E-ISSN: 2073-3720
https://doi.org/10.33997/j.afs.2024.37.4.005

Stock Identification of Brown-Striped Snapper, *Lutjanus vitta* (Qouy and Gaimard, 1824), Inferred From Otolith Shape Analysis off the Coast of Iligan Bay, Mindanao, Philippines

MARICEL TUMAMPOS GUMOLOC[1], IVANE R. PEDROSA-GERASMIO[1,2,*]
[1]Marine Science Department, College of Science and Mathematics, Mindanao State University Iligan Institute of Technology, Tibanga, Iligan City, Philippines
[2]Marine Biodiversity Laboratory, Premier Research Institute of Science and Mathematics, Mindanao State University Iligan Institute of Technology, Tibanga, Iligan City, Philippines

*E-mail: ivane.gerasmio@g.msuiit.edu.ph |Received: 09/01/2024; Accepted: 17/10/2024

Abstract

Otolith shape analysis is an effective method that has been used to separate stocks based on their phenotypic traits. The brown-striped snapper, *Lutjanus vitta* (Quoy and Gaimard, 1824), is an important component of the fish catch in Iligan Bay, Mindanao, Philippines. Trends in catch and catch per unit effort, however, had been declining because of overfishing of this economically significant species as a result of continuous human exploitation. To accurately identify and manage fish, one must be aware of the stock structure of a species. The aim of this study was to contribute to the knowledge on the stock identification of *L. vitta* using otolith morphometrics and shape analysis. A total of 90 individuals of *L. vitta* were collected from the three selected sampling locations in Iligan Bay: Iligan City, Kapatagan, and Oroquieta City. Results of ANOVA tests investigating otolith shape differences between areas showed that mean values among sites differed significantly for the following shape indices: ellipticity ($F = 20.93$, $P < 0.001$), aspect ratio ($F = 19.45$, $P < 0.001$), and circularity ($F = 24.72$, $P < 0.001$). A canonical analysis of principal components verified the separation of otolith morphometric parameters among locations, with 73.9 % of the variation explained by the first axis (PC1). However, otolith shape did not differ significantly between sexes ($P > 0.05$). Furthermore, the differences in otolith shape among the three locations suggests a spatial structuring of *L. vitta* in Iligan Bay ($P = 0.001$) and necessitates local management and policies to enable sustainable management of the fisheries.

Keywords: *Lutjanus vitta*, otolith shape, stock identification, fisheries, Iligan Bay

Introduction

Fisheries management requires a thorough understanding of the spatial and temporal variation in fish stock structures among both target and non-target species (Begg and Waldman, 1999; Cadrin et al., 2014). A fish species' response to fishing pressure and environmental changes can be understood through reliable scientific data on population dynamics, which is essential for species resilience (Kerr et al., 2017). Stock, in the context of fisheries, refers to a population of organisms with a shared gene pool that is distinct enough to be regarded as a self-sustaining system suitable for management (Larkin, 1972; Waldman, 2005; Cadrin et al., 2023). The concept of a stock is crucial for understanding the dynamics and management of fish populations, as different stocks of the same species can exhibit varying migration patterns, growth, reproductive dynamics, and other characteristics that require targeted management strategies (Kallio-Nyberg et al., 2002; Begg and Cadrin, 2016). As a result, stock identification develops into an important multidisciplinary area for fisheries management, incorporating several complementary methodologies such as genetic, mark-recapture methods, biometrics, stable isotopes, morphology and chemical investigations of tissues and hard components (Cadrin and Friedland, 1999; Begg and Cadrin, 2016; Barnuevo et al., 2023).

The mechanism of fish population differentiation and habitat connectivity in the marine environment is complex, shaped by fish behaviour (such as natal homing, spawning, and life history strategies) as well as oceanographic and environmental features (Pedrosa-Gerasmio et al., 2015; Marini et al., 2021). These factors can influence fish populations from the earliest stages of life by promoting egg and larval retention or dispersal, acting as significant selective pressures for stock identification. This results in various population units that may respond differently to exploitation and need to be managed independently (Galli and Norbis, 2013).

