BRILL

Animal Biology 73 (2023) 195–211

Animal Biology
brill.com/ab

Ontogenetic changes in sagitta otoliths of whitemouth croaker *Micropogonias furnieri* (Acanthuriformes: Sciaenidae) and its implication in acoustic communication

José E. Verocai[1,*], **Antoni Lombarte**[2,*] **and Walter Norbis**[3,4]

[1] Instituto de Ecología y Ciencias Ambientales, Facultad de Ciencias, Montevideo 11400, Montevideo, Uruguay

[2] Institut de Ciències del Mar – Consejo Superior de Investigaciones Científicas (CSIC) – Barcelona, Passeig Marítim de la Barceloneta, 37, 08003 Barcelona, Catalonia, Spain

[3] Instituto de Biología, Facultad de Ciencias, Montevideo 11400, Montevideo, Uruguay

[4] Departamento de Biología, Direccion Nacional de Recursos Acuáticos, MGAP, 11200, Montevideo, Uruguay

*Corresponding author; e-mail: otolito21@gmail.com; toni@icm.csic.es

ORCID iDs: Verocai: 0000-0001-6240-4797; Lombarte: 0000-0001-5215-4587; Norbis: 0000-0002-7819-3450

Received 22 September 2022; final revision received 28 January 2023; accepted 16 February 2023; published online 29 March 2023; published in print 19 June 2023

Abstract

The otoliths of teleosts are part of the inner ear. They are related to the balance system and are also mechanical components of sound transmission, responsible for hearing in fish. Their morphology is determined by the function they perform and is also influenced by environmental factors. In this context, our goal was to: analyse the possible morphological changes of sagitta otoliths of *Micropogonias furnieri* (Desmarest, 1823) during its ontogeny, based on six biometric indexes. We analysed 162 sagittae otoliths of *Micropogonias furnieri* collected in the Rio de la Plata estuary. The variation of the indexes was associated with the different size groups. Rectangularity and aspect ratio [otolith length/total fish length (OL/TL) and sulcus area/otolith area (SA/OA)] were the indexes that contributed the most to the discriminant function. Biometric index differences were related to diverse juvenile environments. The observed shift in the SA/OA relationship could be attributable to a need to respond to a change in the perception of sound (pulse and frequency) experienced by individuals larger than 20 cm that begin to frequent the same areas as spawning adults. In this sense, the ontogenetic morphological change of sagitta otoliths plays an important role in perception and acoustic communication for this species.

Published with license by Koninklijke Brill NV | DOI: 10.1163/15707563-bja10105
 | ISSN: 1570-7555 (print) 1570-7563 (online)

Keywords
Biometric indexes; *Micropogonias furnieri*; ontogenetic; otolith; Rio de la Plata estuary; whitemouth croaker

Introduction

The sagitta otoliths of teleost fishes are structures composed of calcium carbonate (aragonite, vaterite), organic matter and trace elements (Simkiss, 1974; Morales-Nin, 1987; Gauldie & Nelson, 1990). They are located inside the skull, in the otic capsule, being part of the inner ear, are mechanical components of sound transmission, and also function as an organ of equilibrium (Fay & Coombs, 1983; Campana & Casselman, 1993; Popper & Platt, 1993). In addition to being genetically established, its morphology is also determined by its function (Gauldie, 1988; Lombarte et al., 2006; Tuset et al., 2008). It is also influenced by intrinsic and extrinsic factors, such as reproduction, feeding, depth, salinity and water temperature (Casselman, 1990; Lombarte & Lleonart, 1993; Lombarte et al., 2003). Owing to its relevance as a sensory organ, the morphology of sagitta otoliths in sciaenids has been described in numerous works. The main reason is that most members of this family are characterized by being specialized in acoustic communication, finding a relationship between the structure of the otolith, the swim bladder and the auditory function (Ramcharitar et al., 2001, 2004, 2006; Cruz & Lombarte, 2004; Ramcharitar & Popper, 2004).

In that context, the study of otolith biometric indexes (shape) variation during fish growth has implications for the function they fulfill throughout development, particularly in long-lived species such as *Micropogonias furnieri* (Desmarest, 1823) (Norbis & Verocai, 2005) (42 years) which presents the largest otoliths among the Rio the Plata estuary sciaenids (Chao, 1978; Baldás et al., 1997).

Despite the existence of other very suitable methods to analyze otolith shape for species classification (Tuset et al., 2021), we investigate the morphological variation of the sagitta otoliths based on the relationships that exist among different otolith biometric proportions (indexes), with a detailed anatomical description that helps to interpret its ontogenetic variation. These will contribute to knowing the morphometric relationships between fish total length (TL) and otolith (Tuset et al., 2003; Carvalho et al., 2015; Lombarte & Tuset, 2015; Giménez et al., 2016). *Micropogonias furnieri* sagitta morphometry has been described in previous works using some of these biometric indexes (Corrêa & Vianna, 1992; Volpedo & Echeverria, 1999; Waessle et al., 2003; Rossi-Wongtschowski et al., 2014). However, otolith ontogenetic development described by biometric proportions (indexes), (Tuset et al., 2008), has not been employed for the study of this species.

The whitemouth croaker *Micropogonias furnieri* is a coastal sciaenid (depth $<$ 50 m), benthic and euryhaline species. It is distributed from the Yucatan Peninsula (Mexico), along the east coast of South America to the Gulf of San Matías (Argentina) (41°S) (Isaac, 1988). It is particularly abundant in the Rio de la Plata estuary and it is the main resource of artisanal and industrial coastal fisheries in southern Brazil, Uruguay and Argentina (Norbis, 1995; Haimovici &d Ignacio, 2005; Norbis et al., 2006). It is a benthic predator that feeds mainly on crustaceans (crabs) and polychaetes (Puig, 1986; Masello et al., 2002; Olsson et al., 2013). Their spawning period spans from October to March (austral summer) and spawning occurs mainly in the frontal region of the Rio de la Plata estuary and the Uruguayan Atlantic coast (Macchi et al., 1996, 2003; Vizziano, 2002). Like many sciaenids, *M. furnieri* also produces sound. Two different types can be identified: advertisement calls during the reproductive season (only by males) and disturbance calls (by both sexes) (Tellechea et al., 2010). Due to its great economic importance, the whitemouth croaker is the most studied fish in the Rio de la Plata; however, there are few works that focus directly on the ontogenetic trajectory of the otolith using biometric indexes.

The goal of this work was knowing whether the whitemouth croaker sagitta otolith changes its shape in correlation with the five length categories of the fish, in addition to knowing if changes can be interpreted as morphological and functional variations of these biometric indexes.

