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Relationship between fish length and otolith morphological characteristics of *Sargocentron spiniferum* (Forsskål, 1775) from the southern Red Sea

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Abstract: Otolith morphology analysis is one of the main tools used for fish or fish stock identification. Moreover, otolith shape can also be used in fish dietary studies (stomach content) for the identification of prey fishes and their size according to the relationship between fish and otolith sizes. In the present study, the relationship between fish length and otolith morphological dimensions was investigated for the sabre squirrelfish, *Sargocentron spiniferum* (Forsskål, 1775) (family: Holocentridae). Samples of 185 fish were collected from the coast of the Red Sea, Egypt. To analyze the relationship between fish and otolith, otolith morphometric measurements (length, width, area, perimeter, weight, sulcus, and ostium) and shape factors (aspect ratio, compactness, form factor, rectangularity, roundness, ellipticity, squareness) describing outline shape were extracted using image analysis. Generalized linear models were applied for the relationship between body length and each otolith morphology feature. From the relationships between the total length of fish and fourteen morphology features, only otolith length, caudal length, and squareness were significantly correlated with fish size. Our results provide more information for the relationship between fish length and otolith morphometric features.

Keywords: Fish size; otolith shape; head side; *Sargocentron spiniferum*; Red Sea; Egypt.

1. Introduction

The sabre squirrelfish *Sargocentron spiniferum* (Forsskål, 1775) is a member of the family Holocentridae, which is mainly distributed in the Indo-Pacific from the Red Sea and East Africa to the Hawaiian Islands and Ducie Islands extending south to Australia. It is also distributed in New Caledonia, north to southern Japan and the Ogasawara Islands [1, 2]. This species inhabits different reef areas between reef flats in lagoons and seaward reefs at a depth of 122 m. This fish occurs under ledges during the day [3], and when smaller in size, this fish inhabits shallow, protected areas. It is a nocturnal fish that feeds on crabs, shrimp, and small fishes [4].

Many researchers have used the clear and distinct growth rings of sagittal otoliths to study the age and growth of fishes. Otoliths are calcified structures located at the right and left inner ears of fishes, which are useful in the study of fish biology, ecology, and fisheries science [5-7]. Otoliths are also used to estimate movement, varied habitat, population dynamics, and trophic level for fish species [7-15].

Otolith morphology is used to recognise fish species in taxonomic, phylogenetic, paleoichthyological and dietary studies [16-19]. Within species, otolith shape is also used to identify stocks. A recent study showed, however, that directional asymmetry between right and left otoliths within individuals could affect the results from the otolith shape analysis as tool to identify the stocks [20]. Consequently, the aim of the present work was to estimate the relationship between fish size and otolith outline features according to the...
location of the otolith, i.e., left versus right inner ear for *S. spiniferum* from the Egyptian waters of the Red Sea.

2. Materials and Methods

Fish species were randomly collected from the southern Red Sea at the Shalateen fishing port, which is located 520 km south of Hurghada (Figure 1), Egypt, between March 2018 and February 2019.

The fish were obtained from the commercial catch of the hook and line fishery at Shalateen fishing ground. In the laboratory, total fish length (TL) was measured to the nearest 0.1 mm, then the sex was determined from macroscopic observation. The total length of the species ranged between 17.7 and 45.8 cm. Sagittal otoliths (370 paired left and right otoliths) were extracted from the inner ear of 185 *S. spiniferum*, cleaned and dried. Otolith weight (OW) for each head side was measured using a digital balance AS220 k/1 to the nearest 0.0001 g. Otolith images were captured using a Euromex CMEX-10 PRO camera with a stereomicroscope. The following otolith measurements: length (OL, mm), width (OH, mm), area (OA, mm²), perimeter (OP, mm), sulcus length (SU), ostium length (OS) were taken using image processing systems (Image J analysis software, [21]; detailed descriptions is in Figure 2).
Figure 2. Diagram and scheme of the otolith of *Sargocentron spiniferum* illustrating various features of the otolith measurements.