Otolith chemistry (Koolkalya et al., 2020) and morphometric parameters and shape analysis have been used as a tool for discriminating between fish stocks (Soeth et al., 2019; Wujdi et al., 2022). Otoliths are crucial organs that are involved in a variety of fish functions, particularly hearing and balancing (Popper and Coombs, 1982). They maintain track of the many stages of fish life in the surrounding environment. Otoliths can therefore be thought of as the "flight recorders" of fish, similar to the black box of an airplane (Lecomte-Finiger, 1992). Due to their constant growth and metabolic inertness, otoliths serve as significant natural tags in research on fish stock identification. Information contained in the otolith can be used to assess a fish species' stock structure, spatial distribution, and range in an environment. On the other hand, chemical and otolith-form stock delineation methods are available (Koolkalya et al., 2020).

Otolith morphometric parameters and shape analysis, a species-specific and stable method, effectively distinguish stocks or population components based on phenotypic traits that change minimally with growth (Campana and Casselman, 1993; Soeth et al., 2019; Wujdi et al., 2022). Otoliths are vital organs involved in fish functions such as hearing and balancing (Popper and Coombs, 1982). Due to their continuous growth and metabolic inertness, otoliths serve as natural tags in fish stock research. Geographic variations in otolith morphology likely reflect population differences, indicating that distinct geographical areas are partially occupied during fish life histories (Casselman et al., 2011). Several stock discrimination studies have used otolith morphology (Cardinale et al., 2004; Stransky and MacLellan, 2005), with classification success rates for interstock separation varying from 60 % to 95 % depending on the species. Otolith shape analysis has been extensively and successfully used in stock identification studies of many marine fish species. The quantitative description of the otolith contour is made possible by the otolith shape analysis, which produces two-dimensional images that can be statistically compared (Lestrel, 1997).

Many studies have documented a global decline in marine fish populations, including in Southeast Asia (Golden et al., 2016; Teh et al., 2017). For instance, numerous snapper species are also declining or facing depletion, reflecting global trends in marine fisheries (Morris et al., 2000; Freitas et al., 2011). In the Philippines, there are 56 species of snappers, locally known as 'maya-maya', out of approximately 110 species reported globally (see FishBase). One commercially important snapper species, the brown-striped snapper, *Lutjanus vitta* (Quoy and Gaimard, 1824), has already been reported to be overexploited in some areas in the Philippines (Palla et al., 2016). *Lutjanus vitta* is a medium-sized marine fish primarily found on reefs and widely distributed across the Indo-Pacific region, from the Seychelles to New Caledonia in the south and the Ryukyu Islands in the north (Allen, 1985). It inhabits seas ranging from 40 to 120 m in depth, with larger specimens typically found in deeper waters (Davis and West, 1992).

With declining fish catches, effective management of marine fish resources is crucial to replenish fish resources. It is essential to differentiate fish populations as distinct stocks to avoid consequences such as reduced genetic diversity, diminished spawning capabilities, and ecological challenges (Hilborn and Walters, 2004). Fish stock identification is also essential for ecological insights into population dynamics (Mérigot et al., 2007; Halim et al., 2021).

Therefore, this study investigated stock identification of the brown-striped snapper *L. vitta* from Iligan Bay, Mindanao, Philippines. Specifically, we examined variations in otolith morphometric parameters and shape among fish sampled from three locations in Iligan Bay, Mindanao, Philippines: Iligan City, Kapatagan, and Oroquieta City.

Materials and Methods

Ethical approval

This study diligently adhered to relevant guidelines, whether international, national, or institutional, regarding the care and use of animals. The fish samples used were obtained from vendors who practice sustainable and humane fishing, reducing the negative effects on animal welfare.

Study site

Iligan Bay is a bay on the Philippine Island of Mindanao. It is the largest bay in the western portion of Northern Mindanao that connects to the Bohol Sea, with an extent of 1,811.16 km^2(PSA, 2018) and surrounded by the three provinces of Misamis Oriental, Lanao del Norte, and Misamis Occidental (Fig. 1).

Sample collection

A total of 90 non-live fresh samples of *L. vitta* were purchased from the fish markets of Iligan City in Lanao del Norte, Oroquieta City in Misamis Occidental, and lastly, in the municipality of Kapatagan, Lanao del Norte, from April to May 2023. Interviews with the fish