Methods

The otoliths of *M. furnieri* caught in the Rio de la Plata estuary (fig. 1) were obtained in 2018 from samplings of industrial landings carried out by Direccion Nacional de Recursos Acuaticos (DINARA) and artisanal fisheries. The total length (TL) (cm) was recorded for each individual. Based on the distribution of lengths (Waessle

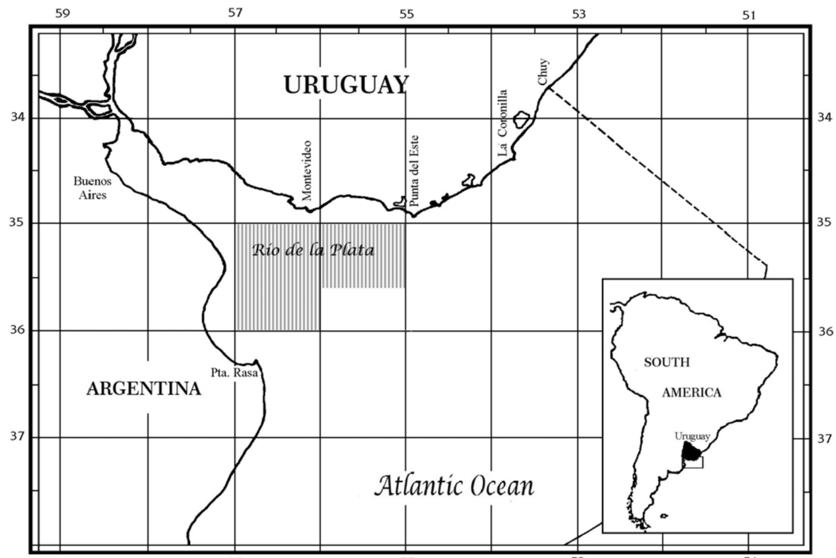

Figure 1. Study area indicating (shaded area) the sampling site in the Rio de la Plata estuary.

et al., 2003; Verocai, unpublished data) five groups (G) were determined (G1, 5.0 to 9.9 cm; G2, 10.0 to 14.9 cm; G3, 15.0 to 19.9 cm; G4, 20.0 to 29.9 cm; and G5, 30.0 cm and greater). Sagitta otoliths were extracted and cleaned with 98% isopropyl alcohol and dry-stored in paper envelopes.

Otolith measurements

The medial surface of the right sagitta otolith was photographed with a digital camera attached to a binocular loupe (Olympus SZ61, Lumenera Infinity 1 camera). All images were analyzed using Image J v.1.50 software. Otoliths were described morphologically, according to the terminology proposed by Tuset et al. (2008). Otolith length (OL) and maximum otolith width (OH) were recorded in centimeters. Perimeter (OP), area of the medial surface of the otolith (OA) and area of the acoustic sulcus (SA) were also measured (fig. 2). Potential equations were used so as to analyze the relationships between TL and OL and between TL and OH. Also, the relationships between SA and TL, SA and OH and SA/OA and TL were analyzed. For these relationships, allometric coefficients were calculated using the log-linear version of the equation $y = aX^b$, and tested for significant deviations from isometry ($b = 1$ for SA vs OA comparisons, and $b = 2$ for OA vs TL and SA vs TL).

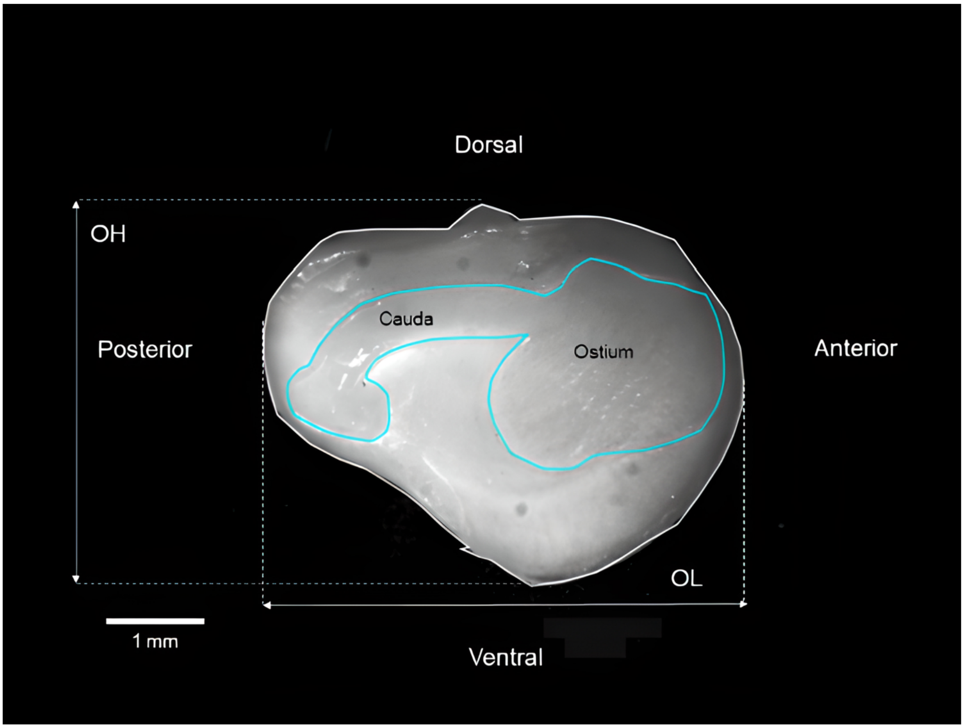

Figure 2. Measuremens made on the inner face of sagitta otoliths of *Micropogonias furnieri*: OH, otolith height; OL, otolith length; sulcus (ostium and cauda).

Six biometric indexes were used for each of the five length classes: rectangularity (REC) (describing variations in length and width with respect to area, with values of 1 when it is a perfect square) [OA/(OL × OH)]; circularity (CIR), which provides information on the similarity to a circle, with a value of 1 when it is a perfect circle ($4OA/OP^2$) (Takashimizu & Iiyoshi, 2016); aspect ratio (OH/OL), which indicates to what extent the otolith is circular or elongated (Francus, 1999); (OL/TL); relative surface area of the acoustic groove (SA/OA); and ellipticity (ELE) [(OL − OH)/(OL + OH)] (Tuset et al., 2003, 2008). In this work, an interpretation of the meaning of the indexes in relation to the ontogenetic development of otoliths was made considering anatomical changes and taking into account size categories.

Statistical analysis

In order to determine whether each of the shape indexes presented differences among length groups, the Kruskal-Wallis nonparametric analysis of variance was used (Sokal & Rohlf, 1995). If significant differences were found among groups, the Mann-Whitney test corrected by the Bonferroni criterion was used, so as to determine among which groups there were differences (Sokal & Rohlf, 1995). Canonical Discriminant Analysis (CDA) was used to compare the five length groups according to the six indexes. It was also used to determine which indexes contributed more to group discrimination. The CDA assesses the separation among individuals by stage through canonical and discriminant axes, evaluating the presence of groups in a multivariate space by maximizing the variations among them. Wilk's lambda was used to test the significant differences among groups (Manly & Alberto, 2016). Wilks' lambda is a measure of how well each function separates cases into groups. This statistic varies between 0 and 1; smaller values of Wilks' lambda indicate greater discriminatory ability of the function. This statistic follows an asymptotic Snedecor F distribution with the number of groups and variables in the numerator, and the degrees of freedom of the dispersion matrices in the denominator (Rao, 1952). PERMANOVA (Anderson, 2001) was also used to compare size groups according to the indexes and to test for significant differences among them. Mahalanobis distance was used and $p = 0.05$, in all cases. All statistical analyses were performed using Past software (Version 4.07) (Hammer et al., 2001).

Results

A total of 164 individuals were analyzed (length range: 5.5 to 46.0 cm). A general pattern of sagitta otolith shape was determined. They presented a dorsal areal depression, a rounded anterior region, a concave external face, and a convex internal face. It had no rostrum nor antirostrun. The acoustic sulcus had the following characteristics: position: supramedial; opening: pseudo-ostial; orientation: horizontal; morphology: heterosulcoid, laterally bent and concave ostium, and a tubular and notoriously curved cauda in adults, more than in young fish (fig. 2).

