxonomists to evaluate the level of intra-specific morphological plasticity or inter-specific phylogenetic proximity (Tuset et al., 2016; Granados-Amores et al., 2020; Mereles et al., 2021; Damadi et al., 2024). Otoliths are considered as efficient tool for identifying fish species because of the high degree of species-specific morphological characteristics (Torres et al., 2000; Lombarte and Cruz, 2007; Tuset et al., 2012). Three pairs of otoliths (sagittae, lapilli, and asteriscus) are calcareous structures found in the inner ear of teleosts, and are associated with hearing, the sense of balance, gravity and acceleration (Popper and Coombs, 1982; Campana, 1999). Genetic and environmental factors affect the otolith morphology (Tuset et al., 2003) and these hard structures (especially the sagittae) have been widely used in fishery research for species discrimination (Zischke et al., 2016; Cerda et al., 2021), fish population structure and stocks separation (Duncan et al., 2018; Moreira et al., 2019; Moura et al., 2020), sexual dimorphism (Maciel et al., 2019), migration patterns (Sousa et al., 2016), ecology and ontogenetic studies (Volpedo et al., 2008; Sadighzadeh et al., 2014; Biolé et al., 2019).

Traditional morphometry and geometric methods have been used widely to describe the shape of the otolith (Pavlov, 2016; Bose et al., 2020). Traditional morphometry method provides only indirect assessments of the differences in shapes and a geometric method has greater potential for capturing all the shape variations and small-scale individual differences in the otolith shape (Pavlinov and Mikeshina, 2002). The direction of geometric morphometrics based on the analyses of closed contours and, in particular, Elliptic Fourier Analysis (Kuhl and Giardina, 1982) is used more often in many studies (Tracey et al., 2006; Agüera and Brophy, 2011; Moreira et al., 2019). Elliptical Fourier Analysis is a rapid, objective, and semi-automated method to obtain accurate information to compare otolith shapes. Also, shape indices can be used to evaluate the shape of objects which are simpler to calculate comparing the elliptical Fourier descriptors (Smith, 1992; Tuset et al., 2003).

## Materials and Methods

### Fish Collection

A total of 170 Sillaginid specimens were collected from coastal waters of the Oman Sea (Chabahar Bay; 25°18′ 47′′ N 60°30′47′′ E) and the Persian Gulf (Hormuz Island; 27°10′ 56′′ N 56°19′09′′ E) in the south of Iran during occasional sampling from March to August 2021 (Figure 1) by beach seine. Taxonomical identification of specimens was done based on McKay (1985 and 1992). Total lengths (TL) were measured by a clipper to the nearest 0.1 mm accuracy. Specimens identified as *Sillago arabica* (TL=115–152 mm, n=30), *S. attenuata* (TL=125–165 mm, n=30), *S. indica* (TL=110–169 mm, n=45), *S. sihama* (TL=140–202 mm, n=34) and *Sillaginopodys chonropus* (TL=127–192 mm, n=24).