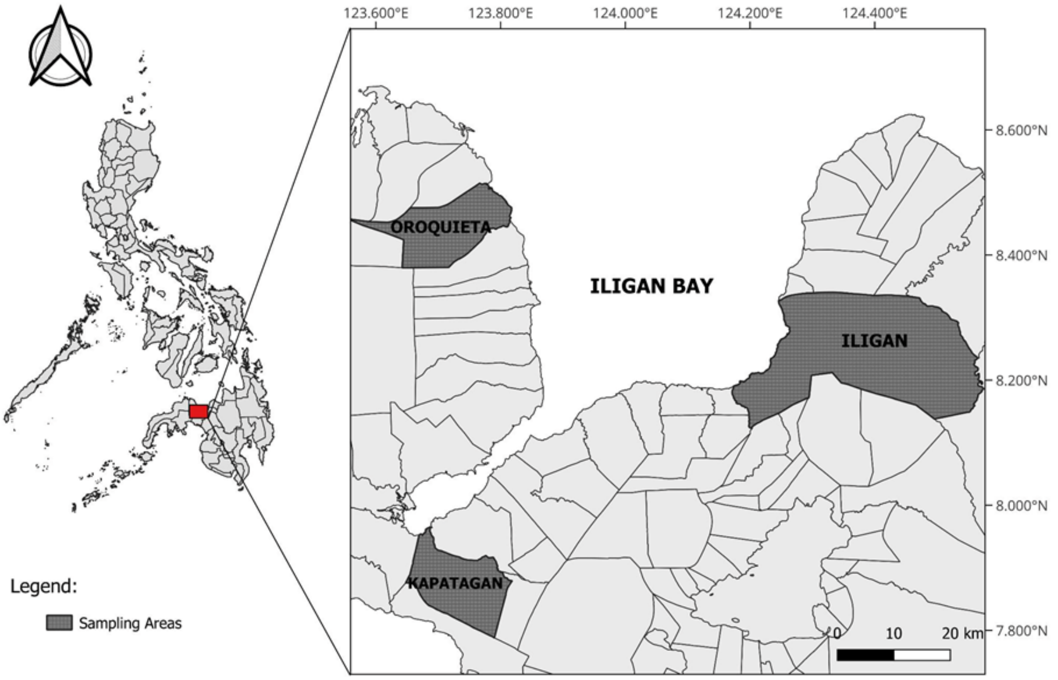

Fig. 1. Map with the sampling locations from which samples of *Lutjanus vitta* were collected for otolith shape analysis. The dark grey colour indicates the selected sites in Iligan Bay, Mindanao, Philippines.

vendors in the market were conducted prior to sample collection to verify that the specimen originated from the designated sampling locations. All the samples were stored frozen and brought to the laboratory for further analysis. Upon transport to the laboratory, the standard length (SL) and total length (TL) and fork length (FL) of fish individuals were measured to the nearest 0.1 cm using a digital caliper (Ole, Philippines), while the total weight (BW) was taken in 0.1 g using a digital weighing scale (Bestguard, China).

Otolith extraction and storage

The largest of the three pairs of otoliths, sagittate, are the ones that are typically employed for otolith analysis (Campana and Casselman 1993). Consistent with the methods in Barnuevo et al. (2023), the sagittal otoliths were extracted from the samples, first, by locating the optic capsule in the post-ventral portion of the neurocranium. A shallow incision was carefully made in the central region of the optic capsule, gently breaking it open to reveal the sagittal otoliths, which were subsequently extracted using fine forceps. The collected otoliths were pre-cleaned with distilled water, submerged it in for 20 to 30 seconds to remove remaining blood and muscle debris, and immersed in absolute ethanol until air-dried. And then, the samples were stored in small plastic vials. The weights of individual otoliths (OW) were measured using analytical balance (BSM220.4, ZhuoJin Electronic, China).

Image acquisition

Otoliths were positioned on their non-sulcus side with the rostrum pointed to the left. Otoliths were photographed using a camera phone (Realme 8 5G, China) in two dimensions using a stereomicroscope at 2× magnification. Bright two-dimensional objects were produced utilising high contrast digital photographs obtained using reflected light against a dark background.

Shape analysis

To visualise the differences in the otolith shape of brown-striped snapper from Iligan Bay, images were subjected to shape analysis using the shapeR 0.1-5 package (Libungan and Pálsson, 2015) in RStudio version 4.2.1 to generate Wavelet and Fourier coefficients. Additional packages used were vegan 2.6-4 (Oksanen et al., 2022) and ggplot2 3.3.3 (Wickham, 2016) for creating high resolution plots. Due to the difficulties encountered in detecting the outlines from raw otolith images, even when increasing the "detect. outline" threshold (0.1–0.5), the otolith images were processed using Adobe Photoshop CS6 13.0.6 (Adobe, USA) to replace the background with a uniform black colour (Fig. 2). This added step increased the detectability of the otolith outlines (at threshold = 0.1–0.2), as the possible effects of glares on the otolith surface was eliminated.