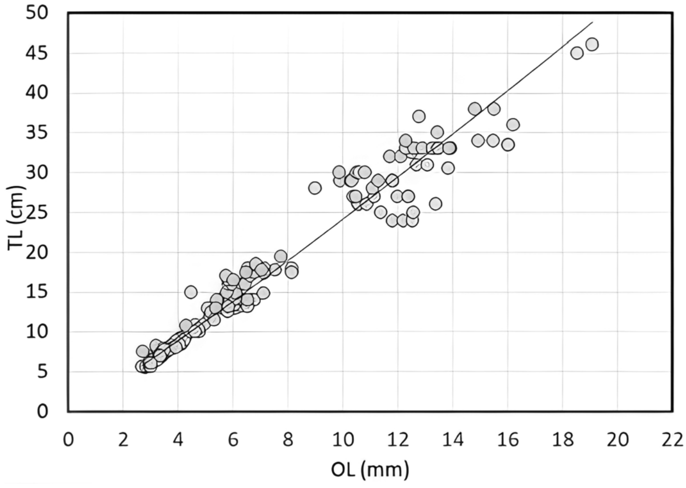

Figure 3. Relationship between otolith length and fish length (potential fit).

The OL vs TL relationship was potentially and significantly different from 1, (positive allometry) (TL $= 1.9804 \times$ OL$^{1.086}$, $S_b = 0.015$, $r = 0.985$, $N = 162$, $tc = 71.83$, $P < 0.00001$) and the OH vs TL was significant but the slope was equal to 1 (isometry) (fig. 3), (TL $= 3.041 \times$ OH$^{0.994}$, $S_b = 0.024$, $r = 0.955$, $N = 162$, $tc = 41.20$, $P < 0.00001$) (fig. 4). The average SA/OA ratio for all individuals was 0.393 ($\pm$0.0427). The SA presented a significant relationship with OA ($b = 1.063$, $S_b = 0.0067$, $r = 0.996$, $N = 162$, $tc = 157.61$, $P < 0.00001$) and also presented a positive and significant allometric growth. The relationship between OA and TL was positive and significant ($b = 1.739$, $S_b = 0.026$, $r = 0.982$, $N = 162$,

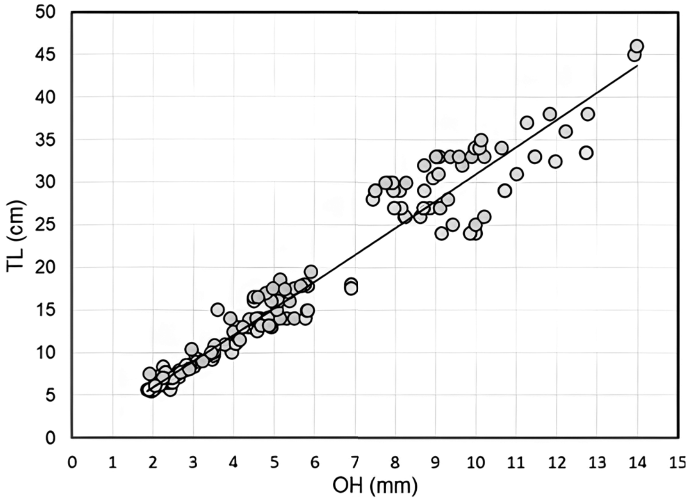

Figure 4. Relationship between otolith heigth and fish length (potential fit).

$tc = 67.02$, $P < 0.00001$) with negative and significant allometry. The relationship between SA and TL was positive and significant ($b = 1.855$, $S_b = 0.027$, $r = 0.983$, $N = 162$, $tc = 67.63$, $P < 0.0001$), with negative and significant allometry. These two variables are associated with $\log_{10}$ TL, showing a positive and significant relationship ($N = 162$, $b = 3.29$, $S_b = 0.331$, $r = 0.618$, $tc = 9.94$ $P < 0.00001$) indicating that size is an important predictor of the relationship between sulcus area and otolith area (SA/OA).

Biometric shape indexes

The Kruskal-Wallis test showed significant differences among length groups (G1 to G5) (table 1). The Mann-Whitney test showed there were significant differences between the different groups. The OL/TL index exhibits significant differences between G1, G2 and the other groups (G3, G4 and G5). The REC and SA/OA indexes showed significant differences between G1 and G2 to G5. In addition, G2 showed significant differences with the G4 and G5 groups, respectively. CIR showed differences between groups G1, G3 and G5 (fig. 5).

The CDA showed significant differences among the analyzed groups (Wilks $= 0.306$, $F_{24,548} = 9.227$, $P = 1.043\ \mathrm{e}{-27} < 0.05$, Pillai trace $P = 0.893$, $F_{24,260} = 7.67$, $P = 6.407\ \mathrm{e}{-23} < 0.05$). G1 and G2 present significant differences between them and also with the other groups ($P < 0.0001$), G3, G4 and G5 did not show significant differences among them ($P > 0.005$). This showed that the otolith biometric indexes presented the greatest changes in the early stages of the fish life (up to 15 cm) and little variation after this stage. The percentage of correct classification was 62.28%; when applying cross-validation, 56.29%. The first factor accumulated 76.22% of the explained variance, separated the length groups along the axis and, in turn, showed an order from negative to positive values. The second factor explained 14.19% (together more than 90%).

Table 1.
Kruskal-Wallis test for the six biometric indexes of the sagitta otolith of *Micropogonias furnieri*.

Index	*H*	*P*
CIR	21.28	0.000
REC	69.94	0.000
ELE	40.82	0.000
OH/OL	40.97	0.000
OL/TL	88.74	0.000
SA/OA	59.28	0.000

CIR, circularity; ELE, ellipticity; *H*, Kruskal-Wallis statistic; OH/OL, otolith height/otolith length; OL/TL, otolith length/total fish length; *P*, probability; REC, rectangularity; SA/OA, sulcus area/otolith area.

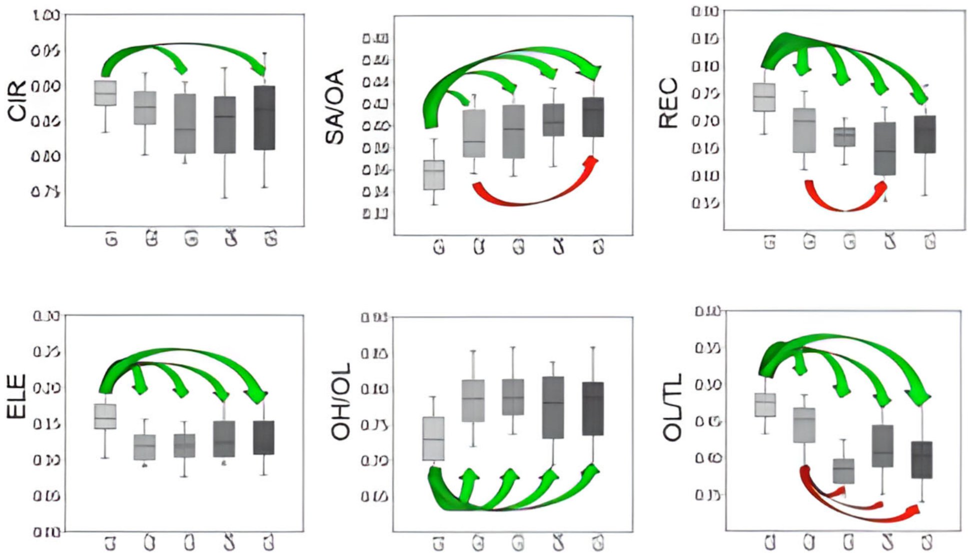

Figure 5. Mann-Whitney test corrected by the Bonferroni criterion for the six indexes separated by length group (G1 to G5): CIR, circularity; ELE, ellipticity; OH/OL, otolith height/otolith length; OL/TL, otolith length/total fish length; REC, rectangularity; SA/OA, sulcus area/otolith area. Arrows (G1, green; G2, red) indicate significant differences between groups (Past version 4.06).