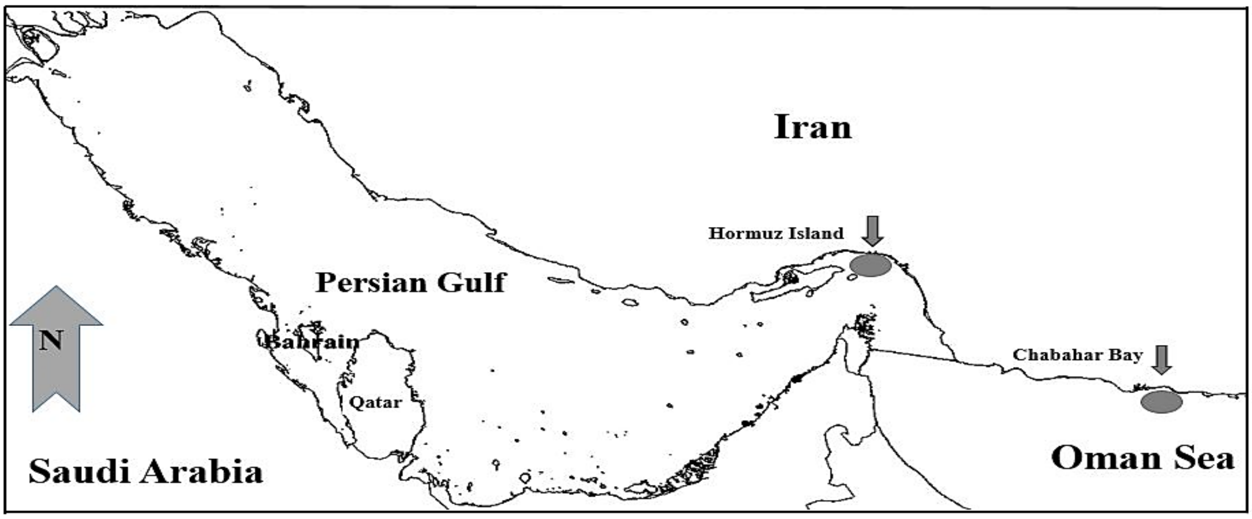


**Figure 1.** Sampling sites of Sillaginid specimens from the Persian Gulf and the Oman Sea.

**Scanning Electron Microscopy**

The cleaned otoliths were fixed on a specimen holder using sticker tape and coated with a 30-nm layer of gold and scanned with the TESCAN VEG A3 SEM. The morphological descriptions of the otoliths were based on the terminology proposed by Tuset et al (2008).

**Otolith Morphology and Shape Indices**

Fish otoliths were rinsed with 3% $H_2O_2$ (for 15 min) and distilled water after removing from each fish specimen, then dried and stored in polyethylene vials. Sagittal otolith from the left side was selected to be photographed under a stereomicroscope (Luxeo 6z, Labomed) with a digital camera (SP-320, Olympus). Four morphometric characters, otolith length (OL), otolith width (OW), perimeter (P) and otolith area (OA), were measured using photos and Digimizer (Ver. 5.6.0). Measurements were used to calculate six shape indices: Form Factor ($4\pi A/P^2$), Aspect Ratio (OL/OW), Roundness ($4A/\pi(OL)^2$), Circularity ($P^2/A$), Rectangularity ($A/(OL\times OW)$), and Ellipticity (OL– OW)/(OL+OW) (Tuset et al., 2003; Lord et al., 2012). As there was no significant difference observed between left and right otoliths (t-test, $p>0.05$), left otolith was selected for the analyses in this study.

**Otolith Outline Analysis**

Variation in otolith shape was assessed using elliptical Fourier analysis (Kuhl and Giardina, 1982; Ferson et al., 1985). The outline of the otolith was traced using the tps Util (1.38) (Rohlf, 2008), tpsDig (2.16) (Rohlf, 2006) and GMTP (Geometric Morphometrics Tools Package) software (Rohlf, 2006; Taravati and Darvish, 2010) The x and y coordinates of 180 landmarks were obtained from points equally spaced along the otolith's outline using tps. Dig (2.16) and the TPS file opened in the PAST statistical program (Hammer et al., 2001). The x- and y-coordinates of Fourier harmonics were made invariant to the otolith size, rotation and starting position of the tracing of the outline using the generalized Procrustes analysis of least squares superimposition method (Ferson et al., 1985; Rohlf and Slice, 1990). The Elliptic Fourier analysis method defines the outline using multiple components known as harmonics. Each harmonic is defined by four coefficients, derived from the projection of each outline point onto the x and y axes. The more harmonics there are, the more accurate the outline description becomes (Kuhl and Giardina 1982). For each numerical image, the software Past 4.16c calculated the Fourier coefficients (EFCc) to make them invariant to the otolith size and its orientation (and position) relative to the start of the outline, which is arbitrarily defined. Additionally, the Fourier power (FP) spectrum was computed to determine the optimal number of harmonics needed for the most accurate reconstruction of the otolith outline (Crampton 1995). The Fourier power (FP) spectrum is calculated using the formulas presented in Morat et al. (2012) and Çol and Yilmaz (2022). As 99.95% of the cumulative power was described by the first three harmonics, the otolith shape of five species was summarized by 120 Fourier coefficients. However, the coefficients derived from the first harmonic were not taken into account, because the outline reconstructed with these coefficients is a simple ellipse, resulting in maximum Fourier power and then masking the information derived from the other harmonics (Crampton 1995). Therefore, 29 harmonics and thus 116 Fourier coefficients were used for the data analysis. The Canonical variate analysis (CVA) was performed with the 116 Fourier coefficients.