Otolith morphometrics

Eight parameters that indicate shape indices were calculated, including five size-related parameters (otolith length, OL; otolith height, OH; otolith perimeter, OP; otolith area, OA; and otolith weight, OW) (Table 1). Using an analytical balance, the OW was calculated to the closest 0.0001 g.

Allometry correction

Fish size was shown to be strongly correlated with

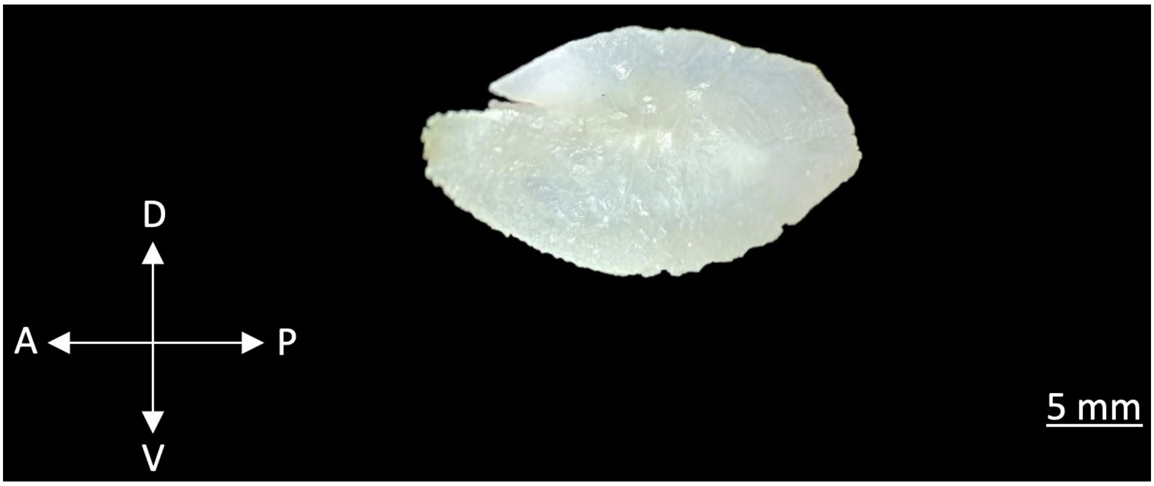

Fig. 2. A representative image of the right otolith from *Lutjanus vitta* that was used to process data in RStudio using the shapeR package. The otolith is shown as A (anterior), P (posterior), V (ventral), and D (dorsal) on the directional map (cross).

Table 1. Size parameters and shape indices used in this study based on Tuset et al. (2003) and Barnuevo et al. (2023).

Size parameters	Shape indices	Equation
Otolith length (OL)	Rectangularity (RE)	RE = OA/(OL × OH)
Otolith height (OH)	Squareness (SQ)	SQ = OA/(OL × OW)
Otolith perimeter (OP)	Ellipticity (EL)	EL = OL - OH/ OL + OH
Otolith area (OA)	Roundness (RO)	RO = 4OA/лOL^2
Otolith weight (OW)	Aspect ratio (AR)	AR = OL/OH
	Form factor (FF)	FF = 4лOA/OP^2
	Compactness (CO)	CO = OP^2/OA
	Circularity (CI)	CI = OP/OA^2

A: area (mm^2), P: perimeter (mm), OL: length (mm) and OW: width (mm).

otolith morphometry and shape indices of *L. vitta*. Correction of data was performed to eliminate the impact of allometry on the final otolith characteristics to provide more reliable comparisons. The formula, which is based on the work of Deepa et al. (2018), is as follows:

$$\mathrm{Ms} = \mathrm{Mo}(\underline{x}/x)^{b}$$

where Ms is the corrected otolith parameter, Mo is the observed otolith size or shape parameter, $\underline{x}$ is the mean total length of all the fish specimens in the group, and x is the total length of the specific fish specimen.