From these size parameters, the following shape indices were calculated: form factor (FF), aspect ratio (AR), circularity (CI) rectangularity (RE), roundness (RO), ellipticity (EL), compactness (C), and squareness (SQ) [21-28]. The formulae of these shape index factors (FF, C, AR, CI, RO, RE, EL, and SQ) are presented in Table 1.
Table 1. Size dimension parameters and shape indices of otolith used to describe *S. spiniferum* otolith characteristics.

| Size parameters     | Shape indices          |
|---------------------|------------------------|
| Length: OL          | Circularity: CI = \( \frac{OP}{OA^2} \) |
| Width: OH           | Ellipticity: EL = \( \frac{OL - OH}{OL + OH} \) |
| Perimeter: OP       | Roundness: RO = \( \frac{4 OA}{\pi OL^2} \) |
| Area: OA            | Aspect ratio: AR = \( \frac{OL}{OH} \) |
| Weight: OW          | Rectangularity: RE = \( \frac{OA}{OL \cdot OH} \) |
| Sulcus: OSL         | Form-factor: FF = \( \frac{4 \pi OA}{OP^2} \) |
| Ostium: OOs         | Compactness: CO = \( \frac{OP^2}{OA} \) |
| Cauda: OCu          | Squareness: SQ = \( \frac{OA}{OL \cdot OW} \) |

Subsequently, the relationship of fish length with each otolith feature (size parameters or shape indices) was modelled according to the side (S):

\[
\text{Log}(TL) = \text{Otolith feature} + \text{Side} + \text{Otolith feature:Side}
\]  

(1)

Statistical analyses were performed in the R statistical environment [29] stats package [30].

3. Results

A general pattern of *S. spiniferum* sagittae can be recognized in adult individuals: the otolith shape of *S. spiniferum* is ovate with sinuate margins, and is elongated, reflecting slower growth of the dorso-ventral axis compared to the antero-posterior axis. The dorso-ventral axis has the highest growth, with with a much higher distance than that observed on antero-posterior axis with the rounded posterior area and the lobed anterior area. The *sulcus acousticus* is ostial with the heterosulcus and ostium formed by a short, funnel-like ostium that opens to the anterior margin, and closed, tubular cauda at least two times larger than the ostium (Figure 2). Descriptive statistics and paired t-test results for left and right otoliths of *S. spiniferum* are given in Table 2.
Table 2. Summary of descriptive statistics and paired t-test results for left and right oto-
 liths of S. spiniferum (n=185).

| Parameters (side) | Minimum  | Maximum  | Mean    | Std. Deviation | paired t test |
|-------------------|----------|----------|---------|----------------|---------------|
|                   | Left     | Right    |         |                |               |
| OL                | 9.54     | 14.11    | 11.91   | 0.91           | 0.0880        |
| Right             | 9.56     | 14.11    | 11.88   | 0.93           |               |
| OH                | 5.75     | 9.97     | 7.55    | 0.86           | 0.3790        |
| Right             | 6.04     | 9.97     | 7.57    | 0.83           |               |
| OW                | 0.0365   | 0.1574   | 0.0648  | 0.0017         | 0.1931        |
| Right             | 0.0255   | 0.1572   | 0.0643  | 0.0017         |               |
| OSL               | 8.18     | 13.75    | 10.47   | 1.11           | 0.0000        |
| Right             | 8.15     | 13.73    | 10.31   | 1.13           |               |
| OOs               | 2.56     | 5.86     | 4.21    | 0.60           | 0.1120        |
| Right             | 2.58     | 5.98     | 4.26    | 0.64           |               |
| OCu               | 4.84     | 8.04     | 6.27    | 0.72           | 0.0000        |
| Right             | 4.87     | 8.03     | 6.05    | 0.72           |               |
| OA                | 43.44    | 91.29    | 66.89   | 0.729          | 0.18          |
| Right             | 44.44    | 91.33    | 66.90   | 0.727          |               |
| OP                | 25.01    | 40.01    | 32.278  | 0.194          | 0.17          |
| Right             | 25.01    | 40.00    | 32.276  | 0.194          |               |
| AS                | 0.500    | 0.730    | 0.633   | 0.040          | 0.135         |
| Right             | 12.680   | 20.680   | 15.697  | 0.038          |               |
| CO                | 0.150    | 0.250    | 0.201   | 1.389          | 0.073         |
| Right             | 0.580    | 1.010    | 0.746   | 1.389          |               |
| FF                | 0.490    | 0.740    | 0.601   | 0.019          | 0.319         |
| Right             | 0.160    | 0.330    | 0.226   | 0.018          |               |
| RE                | 12.680   | 20.680   | 15.697  | 0.072          | 0.791         |
| Right             | 0.580    | 1.010    | 0.746   | 0.069          |               |
| RO                | 0.530    | 0.740    | 0.637   | 0.058          | 0.072         |
| Right             | 12.680   | 20.680   | 15.693  | 0.056          |               |
| EL                | 0.150    | 0.250    | 0.201   | 0.031          | 0.113         |
| Right             | 0.590    | 1.000    | 0.746   | 0.029          |               |
| CI                | 0.490    | 0.780    | 0.605   | 1.389          | 0.073         |
| Right             | 0.150    | 0.310    | 0.223   | 1.389          |               |
| SQ                | 12.680   | 20.680   | 15.693  | 0.072          | 0.791         |
| Right             | 0.590    | 1.000    | 0.746   | 0.069          |               |