**Statistical Analysis**

All the data were first examined for normality and homoscedasticity using Kolmogorov-Smirnov tests and presented as mean values ± standard deviation (±SD). The shape indices were corrected and standardized to eliminate possible body size and allometry effects (Lombarte and Lleonart, 1993). The shape indices were compared using one-way analysis of variance (ANOVA) followed by Duncan's post-hoc multiple comparisons to test the significant ($p<0.05$) differences among species.

The statistical differences among groups detected by CVA were tested with Wilks' lambda ($\lambda$). Mahalanobis distances (Bookstein 1991) were employed to assign each individual to the nearest group according to the group means. The classification success of the discriminant analysis was tested by jack-knife cross-validation. A dendrogram was constructed by hierarchical cluster analysis (UPGMA), based on the Euclidian distance values to assess the degree of similarity between Sillaginid species. All statistical analyses were performed using PAST (Ver. 4.16c) and SPSS. (Ver. 25.0).

## Results

**Scanning Electron Microscopy (SEM) Analysis**

The left sagittal otoliths (both sides of otolith) of five Sillaginid species (*S. arabica*, *S. attenuata*, *S. indica*, *S. sihama* and *Sillaginopodys chondropus*) were examined using a scanning electron microscope (Figure 2). The sagittal shape was oval in *S. attenuata* and *S. arabica* and near oval (or elliptical to rounded triangle) in other species. Most variations in the otolith shapes of these species were observed in the anterior region and ventral margins (Figure 2). The anterior region of sagittal otolith in *S. arabica* and *S. attenuata*, is broad and blunt to rounded, in *Sillaginopodys chondropus* it is slightly curved, in *S. indica* it is more stretched and slightly curved and in *S. sihama* it is elongated and thicker (Figure 2). The ventral margin in *S. arabica* is smooth but in *S. attenuata* it is slightly