Statistical methods

The mean shape of the otoliths from brown-striped snapper *L. vitta* was obtained using the shapeR (Libungan and Pálsson, 2015) and vegan (Oksanen et al., 2022) packages in RStudio using the "detect. outline" function and were plotted using the Wavelet and Fourier coefficients. To further characterise the differences in mean otolith shapes across the sampling stations, an ANOVA-like permutation test with the vegan package (anova.cca) was also run for both smoothed and non-smoothed outlines to assess the significance of constraints using 1000 permutations. To further support these results, a constrained ordination (Canonical Analysis of Principal Coordinates) was performed using the Wavelet coefficients, which are visualised using the ggplot2 package (Wickham, 2016). Multivariate Analysis of Variance implemented in R using RStudio was used to test the significant variations in otolith shape for both sexes.

Results

Mean length and weight

The mean length and weight of *L. vitta* species in three different areas are shown in Table 2. A total of 30 individuals from each location were collected and examined. Kapatagan had the greatest mean length and weight recorded, with a mean length of 20.32 cm and mean weight of 149.14 g, followed by Iligan City species with 19.26 cm mean length and 112.50 g fish weight, and lastly, species from Oroquieta City, which has a mean fish length of 18.47 cm and an average of 100.13 g in fish weight.

Shape indices

Significant shape differences across Iligan, Kapatagan, and Oroquieta were seen in the mean values of three of these indices ($P < 0.001$), i.e., aspect ratio, circularity and ellipticity, while the five other indices, however, did not differ at all or showed no difference (Table 3). Mean values among sites differed

Table 2. Average total length and weight of *Lutjanus vitta* collected from Iligan Bay.

Sampling site	n	Fish length (cm)	Fish weight (g)
Iligan City	30	19.26 ± 1.38	112.50 ± 26.94
Oroquieta City	30	18.47 ± 3.23	100.13 ± 49.84
Kapatagan	30	20.32 ± 4.59	149.14 ± 144.53

Table 3. Results of ANOVA tests investigating differences in otolith size parameters and shape indices across the sampling areas: Iligan City, Kapatagan, and Oroquieta City.

Descriptors	*F*	df	*P*
Otolith length (OL)	24.94	2	2.74e-09
Otolith height (OH)	0.899	2	0.411
Otolith weight (OW)	4.4	2	0.015
Otolith area (OA)	24.99	2	2.66e-09
Otolith perimeter (OP)	18.43	2	2.12e-07
Rectangularity (RE)	2.838	2	0.064
Squareness (SQ)	6.119	2	0.003
Ellipticity (EL)	20.93	2	3.79e-08
Roundness (RO)	6.001	2	0.003
Aspect ratio (AR)	19.45	2	1.04e-07
Circularity (CI)	24.72	2	3.15e-09
Compactness (CO)	1.623	2	0.203
Form factor (FF)	2.238	2	0.093

significantly for ellipticity ($F = 20.93$, $P = 3.79\text{e-}08$), for aspect ratio ($F = 19.45$, $P = 1.04\text{e-}07$), and for circularity ($F = 24.72$, $P = 3.15\text{e-}09$). However, rectangularity ($F = 2.838$, $P = 0.064$), squareness ($F = 6.119$, $P = 0.003$), roundness ($F = 6.001$, $P = 0.003$), compactness ($F = 1.623$, $P = 0.203$), and form factor ($F = 2.238$, $P = 0.093$) did not show any significant difference.

Shape visualisation

Using the average harmonics to reconstruct the shape's outline (Fig. 3), the otolith shape of brown-stripe snapper in Iligan Bay were rectangular and elliptic and amongst the three clusters of locations there were considerable differences identified by discriminant analysis. The length direction of the otolith, particularly between the rostrum and the anti-rostrum, clearly showed the variation between stocks.

Variation in shape between sexes

Possible sex-specific variations in otolith morphology were tested on standardised amplitudes on otoliths with a total length of 3.0 to 5.0 mm using Wavelet and Fourier coefficients. Using Multivariate Analysis of Variance, it was observed that no significant ($F = 70.088$, Pillai T = 0.746, $P < 0.01$) variations in otolith shape between males and females were found for the three populations in Iligan Bay, Mindanao, Philippines.