Morphological coefficients (table 2) indicate that the rectangularity (REC), and aspect ratio (OL/TL and SA/OA) indexes contributed most to the discriminant function, characterizing individuals of length groups G1 and G2 (REC and OL/TL, respectively) and individuals of length groups G3, G4 and G5 (SA/OA). The REC index presented an acute angle with CIR, showed significant differences between

Table 2.
Morphometric variables that contributed the most to discriminate the groups with the first two principal factors of the discriminant analysis that explain 90.41% of the variability.

	Factors	
Indexes	F1	F2
CIR	−2.861	0.890
REC	−12.064	−4.201
ELE	6.202	−17.560
OH/OL	5.794	8.240
OL/TL	−11.030	11.838
SA/OA	10.544	1.674

CIR, circularity; ELE, ellipticity; *H*, Kruskal-Wallis statistic; OH/OL, otolith height/otolith length; OL/TL, otolith length/total fish length; *P*, probability; REC, rectangularity; SA/OA, sulcus area/otolith area.

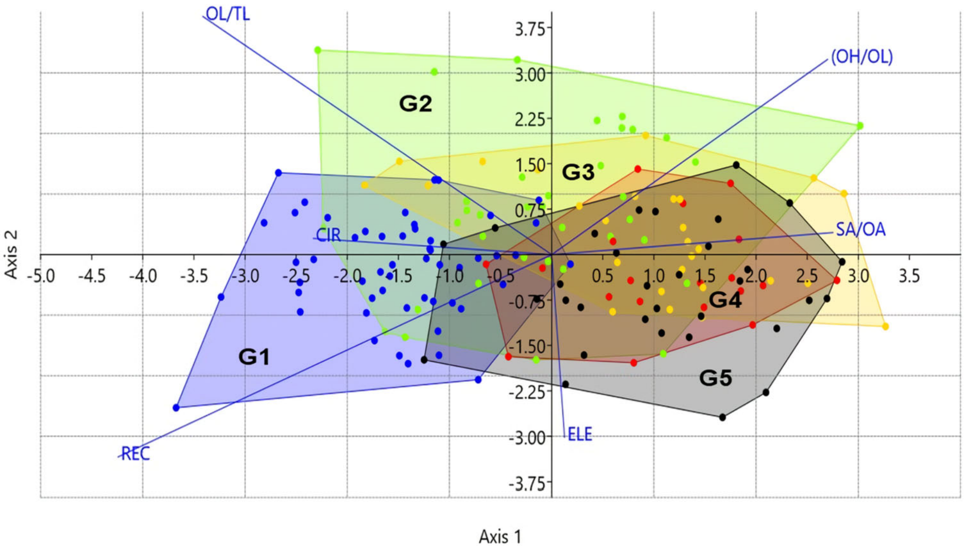

Figure 6. Discriminant analysis of the six indexes used and the five size groups (G1 to G5) considered in this work.

G3 and G5, and contributed to characterize G1 and G2, but to a lesser extent. Otoliths of the G2 group were located along axis 1 as a transition group between the G1 length group and the other groups (G3 to G5). The OL/TL and OH/OL indexes contributed to determine this group. These fish showed rapid otolith length growth, relative to growth in fish length, and a higher (OH/OL) index relative to the four remaining length groups. Length group G3 presented the slowest otolith growth with respect to fish length, being less rectangular and with a higher OH/OL ratio than those in length groups G1 and G2. The G4 and G5 length groups (older than two years) showed more elliptical, less circular and less rectangular otoliths, presenting slower otolith growth in relation to fish length growth and larger sulcus area in relation to the total otolith area (SA/OA, fig. 6). There were significant differences between size groups (PERMANOVA, Permutation N: 9999, $F = 7.086$, $P = 0.0001 < 0.05$) with similar results to those obtained by CDA.

Discussion

This work showed that the biometrical (morphological) changes occurring in the sagitta otolith of *M. furnieri* presented variations associated with different length groups. Otoliths are a permanent record of events occurring in a fish's life history. Furthermore, the way in which otolith growth (thickness and density of deposited material) is related to fish growth is complex, with a dependence between intrinsic otolith characteristics (size, morphology, and growth) and environmental factors (temperature, salinity) (Wilson, 1985, Mosegaard et al., 1988, Lombarte & Lleonart, 1993; Lombarte et al., 2003). These growth patterns in otoliths are reflected in variables associated with the size of the structure that can be quantified by biometric indexes (Secor et al., 1989; Campana & Casselman, 1993; Lombarte et al., 2003; Tuset et al., 2008).

For many fish species, size (length) prediction can be achieved from otolith length (Giménez et al., 2016). In *M. furnieri*, calcium deposition takes place in the anteroposterior axis in individuals larger than 10 cm (Volpedo & Etcheverría, 1999). This results in an OL vs TL relationship tending to isometry. However, in this study, the relationship that explained more than 98% of TL as a function of OL, was potential and allometric. For individuals smaller than 17 cm, the same relationship was found (Waessle et al., 2003). The potential relationship between OH and TL was isometric; this may be a consequence of material deposition in the transverse axis of the otolith, which takes place from early stages in the sagitta development (Volpedo & Etcheverria, 1999; Wassele et al., 2003). The whitemouth croaker shows rapid growth in the first two years of life (Norbis & Verocai, 2005; Borthagaray et al., 2011) as shown in the G1 and G2 length groups. By means of CDA, the indexes that most contributed to characterize these differences with the G3, G4 and G5 groups were REC and OL/TL. Rectangularity can be explained in terms of the development of accessory planes of growth in the otolith. It is a measure of 'boxiness', i.e., how well the otolith shape fills the area of its smallest enclosing rectangle (Galley et al., 2006). Growth of the primordium (otolith core) occurs at a higher rate along the anteroposterior axis than along the dorsal-ventral axis, (Volpedo & Etcheverría, 1999; Waessle et al., 2003). As a result, the otolith becomes longer with age. This deposition of material results in the inability to retain its initial oblong shape (exhibiting small changes in rectangularity with increasing size). Instead, it develops an irregular elongated shape that progressively fills less of its surrounding rectangle (Galley et al., 2006).