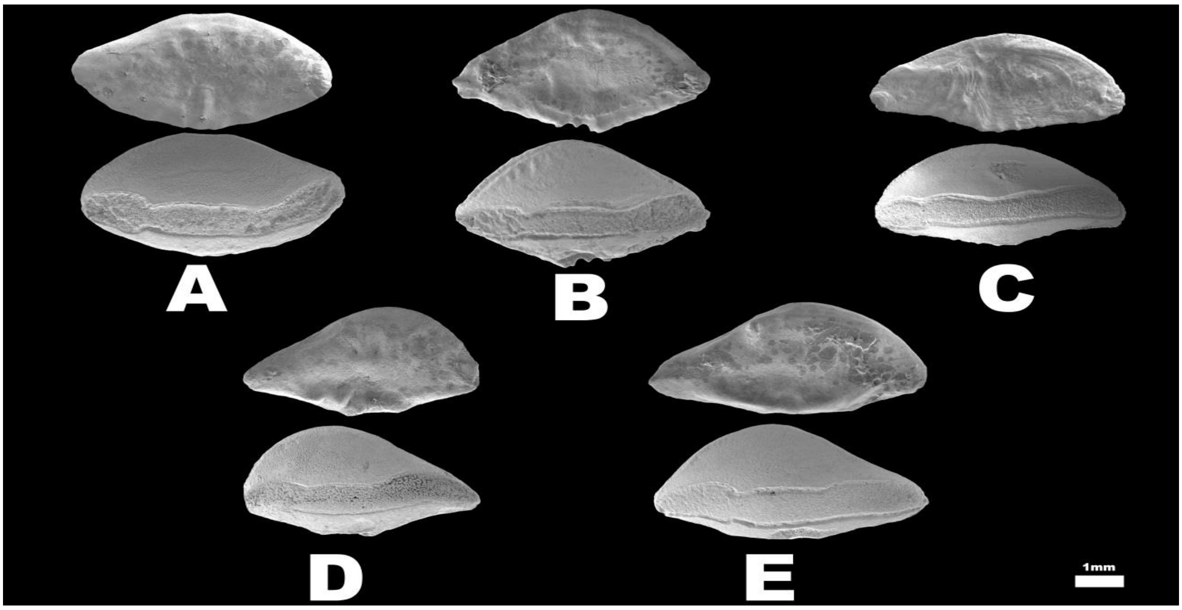


**Figure 2.** SEM imaging of left sagittae inner (top) and outer (down) surface; (A) *S. arabica,* (B) *S. attenuata*, (C) *Sillaginopodys chondropus*, (D) *S. indica* and (E) *S. sihama*

protruding. In *Sillaginopodys chondropus* the ventral margin is straight with an obvious protuberance. For *S. sihama* and *S. indica* dorsal margin of the sagittal otolith is smooth, and there are indistinct wavy protuberances on the ventral margin (Figure 2).

In all species, the sulcus acusticus was homosulcoid. In the *S. arabica* and *S. sihama*, sulcus opening was pseudo-ostiocaudal, while in the *S. indica*, *S. attenuata* and *Sillaginopodys chondropus,* it was ostial. The positions of the sulcus in these species were inframedian. The ostium was typically funnel-like and was shorter than the cauda in all studied species. The cauda was tubular, ending close to the posterior ventral margin. It was slightly curved in *S. attenuata*, *S. indica*, *S. sihama* and *Sillaginopodys chondropus* while in *S. arabica*, cauda was strongly curved (Figure 2 & Table 1).

**Otolith Shape Indices**

The results of the otolith shape indices are presented in Table 2. The roundness index in *S. sihama* and *Sillaginopodys chondropus* were significantly ($p<0.05$) lower than other Silaginid species indicating that the otolith is more elongated and less wide in these species and potentially could be used to distinguish them. The circularity index ranges from 14.53±0.41-16.94±0.73 and was significantly ($p<0.05$) higher in *Sillaginopodys chondropus* in comparison with other species except for *S. sihama*. The rectangularity index range was 0.69±0.25-0.73±0.04 and all the otoliths were non-square. The ellipticity index ranges appeared as 0.25±0.02-0.34±0.02 indicating otoliths are mostly oval or fusiform to spindle-like shape. The *S. attenuata* is potentially distinguishable from other studied species by significantly ($p<0.05$) lower ellipticity index value. Based on the values of the aspect ratio index, otoliths of *Sillaginopodys chondropus* (2.16±0.10) and *S. sihama* (2.04±0.20) were elongated. Significantly the lowest aspect ratio index ($p<0.05$) estimated for *S. attenuata* which could be useful to distinguish this species. *S. arabica* and *S. attenuata* have the highest value of form factor, indicating the flattest edges compared to other species. The results of a multivariate comparison of otolith shape indices with CVA are given in Figure 3. Five

**Table 1**. Terms used to define the ostial region of the sulcus acusticus (Tuset et al., 2008)

| | |
|---|---|
| Homosulcoid | Sulcus with ostium and cauda clearly differentiated, but similar in shape and almost symmetrical |
| Pseudo-ostiocaudal | Sulcus with both ends very close to the opposing margins of the otolith; the openings may be indistinct or reduced to very small, narrow channels. |
| Ostial | Sulcus with an ostium open widely in the anterior margin of the otolith, and with a cauda distinctly closed far away from the posterior margin |
| Inframedian | Sulcus generally positioned below the longitudinal midline of the otolith; the ventral area is noticeably smaller than the dorsal area. |
| Funnel-like | The walls of the ostium are straight or concave, but more or less symmetrical, and spread apart anterior wise from the region of confluence with the cauda. |
| Tubular | The ostium is rather long and its walls are straight or curved, but they follow a fairly parallel path from the region of confluence with the cauda to the anterior tip |