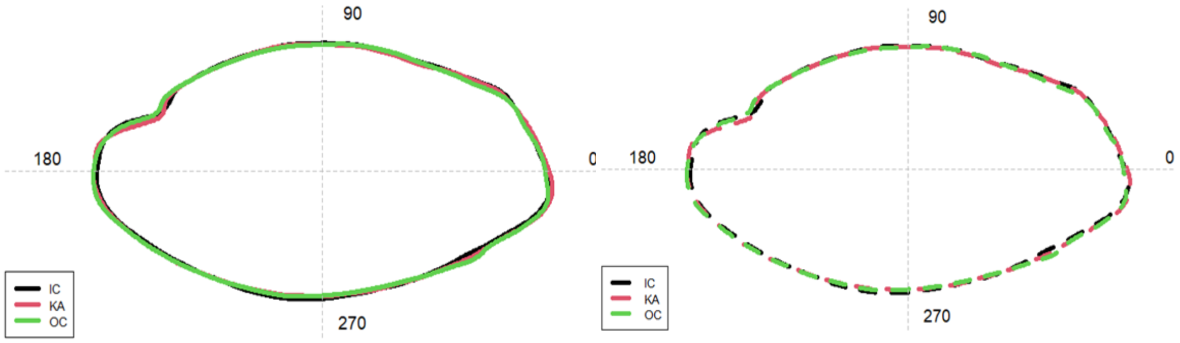

Fig. 3. Mean otolith shape from the spawning ground of Iligan Bay based on Wavelet reconstruction (right) and Fourier descriptors (left) of *Lutjanus* species from Iligan City (IC, n = 30), Kapatagan (KA, n = 30), and Oroquieta City (OC, n = 30).

Variation in shape and size

The canonical analysis of principal (CAP) coordinates provides a summary of the differences in otolith shape across the three populations that the ANOVA (Fig. 4, *P* = 0.001) determined to be significant. Along the first discriminating axis (CAP1), the Iligan City sample differs from Oroquieta City and Kapatagan, whereas the Oroquieta City sample mostly deviates from the Iligan City sample along the second axis (CAP2). In comparison, CAP1 explained 70.9 % and CAP2 contributed 29.1 % of the variation between populations.

The results of the ANOVA-like permutation test (Table 4) revealed significant (*F* = 2.82409, *P* = 0.001) differences in otolith shape across the sampling regions of Iligan City, Kapatagan, and Oroquieta City (Fig. 5) according to permutation testing.

The separation between sampling locations was further supported by the seasonal discrimination, which made use of all standardised otolith characteristics. Figure 6 shows the groupings of *L. vitta* otolith samples that were taken from the three sampling locations. The first two PCA eigenvalues (99.01 %) accounted for most of the total size and shape variance in the otolith across the three locations. Otolith morphometric descriptors such as otolith area (OA), otolith length (OL), otolith height (OH), and otolith perimeter (OP) contributed 73.9 % of variation explained by the first component (PC1), while derived otolith descriptors such as rectangularity (RE), compactness (CO), squareness (SQ), and form factor (FF) with 25.2 % more variation were added by the second component. Kapatagan samples were mainly found in the positive side of PCA1, However, samples from Oroquieta and Iligan City displayed overlapping patterns, suggesting some otolith similarities.

Discussion

Otoliths are utilised in stock identification due to their regular collection and use in traditional stock assessment, which makes their inclusion both reasonable and effective (Wang et al., 2011). Otolith morphology is primarily determined by genetics but is also influenced by environmental variables (Begg and Brown, 2000; Cardinale et al., 2004). In line with the expectation of an adaptive response to environmental conditions, the study's three populations show significant variations in otolith morphometric parameters. Thus, otolith morphometrics serve as a potential indicator for species or stock discrimination and may prove valuable for phenotypic rather than genetic stock differentiation (Pothin et al., 2006).

Individuals from the *L. vitta* species employed in this study revealed considerable variability in shape morphometric parameters between the three sampling locations, particularly between Iligan City, and the other two locations (Kapatagan City and Oroquieta City). Otolith shape index data can be used in subsequent investigations to distinguish stocks since they considerably differ between the three populations. It can be hypothesised that the inter-group differences in otolith form could be caused, in part, by environmental factors experienced by fish in the studied areas. Numerous additional studies have also revealed changes in sagitta shape between fish populations and stocks. For example, Campana and Casselman (1993) reported variances in sagitta shape between *Gadus morhua* stock and discovered that disparities in population growth rates could account for a significant portion of the form variation. Variations in environmental conditions can have a significant impact on otolith growth and consequently, on the morphology of otoliths (Campana and Neilson, 1985). It is well known that otolith shape can be influenced by sex, age, and year class and that growth rate appears to be more significant for regional differences than stock origin (Campana and Casselman, 1993; Hüssy et al., 2016; Moreira et. al., 2019; Vandenbussche et al., 2019).