The littoral shallow waters (<6 m) and deeper waters (>6 m) of the Rio de la Plata estuary present different hydrodynamic and physical-chemical patterns, creating different environments (Guerrero et al., 1997; Nagy et al., 2002, 2008). The *M. furnieri* juvenile's preference for littoral waters (individuals < 20 cm), is related to the protection from predators, food availability and environmental conditions (Allega et al., 2020), all of which promote faster growth. This habitat preference varies during its ontogenetic development; larvae are associated with less brackish and more turbid waters (average < 1 psu) (Wells & Daborn, 1998; Braverman et al., 2009; Allega et al., 2020), and they make migratory movements, from breeding areas to deeper waters (Acha et al., 1999; Martinez & Retta, 2002; Vizziano et al., 2002; Jaureguizar et al., 2016). This is probably one of the causes that affect fish growth, producing modifications in their otoliths to adapt to different environmental conditions. High indexes (OL/TL) would be associated with groups that are found in high-turbidity environment, such as the waters of the Rio de la Plata estuary, needing to enhance other sensory systems related to acoustic communication (Gauldie, 1988; Paxton, 2000; Cruz & Lombarte, 2004). The differences found between groups G1 and G2 and the other groups might be related to different hearing sensitivities. From 12 cm onwards, juvenile *M. furnieri* migrate to deeper and more saline waters (Castello, 1986; Vizziano et al., 2002). Although adults (>32 cm) tolerate a wide range of salinity (0.5-33 psu), the highest concentrations are located in estuarine and marine waters (>25 psu) (Acha el at., 1999; García et al., 2010; Lorenzo et al., 2011). High values of the sphericity index (OH/OL) for all length groups analyzed (G1 to G5) would confirm the hypothesis that individuals with high values of sphericity frequent environments associated with soft substrates (Volpedo & Etcheverría, 2003). G2 fish showed a lower OL/TL ratio than G1 individuals. This indicates a slower growth of the otolith along its anteroposterior axis with respect to the total length of the fish. This variation in the OL/TL ratio may be a consequence of a faster somatic growth so as to access a better food supply and to have a greater possibility of predator escape. The diet composition of *M. furnieri* varies according to the size of the fish. The mouth size increases as the fish increases in length, making them able to access prey with more energetic value (Olsson et al., 2013). Different food items have been linked to morphological differences in teleosts otoliths (Aguirre & Lombarte, 1999). G2 corresponds to individuals that are less than one year old and show great spatial dispersion (Allega et al., 2020) as a consequence of a long reproductive period. This species reproduces from October to March, with two main spawning peaks (Pravia et al., 1995; Macchi et al., 1996; Viziano, 2002). Two groups arrive at the breeding area, with different size and growth characteristics (Norbis & Verocai, 2005), giving rise to the same cohort with a bimodal length distribution (Norbis & Verocai, unpublished data). Fish larger than 20 cm (G4 and G5) presented otoliths with very similar characteristics to each other. They behave as a single group, being more elliptical in shape, together with morphotype I defined by Santos et al. (2017) for individuals that inhabit the Rio de la Plata estuary.

In addition, it was shown that the G4 and G5 groups presented a larger acoustic groove in relation to the total otolith area (SA/OA). The increase in this index could be associated with the development of inner-ear perception and hearing ability. The mechanism of hearing in fish involves a physical interaction between the sagitta otolith and the macula. Therefore, larger fish have more sensory cells than smaller fish (Ramcharitar et al., 2001). The SA/OA ratio indirectly describes how much of the otolith surface area is in contact with the macula (M) through the acoustic sulcus. The ratio of macula area/otolith area (MA/OA) versus auditory threshold indicates that there is a strong relationship between MA/OA and otolith sensitivity at different frequencies. For this reason, fish with a relatively large sulcus may be more sensitive to sounds, with a relationship between the SA/OA ratio and the minimum auditory threshold (Platt & Popper, 1981; Gauldie, 1988). Sciaenids are known for their ability to produce sounds and are also called croakers (Fine et al., 1977, 2004; Myrberg, 1981; Saucier & Baltz, 1993; Ramcharitar et al., 2006). *Micropogonias furnieri* is one of the sciaenid species with the largest otoliths relative to body size (Verocai, unpublished data) and produces two types of sound (disturbance calls by both sexes; advertisement calls only by males) (Tellechea et al., 2010, 2011). The disturbance calls are produced at sizes from 10 cm, with a pronounced change (pulse duration and dominant frequency) from 20 cm, when the fish are still juveniles (size at first maturity, 32 cm, Norbis & Galli, 2013). From 20 cm onwards, these sound characteristics stabilize (Tellechea et al., 2010). The relationship between SA/OA may contribute to the change in sound perception, related to changes that were found in the G1, G2 and G3 groups. These changes in juvenile fish could contribute to maintaining their functionality as sound transducers (Gauldie, 1988). For the G4 and G5 groups, this change in the surface area of the acoustic groove (increased SA/OA rate) may be due to the need of containing more hair sensory cells. This is in order to respond to a change in sound perception experienced by individuals that, from 20 cm onwards, begin to frequent areas with similar characteristics of salinity and depth as spawning individuals (Macchi & Christiansen, 1996; Macchi et al., 1996; Jaureguizar et al., 2003). During the reproductive period, only adult males of *M. furnieri* produce advertisement calls (Tellechea et al., 2010, 2011). As a result, the ontogenetic changes described by means of otolith biometric indexes, would not only respond to environmental factors related to the development of the fish, but they are also associated with the sound perception of conspecifics, including those that participate in the reproduction (Tellechea et al., 2011).

Acknowledgements

We would like to thank the DINARA (Direccion Nacional de Recursos Acuáticos) and artisanal fishermen of Pajas Blancas who provide the otolith samples for this investigation. Also, thanks to Diego Norbis for improving the English text of the manuscript. Walter Norbis is supported by the ANII-SIN (Agencia Nacional de Investigaicon e Innovacion, Sistema Nacional de Investigadores). José Verocai and Walter Norbis also wish to thank the PEDECIBA-Biologia (Programa de Desarrollo de las Ciencias Básicas-Uruguay) for the funding provided. The datasets generated and analysed during the current study are available from the Department of Oceanography and Marine Ecology, Faculty of Sciences, University of the Republic, Montevideo, Uruguay.

References

Acha, E.M., Mianzán, H., Lasta, C.A. & Guerrero, R.A. (1999) Estuarine spawning of the whitemouth croaker *Micropogonias furnieri* (Pisces: Scienidae), in the Río de la Plata, Argentina. *Mar. Freshw. Res.*, 50, 57-65. DOI:10.1071/MF98045.

Aguirre, H. & Lombarte, A. (1999) Ecomorphological comparisons of sagittae in *Mullus barbatus* and *M. surmuletus*. *J. Fish Biol.*, 55, 105-114. DOI:10.1006/jfbi.1999.0974.

Allega, L., Braverman, M., Campodónico, S., Carozza, C.R., Cepeda, G.D., Colonello, J.H., Derisio, C., Di Mauro, R., Firpo, C.A., Gaitán, E.N., et al. (2020) Estado del conocimiento biológico pesquero de los principales recursos vivos y su ambiente; con relación a la exploración hidrocarburífera en la Zona Económica Exclusiva argentina y adyacencias. Instituto Nacional de Investigación y Desarrollo Pesquero (INIDEP), Mar del Plata, Argentina.

Anderson, M.J. (2001) A new method for non-parametric multivariate analysis of variance. *Aust. Ecol.*, 26, 32-46. DOI:10.1111/j.1442-9993.2001.01070.pp.x.

Baldás, M.I., Pérez Macri, G., Volpedo, A.V. & Echeverria, D.D. (1997) Morfología de la sagitta de peces marinos de la costa bonaerense de la Argentina. I. *Carangidae*, *Sciaenidae*, *Mullidae*. *Atlantica*, Rio Grande, 19, 99-112.