**Table 2**. Shape indices values for otolith of five Sillaginid species from the Persian Gulf and the Oman Sea (Mean ± sd)

| Shape index | Species | | | | |
|---|---|---|---|---|---|
| | *S. arabica* | *S. attenuata* | *S. indica* | *S. sihama* | *S. chondropus* |
| Roundness | $0.51±0.03^{b}$ | $0.55±0.02^{c}$ | $0.54±0.1^{bc}$ | $0.45±0.05^{a}$ | $0.43±0.02^{a}$ |
| Rectangularity | $0.73±0.04^{ab}$ | $0.72±0.02^{ab}$ | $0.75±0.10^{b}$ | $0.70±0.04^{a}$ | $0.69±0.25^{a}$ |
| Circularity | $15.22±0.51^{ab}$ | $14.53±0.41^{a}$ | $15.70±1.21^{bc}$ | $16.36±2.08^{cd}$ | $16.94±0.73^{d}$ |
| Ellipticity | $0.29±0.02^{b}$ | $0.25±0.02^{a}$ | $0.28±0.02^{b}$ | $0.32±0.02^{c}$ | $0.34±0.02^{c}$ |
| Form factor | $0.83±0.09^{bc}$ | $0.87±0.02^{c}$ | $0.79±0.06^{b}$ | $0.78±0.09^{ab}$ | $0.74±0.03^{a}$ |
| Aspect ratio | $1.90±0.29^{b}$ | $1.67±0.57^{a}$ | $1.78±0.10^{b}$ | $2.01±0.11^{c}$ | $2.16±0.10^{c}$ |

The different superscript letters in the same row indicate different subset identified by analysis of variance (ANOVA) followed by Duncan's post-hoc multiple comparisons significant difference among groups. The same superscripts indicate insignificant difference.

species are discriminated not distinctly, and the variation of the data is totally described by two variables (CV1=75.4% and CV2=24.6%). The otolith shape indices analysis was the weakest method for species discrimination, with a classification success of 41.5% of cases (Table 3). Only the otoliths of *Sillaginopodys chondropus* and *S. attenuata* were identified correctly above 75%, while *S. arabica*, *S. indica* and *S. sihama* were assigned below 39%.

**Fourier Analysis**

The results of the Fourier analysis of otolith shape are summarized in Figure 4. Regarding Canonical discriminant analysis, the five groups were identified clearly, and the first two variables explained 94% of the total variance among groups (CV1=65.7% and CV2=28.3%). Differences were statistically significant (Wilks' lambda=0.001, p<0.0001).

The clear distinction between species was validated using the matrix of cross-validated values, achieving an overall correct classification rate of 91.7%. Specifically, a 100% correct classification rate was achieved for *S. sihama*, *S. indica*, and *Sillaginopody chondropus* (Table 4).

UPGMA cluster analysis, using Euclidean distance, for otolith shape clearly distinguished the Sillaginid species. The dendrogram is constructed based on Euclidian distance (Figure 5). Otoliths of five examined Sillaginid species were placed into two major groups in cluster analysis, the first group contained the *Sillaginopody chondropus* which was separated from the *S. arabica* and the *S. attenuata* with an oval shape and the second group included *S. indica* and *S. sihama* with an elliptical to rounded triangular otolith shape (Figure 5).

## Discussion

Accurate species identification of commercially landed fish in through fishery monitoring programs by fisheries observers or fishing crew plays an important role in the validation of reported catch by vessels, the collection of useful biological data and define of fishery management strategies for harvested species (Garcia-Vazquez et al., 2012; Williams et al., 2018). Misidentification of Sillaginid fishes by fisheries observers due to similar external morphometric and meristic characteristics of many them has been confirmed through use of genetic markers (Cheng et al., 2021; Saha et al., 2022). Such misidentification is a concern for stock assessments and conservation status of these species (Yang et al., 2020; Cheng et al., 2021). For these five Sillaginides, species identification (especially *S. sihama* and *S. indica*) at sea is particularly confusing and challenging, due in part to subtleties in body morphological variation among the species. Our results demonstrate that otolith shape can provide a fast, cost-effective, and accurate method for identification and discrimination of Sillaginid species and verifying at-sea identifications.