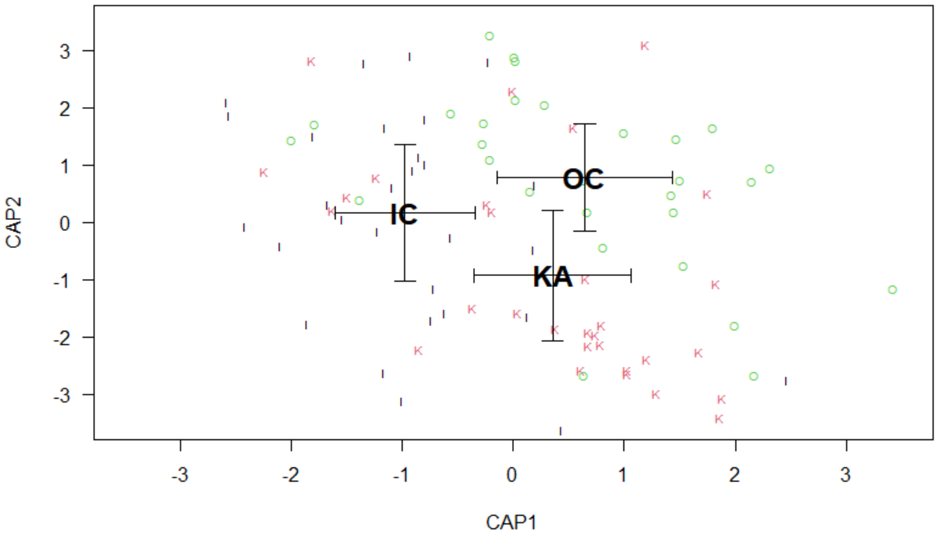

Fig. 4. Otolith shape of samples from three *Lutjanus vitta* populations in Iligan Bay (n = 90) using canonical analysis of principal coordinates with the Wavelet coefficients. IC = Iligan City, OC = Oroquiety City, KC = Kapatagan City.

Table 4. Summary of the ANOVA-like permutation test of otolith shape based on 1000 iterations of elliptical Fourier coefficients for *Lutjanus vitta* (brown-striped snapper) across three sampling locations.

Comparison	df	Variance	F	*P*
All locations	2	1.381	2.824	0.001

Fig. 5. *Lutjanus vitta* representative sagittal otolith pictures, right and non-sulcus side collected from Iligan City (left), Kapatagan (center), and Oroquieta City (right).