Otolith parameters: OL, length, OH- width, OW- weight, OSL- sulcus length, OOs- ostium length, OCu- caudal length, OA- area, OP- perimeter; lengths in mm, weight in g, area in mm².

Analysis of the relationships between fish length and fourteen otolith shape descriptors using a generalized linear model showed that there is a significant relationship between eight otolith parameters (ostium length, caudal length, otolith area, otolith perimeter, compactness, form factor, circularity, and squareness) with the total length of fish.
Only the relationship of body length with otolith length, caudal length and squareness (P<0.05) was significant for right and left otolith (Table 3).

**Table 3.** Generalized linear models for the relationship between fish length and each otolith variable for *S. spiniferum* from the Red Sea with the significance (TL) and Side effect (observed from the interaction between Otolith feature and side in the GLM). The relationship between TL and each otolith descriptor was showed according to the significance of side effect, OL, otolith length; OH, otolith width; SU, sulcus; OS, ostium; OA, otolith area; OP, otolith perimeter, FF, form factor; AR, aspect ratio; C, compactness; RE, rectangularity; RO, roundness; EL, ellipticity; CI, circularity; SQ, squareness.

| Otolith descriptor | TL | Side effect | Relationship between TL and otolith descriptor | R²  |
|--------------------|----|-------------|-----------------------------------------------|-----|
| OL                 | 0.482 | 0.010 | Left side TL = 0.719+ 0.065OL  
Right side TL = 0.636+ 0.072OL | R² = 0.665 |
| OH                 | 0.274 | 0.148 | TL = 0.896+ 0.078OH  
R² = 0.736 |
| Sulcus length      | 0.078 | 0.336 | TL = 0.904+ 0.057SU  
R² = 0.777 |
| Ostium length      | 0.008 | 0.621 | TL = 1.116+ 0.087OS  
R² = 0.586 |
| Caudal length      | < 0.001 | < 0.001 | Left side TL = 1.117+ 0.069CA  
Right side TL = 1.047+ 0.007CA  
R² = 0.384  
R² = 0.806 |
| OA                 | < 0.001 | 0.446 | TL = 1.095+ 0.006OA  
R² = 0.851 |
| OP                 | 0.007 | 0.986 | TL = 0.689+ 0.025OP  
R² = 0.813 |
| AR                 | 0.973 | 0.853 | TL = 1.063+ 0.675AR  
R² = 0.138 |
| C                  | 0.023 | 0.703 | TL = 1.331+ 0.010C  
R² = 0.037 |
| FF                 | 0.001 | 0.388 | TL = 1.656- 0.833FF  
R² = 0.043 |
| RE                 | 0.301 | 0.642 | TL = 1.807- 0.427RE  
R² = 0.166 |
| RO                 | 0.523 | 0.263 | TL = 1.734- 0.327RO  
R² = 0.111 |
| EL                 | 0.574 | 0.091 | TL = 1.686- 0.872EL  
R² = 0.132 |
| CI                 | < 0.001 | 0.802 | TL = 1.331+ 0.010CI  
R² = 0.037 |
| SQ                 | < 0.001 | < 0.001 | Left side TL = 1.734- 0.328SQ  
Right side TL = 1.785-0.396SQ  
R² = 0.111  
R² = 0.138 |