Many studies have shown differences in otolith shape between morphologically similar species (Zhuang et al., 2015; Pavlov, 2016; Cerda et al., 2021; Mereles et al., 2021; Moore et al., 2022). To the best of our knowledge, this study is the first study to use otolith shape to validate Sillaginid species identifications. While shape indices were only partially useful for distinguishing *S. attenuata* from other species, Elliptical Fourier analysis is considered to be powerful and has great potential for capturing all the shape variations and provides an efficient method for describing otolith shapes (Cerda et al., 2021; Ghanbarifardi and Zarei, 2021). The accuracy of classification in this study was 94%. The high accuracy of classification in our research-collected otoliths indicate that the approach can be applied to verify Sillaginid species identifications in fishery-sourced samples to species level with a high degree of confidence. Similarly, high accuracy was documented in many studies with different fish species such as Macrouridae (Moore et al., 2022), Scorpaena (Yedier and Bostanci, 2021), Gobiidae (Bani et al., 2013) and Serranidae (Stransky and MacLellan, 2005). According to this, otolith shape is introduced as a new tool for taxonomic identification of Sillaginides from the southern water of Iran.

Differences in otolith shape and morphology among Sillaginid species may result from genetics and environmental factors. Genetic differences may resolve as different phenotypic traits (such as morphometric variation), which are likely to be more pronounced among species than populations of the same species

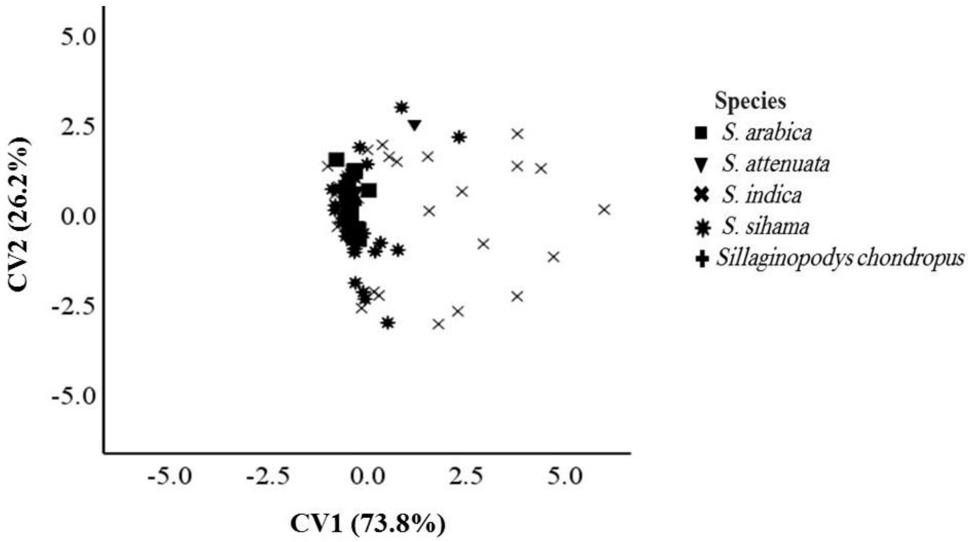


**Figure 3.** Distribution of partial warp scores obtained from shape indices CVA for *S.arabica, S. attenuata, S. indica, S. sihama* and *Sillaginopodys chondropus.*

**Table 3**. Cross-validated classification matrix of the CVA among Sillaginid species base on the shape indices

| Species | *S. arabica* | *S. attenuata* | *S. indica* | *S. sihama* | *Sillaginopodys chondropus* |
|---|---|---|---|---|---|
| *S. arabica* | 32.1 | 27.6 | 0 | 30 | 10.3 |
| *S. attenuata* | 20 | 75 | 5 | 0 | 0 |
| *S. indica* | 12.9 | 29 | 38.70 | 6.8 | 12.6 |
| *S. sihama* | 14.3 | 17.1 | 2.9 | 11.4 | 54.3 |
| *Sillaginopodys chondropus* | 5 | 0 | 0 | 15 | 80 |