Borthagaray, A.I., Verocai, J. & Norbis, W. (2011) Age validation and growth of *Micropogonias furnieri* (Pisces – Sciaenidae) in a temporally open coastal lagoon (South-western Atlantic – Rocha – Uruguay) based on otolith analysis. *J. Appl. Ichthyol.*, 27, 1212-1217. DOI:10.1111/j.1439-0426.2011.01778.x.

Braverman, M.S., Acha, E.M., Gagliardini, D.A. & Rivarossa, M. (2009) Distribution of whitemouth croaker (*Micropogonias furnieri*, Desmarest 1823) larvae in the Río de la Plata estuarine front. *Est. Coast. Shelf Sci.*, 82, 557-565. DOI:10.1016/j.ecss.2009.02.018.

Campana, S.E. & Casselman, J.M. (1993) Stock discrimination using otolith shape analysis. *Can. J. Fish. Aquat. Sci.*, 50, 1062-1083. DOI:10.1139/f93-123.

Carvalho, B.M., Vaz-dos-Santos, A.M., Spach, H.L. & Volpedo, A.V. (2015) Ontogenetic development of the sagitta otolith of the anchovy, *Anchoa tricolor*, in a subtropical estuary. *Sci. Mar.*, 79, 409-418. DOI:10.3989/scimar.04218.31A.

Casselman, J.M. (1990) Growth and relative size of calcified structures of fish. *Trans. Am. Fish. Soc.*, 119, 673-688. DOI:10.1577/1548-8659(1990)119<0673:GARSOC>2.3.CO;2.

Castello, J.P. (1986) Distribución, crecimiento y maduración sexual de la corvina juvenil (*Micropogonias furnieri*) en el estuario de la “Lagoa dos Patos”, Brasil. *Physis*, 44, 21-36.

Chao, L.N. (1978) A basis for classifying Western Atlantic Sciaenidae (Teleostei: Perciformes). *NOAA Tech. Rep. Circul.*, 415, 1-65.

Corrêa, M.F.M. & Vianna, M.S. (1992) Catálogo de otólitos de Scianidae (Osteichthyes-Perciformes) do litoral do estado do Paraná, Brasil. *Neritica*, 7, 13-41. DOI:10.5380/rn.v7i1-2.85034.

Cruz, A. & Lombarte, A. (2004) Otolith size and its relationship with colour patterns and sound production. *J. Fish. Biol.*, 65, 1512-1525. DOI:10.1111/j.1095-8649.2004.00558.

Desmarest, A.-G. (1823) Première décade ichthyologique ou description complète de dix espèces de poissons nouvelles, ou imparfaitement connues, habitant la mer qui baigne les côtes de l’ile de Cuba. *Mer. Soc. Linn. Paris*, 2, 1-50.

Fay, R.F. & Coombs, S. (1983) Neural mechanisms in sound detection and temporal summation. *Hear. Res.*, 10, 69-92. DOI:10.1016/0378-5955(83)90018-7.

Fine, M.L., Winn, H.E. & Olla, B. (1977) Communication in fishes. In: T. Sebeok (Ed.) *How Animals Communicate*, pp. 472-518. Indiana University Press, Bloomington, IN, USA.

Fine, M.L., Schrinel, J. & Cameron, T.M. (2004) The effect of loading on disturbance sounds of the Atlantic croaker *Micropogonius undulatus*: air versus water. *J. Acoust. Soc. Am.*, 116, 1271-1275. DOI:10.1121/1.1736271.

Francus, P. (1999) Using image analysis to estimate quantitatively some microstructural parameters of detrital sediments. *Geol. Belg.*, 2, 173-180. DOI:10.20341/gb.2014.016.

Galley, E.A., Wright, P.J. & Gibb, F.M. (2006) Combined methods of otolith shape analysis improve identification of spawning areas of Atlantic cod. *ICES J. Mar. Sci.*, 63, 1710-1717.

García, M.L., Jaureguizar, A.J. & Protogino, L.C. (2010) From fresh water to the slope: fish community ecology in the Río de la Plata and the sea beyond. *Lat. Am. J. Aquat. Res.*, 38, 81-94. DOI:10.3856/vol38-issue1-fulltext-8.

Gauldie, R.W. (1988) Function, form and time-keeping properties of fish otoliths. *Comp. Biochem. Physiol. A Mol. Integr. Physiol.*, 91, 395-402. DOI:10.1016/0300-9629(88)90436-7.

Gauldie, R.W. & Nelson, D.G.A. (1990) Otolith growth in fishes. *Comp. Biochem. Physiol. A Mol. Integr. Physiol.*, 97, 119-135. DOI:10.1016/0300-9629(90)90159-P.

Giménez, J., Manjabacas, A., Tuset, V.M. & Lombarte, A. (2016) Relationships between otolith and fish size from Mediterranean and north-eastern Atlantic species to be used in predator-prey studies. *J. Fish Biol.*, 89, 2195-2202. DOI:10.1111/jfb.13115.

Guerrero, R.A., Acha, E.M., Framiñan, M.B. & Lasta, C.A. (1997) Physical oceanography of the Río de la Plata Estuary, Argentina. *Cont. Shelf. Res.*, 17, 727-742. DOI:10.1016/S0278-4343(96)00061-1.

Haimovici, M. & Ignacio, J.M. (2005) *Micropogonias furnieri*, (Desmarest, 1823). In: M.C. Cergole, A.O. Ávila-da-Silva & C.L.D.B. Rossi-Wongtchowski (Eds) *Análise das principais pescarias commerciais da Região Sudeste-Sul do Brasil: dinâmica populacional das espécies em explotaçao*, pp. 101-107. Instituto Oceanográfico, Sao Paulo, Brazil.

Hammer, Ø., Harper, D.A.T. & Ryan, P.D. (2001) PAST: paleontological statistics software package for education and data analysis. *Paleontol. Electronica*, 4, 9.

Isaac, V.J. (1988) Synopsis of biological data on the whitemouth croaker *Micropogonias furnieri* (Desmarest, 1823). *FAO Fish. Synopt.*, 150, 35 pp.

Jaureguizar, A.J., Menni, R., Bremec, C., Mianzan, H. & Lasta, C. (2003) Fish assemblage and environmental patterns in the Río de la Plata estuary. *Estuar. Coast. Shelf Sci.*, 56, 921-933. DOI:10.1016/S0272-7714(02)00288-3.

Jaureguizar, A.J., Solari, A., Cortés, F., Milessi, A.C., Militelli, M.I., Camiolo, M.D., Luz Clara, M. & García, M. (2016) Fish diversity in the Río de la Plata and adjacent waters: an overview of environmental influences on its spatial and temporal structure. *J. Fish. Biol.*, 89, 569-600. DOI:10.1111/jfb.12975.

Lombarte, A. & Lleonart, J. (1993) Otolith size changes related with body growth, habitat depth and temperature. *Environ. Biol. Fishes*, 37, 297-306. DOI:10.1007/BF00004637.

Lombarte, A. & Tuset, V. (2015) Morfometria de otólitos. In: A.V. Volpedo & A.M. Vaz-Dos-Santos (Eds) *Métodos de estudo com otólitos: princípios e aplicações*, pp. 269-302. CAFP-BA-PIESCI, Buenos Aires, Argentina.