The correlation between fish total length and otolith morphology showed that with an increase in the total length, the following factors of otolith morphology also increase: length, width, sulcus length, ostium length, caudal length, area, and perimeter (Figure 3). These relationships between body length and otolith measurements (left and
right) were best fitted as linear regression. The relationship between body length and the aspect ratio was, however, close to 1, confirming that the otolith of *S. spiniferum* was oval. There is a positive relationship between total length and the aspect ratio, and the compactness and circularity values. The form factor, rectangularity, roundness, ellipticity, and squareness values decreased as the TL decreased.

![Graphs showing relationships between body length and otolith morphological structures](image)

**Figure 3.** Relationships of body length with the otolith morphological structures (red points = left otolith, and green points = right otolith) according to the side of *S. spiniferum* captured from Shalateen, Red Sea, Egypt.

### 4. Discussion

The sabre squirrelfish *Sargocentron spiniferum* (Forsskål, 1775) contributes to important fisheries, especially the small artisanal fisheries at Shalateen fishing ground, Red Sea, Egypt. Basic data on the biology and dynamics of the species are essential for successful stock assessment and consequently in fisheries management. The observed fish length and shape of the otolith in this study should encourage more research to verify the essential role of otolith morphometric measurements in fish stock identification. The strong correlation between the somatic length and otolith size suggests that somatic growth has a significant influence on the otolith growth [9,31]. The results of this study were not similar to those previously obtained for the redcoat *S. rubrum* [26], where the present study shows greater otolith length, weight and height than *S. rubrum*. Previous studies have focused mainly on the relationship between otolith measurements and fish length [6, 32-36]. In our study, the results of generalized linear models showed that the relationship among these parameters is variable as the otolith length can be affected by the choice of the otolith (significant asymmetry between right and left otoliths). For this species, if the
relationship between fish size and otolith size is used, it is necessary to identify the used side to avoid introducing a bias in the results. Moreover, otolith asymmetry could be considered as a sensitive indicator of fish health that directly affects the fish performance because otoliths are essential to balance and hearing [37-38]. Based on the present data, the relationships between TL and AR, CO, and CI were determined as linear, while the relationships among TL and FF, RE, RO, EL, and SQ were determined as nonlinear. The shape of otolith from different geographical areas is, however, influenced by both environmental parameters (e.g salinity, temperature) and biotic parameters, for example prey availability, and depends on individual genotype [9,39-41]. Consequently, an interaction of environmental and genetic fluctuation generates the morphological variance in otolith shape that may allow the differentiation of stock units. However, the factors that affects the shapes are not fully understood and have not yet been investigated deeply [42]. A recent and on-going work showed that the ontogenetic trajectory of otolith shape could be impacted by the environmental disturbance during the early life stage [43]. The relationships between fish size and otolith shape indices demonstrate the high variability in fish length and morphometric parameters, indicating that the otolith of *S. spiniferum* is rectangular to oval. The results of this study on the fish size and otolith morphometric parameters are useful for further research on verifying the role of otoliths in identification, discrimination and taxonomic classification of fish. The results concur with previous studies showing that otoliths can be widely used for the discrimination and variation of fish species because of their form, diet, weight, and growth [44,45]. Finally, the estimation of the generalized linear models in the present work may be good tool to study the relationship between fish and otolith morphometric features, which are used for fish population dynamics, stomach contents analyses of piscivorous predators, paleontological composition, and yield estimates.

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