41.5% of cross-validated grouped cases correctly classified

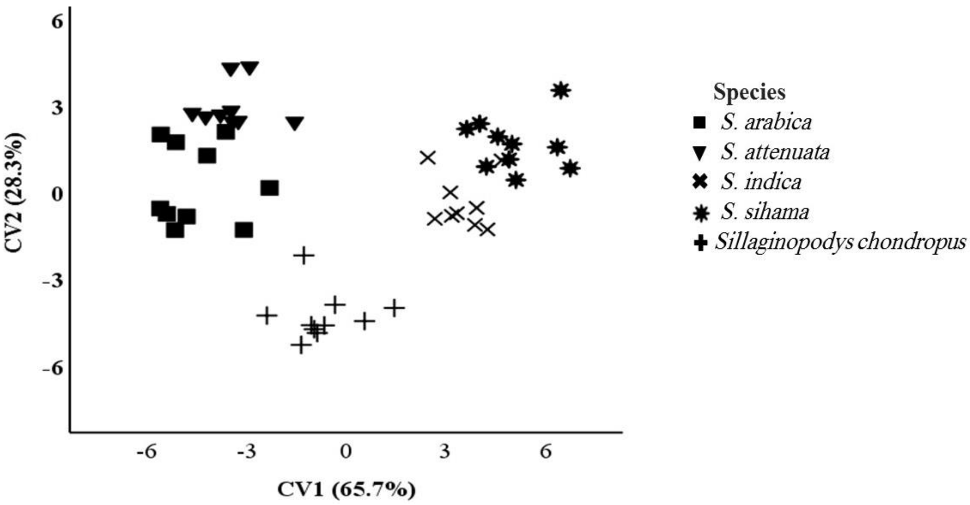


**Figure 4.** CVA of 116 Fourier coefficients for *S.arabica, S. attenuata, S. indica, S. sihama* and *Sillaginopodys chondropus.*

**Table 4**. Cross-validated classification matrix of the CVA among Sillaginid species base on the fourier analysis.

| Species | *S. arabica* | *S. attenuata* | *S. indica* | *S. sihama* | *Sillaginopodys chondropus* |
|---|---|---|---|---|---|
| *S. arabica* | 100 | 0 | 0 | 0 | 0 |
| *S. attenuata* | 11.1 | 88.9 | 0 | 0 | 0 |
| *S. indica* | 0 | 0 | 88.89 | 11.1 | 0 |
| *S. sihama* | 0 | 0 | 20 | 80 | 0 |
| *Sillaginopodys chondropus* | 0 | 0 | 0 | 0 | 100 |