Lombarte, A., Torres, G.J. & Morales-Nin, B. (2003) Specific *Merluccius* otolith growth patterns related to phylogenetics and environmental factors. *J. Mar. Biol. Assoc. UK*, 83, 277-281. DOI:10.1017/S0025315403007070h.

Lombarte, A., Chic, Ó., Parisi-Baradad, V., Olivella, R., Piera, J. & García-Ladona, E. (2006) A web-based environment from shape analysis of fish otoliths. The AFORO database. *Sci. Mar.*, 70, 147-152. DOI:10.3989/scimar.2006.70n1147.

Lorenzo, M.I., Díaz de Astarloa, J.M., Norbis, W. & Cousseau, M.B. (2011) Long term fish assemblages as units of management in a temperate estuary (Rio de la Plata – SW Atlantic Ocean). *Braz. J. Oceanogr.*, 59, 43-59.

Macchi, G.J. & Christiansen, H.E. (1996) Análisis temporal del proceso de maduracion y determinación de la incidencia de atresias en la corvina rubia (*Micropogonias furnieri*). *Frente Mar.*, 11, 73-83.

Macchi, G.J., Acha, M.E. & Lasta, C.A. (1996) Desove y fecundidad de la corvina rubia *Micropogonias furnieri* Desmarest, 1823 del estuario del Río de la Plata, Argentina. *Bol. Inst. Esp. Oceanogr.*, 12, 99-113.

Macchi, G.J., Acha, E.M. & Militelli, M.I. (2003) Seasonal egg production of whitemouth croaker (*Micropogonias furnieri*) in the Río de la Plata estuary, Argentina-Uruguay. *Fish. Bull.*, 101, 332-342.

Manly, B.F.J. & Alberto, J.A.N. (2016) *Multivariate Statistical Methods: a Primer*. 4th Edition. Chapman and Hall/CRC, New York, NY, USA. DOI:10.1201/9781315382135.

Martinez, G. & Retta, S. (2002) Characterization of croaker (*Micropogonias furnieri*) breeding areas along the Uruguayan coastal zone. In: D. Vizziano, P. Puig, C. Mesones & G. Nagy (Eds) *The Río de la Plata Research to Manage the Environment, Fish Resources and the Fishery in the Saline Front*, pp. 139-147. Ecoplata Program, Montevideo, Uruguay.

Massello, A., Scarabino, F., Gamarra, M. & Menafra, R. (2002) Study of feeding habits and stomach contents of *Micropogonias furnieri*. In: D. Vizziano, P. Puig, C. Mesones & G. Nagy (Eds) *The Río de la Plata Research to Manage the Environment, Fish Resources and the Fishery in the Saline Front*, pp. 147-161. Ecoplata Program, Montevideo, Uruguay.

Morales-Nin, B. (1987) The influence of environmental factors on microstructure of otoliths of three demersal fish species caught off Namibia. *S. Afr. J. Mar. Sci.*, 1, 255-262. DOI:10.2989/025776187784522207.

Mosegaard, H., Svedäng, H. & Taberman, K. (1988) Uncoupling of somatic and otolith growth rates in Arctic char (*Salvelinus alpinus*) as an effect of differences in temperature response. *Can. J. Fish. Aquat. Sci.*, 45, 1514-1524. DOI:10.1139/f88-180.

Myrberg, A.A. (1981) Sound communication and interception in fishes. In: W.N. Tavolga, A.N. Popper & R.R. Fay (Eds) *Hearing and Sound Communication in Fishes*, pp. 395-428. Springer-Verlag, New York, NY, USA. DOI:10.1007/978-1-4615-7186-5_20.

Nagy, G.J., Pshennikov-Severova, V. & Robatto, P. (2002) Monthly variability of salinity in the frontal zone of the Río de la Plata off Montevideo, in response to consecutive ENSO fluctuations and the flow of the Uruguay River (1998-2000). In: D. Vizziano, P. Puig, C. Mesones & G.J. Nagy (Eds) *The Río de la Plata: research to manage the environment, fish resources and the fishery in the saline front*, pp. 21-30. EcoPlata Program, Montevideo, Uruguay.

Nagy, G.J., Severov, D.N., Pshennikov, V.A., De los Santos, M., Lagomarsino, J.J., Sans, K. & Morozov, E.G. (2008) Río de la Plata estuarine system: relationship between river flow and frontal variability. *Adv. Space Res.*, 41, 1876-1881. DOI:10.1016/j.asr.2007.11.027.

Norbis, W. (1995) Influence of wind, behaviour and characteristics of the croacker (*Micropogonias furnieri*) artisanal fishery in the Rio de la Plata (Uruguay). *Fish. Res.*, 22, 43-58. DOI:10.1016/0165-7836(94)00310-S.

Norbis, W. & Galli, O. (2013) Spatial co-occurrence of two Sciaenid species (*Micropogonias furnieri* and *Cynocion guatucupa*) subject to fishing in the Río de la Plata and oceanic coast of Uruguay: ecological or technological interdependence? *Bol. Inst. Pesca*, 39, 137-148.

Norbis, W. & Verocai, J. (2005) Presence of two whitemouth croaker (*Micropogonias furnieri*, Pisces: Sciaenidae) groups in the Río de la Plata spawning coastal area as consequence of reproductive migration. *Fish. Res.*, 74, 134-141. DOI:10.1016/j.fishres.2005.03.005.

Norbis, W., Paesh, L. & Galli, O. (2006) Los recursos pesqueros de la costa de Uruguay: ambiente, biología y gestión. In: R. Menafra, L. Rodríguez-Gallego, F. Scarabino & D. Conde (Eds) *Bases para la conservación y el manejo de la costa Uruguaya*, pp. 197-209. Vida Silvestre Uruguay, Montevideo, Uruguay.

Olsson, D., Forni, F., Saona, G., Verocai, J. & Norbis, W. (2013) Temporal feeding habits of the whitemouth croaker *Micropogonias furnieri* in a shallow coastal lagoon (southwestern Atlantic Ocean, Uruguay). *Cienc. Mar.*, 39, 265-276. DOI:10.7773/cm.v39i3.2181.

Paxton, J.R. (2000) Fish otoliths: do sizes correlate with taxonomic group, habitat and/or luminescence? *Phil. Trans. R. Soc. Lond. B Biol. Sci.*, 355, 1299-1303. DOI:10.1098/rstb.2000.0688.

Platt, C. & Popper, A.N. (1981) Fine structure and function of the ear. In: W.N. Tavolga, A.N. Popper & R.R. Fay (Eds) *Hearing and Sound Communication in Fishes*, pp. 3-38. Springer Verlag, New York, NY, USA. DOI:10.1007/978-1-4615-7186-5_1.

Popper, A.N. & Platt, C. (1993) Inner ear and lateral line. In: D. Evans (Ed.) *The Physiology of Fishes*, pp. 99-136. CRC Press, Boca Ratón, FL, USA.

Pravia, M.A., García, C., Ares, L. & Berois, N. (1995) Estimación de la fecundidad y determinación del tipo de desova de la corvina blanca *Micropogonias furnieri* (Teleostei: Sciaenidae). *Rev. Bras. Biol.*, 55, 13-25.

Puig, P. (1986) Análisis de contenidos estomacales de corvina blanca (*Micropogon opercularis*) (Sciaenidae, Perciformes). Verano 1984. *Com. Téc. Mix. Fr. Mar.*, 1, 333-340.

Ramcharitar, J. & Popper, A.N. (2004) Masked auditory thresholds in sciaenid fishes: a comparative study. *J. Acoust. Soc. Am.*, 116, 1687-1691. DOI:10.1121/1.1771614.

Ramcharitar, J., Higgs, D.M. & Popper, A.N. (2001) Sciaenid inner ears: a study in diversity. *Brain Behav. Evol.*, 58, 152-162. DOI:10.1159/000047269.

Ramcharitar, J., Deng, X., Ketten, D. & Popper, A.N. (2004) Form and function in the unique inner ear of a teleost: the silver perch (*Bairdiella chrysoura*). *J. Comp. Neurol.*, 475, 531-539. DOI:10.1002/cne.20192.

Ramcharitar, J., Higgs, D.M. & Popper, A.N. (2006) Audition in sciaenid fishes with different swim bladder-inner ear configurations. *J. Acoust. Soc. Am.*, 119, 439-443. DOI:10.1121/1.2139068.

Rao, C.R. (1952) *Advanced Statistical Methods in Biometric Research*. Wiley and Sons, Inc., New York, NY, USA.

Rossi-Wongtschowski, C.L.D.B., Siliprandi, C.C., Brenha, M.R., Gonsales, S.D.A., Santificetur, C. & Vaz-dos-Santos, A.M. (2014) Atlas of marine bony fish otoliths (sagittae) of Southeastern-Southern Brazil Part I: Gadiformes (Macrouridae, Moridae, Bregmacerotidae, Phycidae and Merlucciidae); Part II: Perciformes (Carangidae, Sciaenidae, Scombridae and Serranidae). *Braz. J. Oceanogr.*, 62, 1-103.

Santos, R.S., Costa, M.C., Queiroz, C. & Gerson, F. (2017) Different sagitta otolith morphotypes for the whitemouth croaker *Micropogonias furnieri* in the Southwestern Atlantic coast. *Fish. Res.*, 195, 222-229. DOI:10.1016/j.fishres.2017.07.027.

Saucier, M.H. & Baltz, D.M. (1993) Spawning site selection by spotted seatrout, *Cynoscion nebulosus*, and black drum, *Pogonias cromis*, in Louisiana. *Environ. Biol. Fish.*, 36, 257-272. DOI:10.1007/BF00001722.

Secor, D.H., Dean, J.M. & Baldevarona, R.B. (1989) Comparison of otolith growth and somatic growth in larval and juvenile fishes based on otolith length/fish length relationships. *Rapp. P-V Réun. Cons. Int. Explor. Mer.*, 191, 431-438.

Simkiss, K. (1974) Calcium metabolism of fish in relation to ageing. In: T.B. Bagenal (Ed.) *Ageing of Fish*, pp. 1-12. Unwin Brothers, Surrey, UK.

Sokal, R.R. & Rohlf, F.J. (1995) *Biometry: the Principles and Practice of Statistics in Biological Research*. 3rd Edition. WH Freeman and Company, New York, NY, USA.

Takashimizu, Y. & Iiyoshi, M. (2016) New parameter of roundness *R*: circularity corrected by aspect ratio. *Prog. Earth Plan. Sci.*, 3, 2. DOI:10.1186/s40645-015-0078-x.

Tellechea, J.S., Martinez, C., Fine, M.L. & Norbis, W. (2010) Sound production in whitemouth croaker and relationship between fish size and disturbance call characteristics. *Environ. Biol. Fish.*, 89, 163-172. DOI:10.1007/s10641-010-9709-7.

Tellechea, J.S., Bouvier, D. & Norbis, W. (2011) Spawning sounds in whitemouth croaker (Sciaenidae): seasonal and daily cycles. *Bioacoustics*, 20, 159-168. DOI:10.1080/09524622.2011.9753641.

Tuset, V.M., Lozano, I.J., González, J.A., Pertusa, J.F. & García-Díaz, M.M. (2003) Shape indices to identify regional differences in otolith morphology of comber, *Serranus cabrilla* (L.,1758). *J. Appl. Ichthyol.*, 19, 88-93. DOI:10.1046/j.1439-0426.2003.00344.x.

Tuset, V.M., Lombarte, A. & Assis, C.A. (2008) Otolith atlas for the western Mediterranean, north and central eastern Atlantic. *Sci. Mar.*, 72S1, 7-198. DOI:10.3989/scimar.2008.72s17.

Tuset, V.M., Otero-Ferrer, J.L., Siliprandi, C., Manjabacas, A., Marti-Puig, P. & Lombarte, A. (2021) Paradox of otolith shape indices: routine but overestimated use. *Can. J. Fish. Aquat. Sci.*, 78, 681-692. DOI:10.1139/cjfas-2020-0369.

Vignon, M. (2012) Ontogenetic trajectories of otolith shape during shift in habitat use: interaction between otolith growth and environment. *J. Exp. Mar. Biol. Ecol.*, 420, 26-32. DOI:10.1016/j.jembe.2012.03.021.

Vignon, M. (2015) Disentangling and quantifying sources of otolith shape variation across multiple scales using a new hierarchical partitioning approach. *Mar. Ecol. Prog. Ser.*, 534, 163-177. DOI:10.3354/meps11376.

Vignon, M. & Morat, F. (2010) Environmental and genetic determinant of otolith shape revealed by a non-indigenous tropical fish. *Mar. Ecol. Progr. Ser.*, 411, 231-241. DOI:10.3354/meps08651.

Vizziano, D. (2002) Reproductive cycle of the white croaker *Micropogonias furnieri* in the frontal zone of the Río de la Plata. In: D. Vizziano, P. Puig, C. Mesones & G.J. Nagy (Eds) *The Río de la Plata research to manage the environment, fish resources and fishery in the saline front*, pp. 103-111. EcoPlata Program, Montevideo, Uruguay.

Vizziano, D., Saona, G., Franco, J. & Nagy, G.J. (2002) Environmental characterization of the spawning area of the white croaker, *Micropogonias furnieri*, in the frontal zone of the Río de la Plata. In: D. Vizziano, P. Puig, C. Mesones & G.J. Nagy (Eds) *The Río de la Plata: research to manage the environment, fish resources and the fishery in the saline front*, pp. 113-124. EcoPlata Program, Montevideo, Uruguay.

Volpedo, A. & Echeverría, D.D. (2003) Ecomorphological patterns of the sagitta in fish on the continental shelf off Argentine. *Fish. Res.*, 60, 551-560. DOI:10.1016/S0165-7836(02)00170-4.

Volpedo, A.V. & Echeverría, D.D. (1999) Diferencias morfológicas en las sagittae de adultos de *Micropogonias furnieri* (Sciaenidae) de la costa bonaerense. *Thalassas*, 15, 19-24.

Waessle, J.A., Lasta, C.A. & Favero, M. (2003) Otolith morphology and body size relationships for juvenile Sciaenidae in the Río de la Plata estuary (35-36°S). *Sci. Mar.*, 67, 233-240. DOI:10.3989/scimar.2003.67n2233.

Wells, P.G. & Daborn, G.R. (1998) *Rio de la Plata. An environmental overview; an EcoPlata Project background report*. Dalhousie University, Halifax, Nova Scotia, Canada.

Wilson, R.R. (1985) Depth-related changes in sagitta morphology in six macrourid fishes of the Pacific and Atlantic Oceans. *Copeia*, 1985, 1011-1017. DOI:10.2307/1445256.