91.7% of cross-validated grouped cases correctly classified

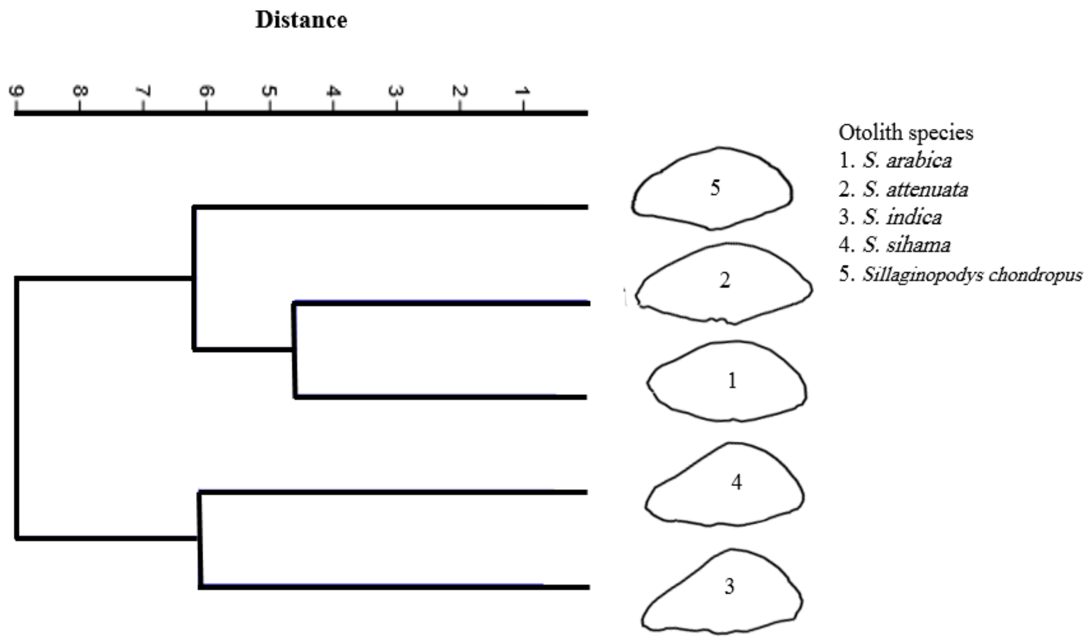


**Figure 5.** The dissimilarity of Sillaginidae species based on the Euclidian distance, grouping by hierarchical cluster analysis (UPGMA). 1. *S. arabica*, 2. *S. attenuata*, 3. *S. indica*, 4. *S. sihama* and 5. *Sillaginopodys chondropus.*

(Cardinale et al., 2004). Consistent with patterns in otolith general otolith shape (especially ventral and anterior part) which is specific for each species. Indeed, several studies have shown that environmental factors could influence the differences in otolith shape between species (Gagliano and McCormick, 2004; Hüssy, 2008; Vignon and Morat, 2010; Ghanbarifardi et al., 2020). Also, age of species could affect the otolith shape which is particularly noticeable between age-1 and older fish, likely due to rapid growth during the first year of life (Neves et al., 2024). To avoid age bias, smaller size fish specimens were omitted from analysis.

According to Zorica et al. (2010) and Bose et al. (2020), the otolith shape might be similar in species inhabiting a similar environment. The ecology of the Sillaginid species studied here is poorly known. All studied species inhabit coastal waters with similar environmental conditions in their area of distribution. However, additional studies are required to evaluate the influence of genetics and their interactions with environmental factors to affect the otolith shape among the studied species.

This study also described the ultrastructure of sagittae in the five Sillaginid species using SEM, for the first time in the Persian Gulf and the Oman Sea. Scanning electron microscopy revealed that in addition to the main variability in the sagittal area (ventral and anterior part), in terms of some characteristics of the sulcus acusticus (the sulcus opening and the magnitude of the posterior curvature), there are also differences among Sillaginid species. The pseudo-ostiocaudal mode opening was observed clearly in *S. arabica* and *S. sihama* and strongly curved cauda were observed only in *S. arabica*. The sulcus acusticus was shown to have a species-specific character and can be useful for differentiating fish species (Gaemers, 1984; Nolf, 1985; Torres et al., 2000). Previous studies were able to successfully use sulcus morphology to discriminate fish species (Granados-Amores et al., 2020; D'Iglio et al., 2021). Since our study discovered differences in the sulcus acusticus shape of otoliths in the Sillaginid species, it seems possible to establish species discriminate fish species within this family as well using otoliths.

## Conclusion

Sagittal otolith shape descriptors and some characteristics of sulcus acusticus are effective tools for the delimitation of Sillaginid species and it is presented as a new tool for the taxonomic identification of this family in the Persian Gulf and the Oman Sea.

## Ethical Statement

All applicable international, national, and/or institutional guidelines for the care and use of animals were followed.

## Conflict of Interest

The authors declare that they have no known competing financial interests or personal relationships that could have appeared to influence the work reported in this paper.

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## Author Contribution

Hashem Khandan Barani: Formal analysis; methodology; investigation; writing-original draft; visualization. Mohammad Sadegh Alavi-Yeganeh: Data curation; supervision; project administration; resources; writing–review and editing. Alireza Riyahi Bakhtiari: validation; writing–review and editing. Mehdi Ghanbarifardi: Investigation; methodology; validation; visualization; writing – review and editing.

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