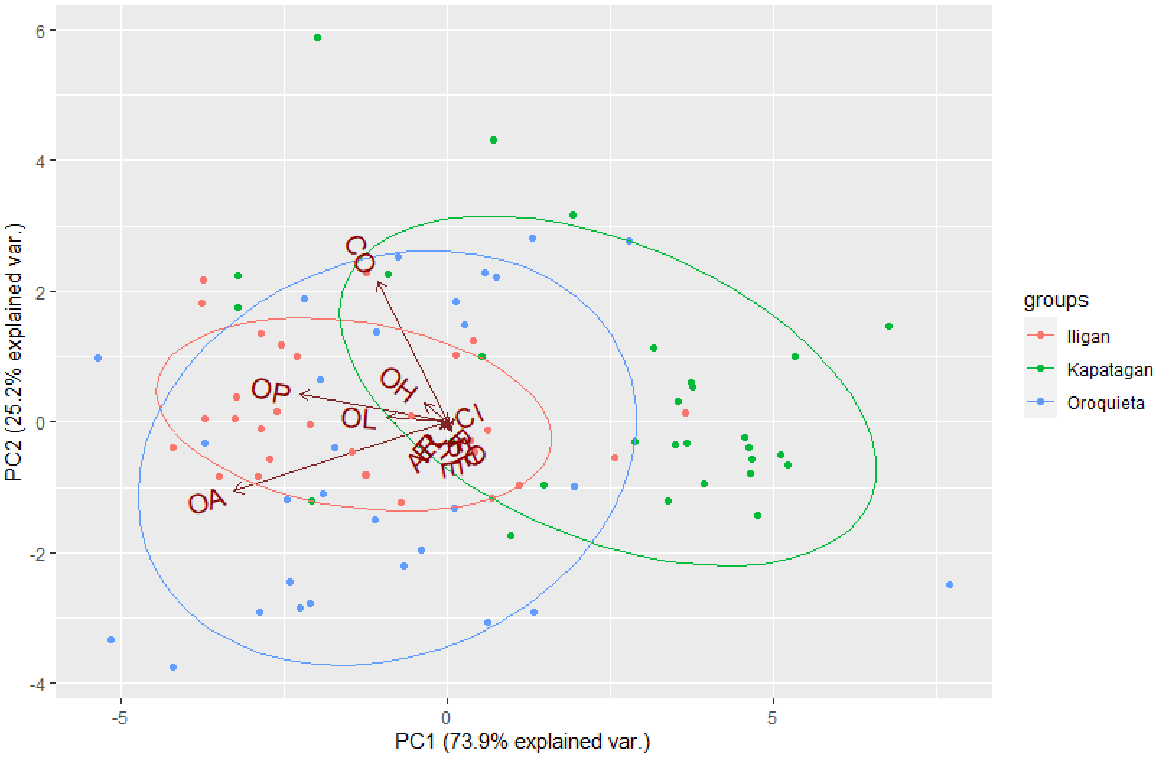

Fig. 6. Principal component analysis (PCA) plot for otolith size and shape indices of *Lutjanus vitta* from the three locations (n = 90). OL- otolith length, OA - otolith area, OP - otolith perimeter, OH - otolith height, OW - otolith weight, CO - compactness, CI - circularity, AR - aspect ratio, RE - rectangularity, RO - roundness, SQ - squareness, EL - ellipticity, FF - form factor.

For the three populations of *L. vitta* in Iligan Bay, this study demonstrated no significant variations in otolith form between males and females. Similarly, Moreira et al. (2019) found no relationship between fish otolith area and sex, indicating that otolith form in this species can be analysed without considering sex. According to other research (Cardinale et al., 2004), sex did not alter the otolith shape characteristics for a subset of species. There are numerous studies of species that have also failed to detect any sex differences in otolith shape e.g.; *Scomber scombrus* (Castonguay et al., 1991); lake trout *Salvelinus namaycush* (Simoneau et al., 2000); *G. morhua*, (Cardinale et al., 2004); megrim *Lepidorhombus whiffiagonis* (Mille et al., 2015), vocal toadfish *Porichthys notatus* (Bose et al., 2016), and blue-whiting *Micromesistius poutassou* (Mahé et al., 2016).

Shape indices exhibited significantly varying mean values across different locations, with groupings related to fish size. Mille et al. (2015) found a direct correlation between otolith morphology and the diet of marine fish, linking these morphological changes to growth rates. This suggests that variations in food availability and dietary composition, which can differ markedly between environments, directly influence otolith development. In addition to diet (Strelcheck et al., 2003; Mille et al., 2015), other environmental factors such as temperature, habitat depth, and salinity (Hoff and Fuiman, 1993; De Vries et al., 2002; Longmore et al., 2010; Sala et al., 2013; Tuset et al., 2015) have also been linked to changes in otolith shape. These influences are likely connected to their effects on growth and metabolic processes in fish (Bose et al., 2018). Vignon (2012) reported that changes in habitat conditions lead to significant alterations in otolith morphology, indicating that fish populations adapt their otolith shape in response to local environmental factors. Maciel et al. (2021) also noted that fish exposed to different environmental conditions are likely to exhibit differences in otolith shape, which can be statistically analysed. Moreover, studies by Cardinale et al. (2004) and Vignon and Morat (2010) have demonstrated that the physicochemical characteristics of the environment interact with genetic factors to determine the morphometric properties of otoliths. Understanding these influences is essential for accurate stock identification and effective fisheries management. This underscores the need for comprehensive studies that consider both genetic and environmental factors in otolith shape analysis.

Strong impacts from somatic length suggested that standardising otolith shape parameters for somatic size does not entirely eliminate the evolution of otolith shape with somatic length. Therefore, it is crucial to obtain sufficiently high sample sizes covering all accessible size classes to fully capture these changes and provide accurate predictions (Lin and Al-Abdulkader, 2019). Various fish habitats (Lord et al., 2012), hearing and sound production functions (Cruz and Lombarte, 2004), taxonomic and phylogenetic relationships (Lin and Chang, 2012), and ontogenetic effects at various developmental stages (Vignon, 2012; Vandenbussche et al., 2019) could all contribute to variations in otolith shape.

Identifying stocks and estimating the contribution of each stock in mixed fisheries is crucial for creating appropriate laws for effective fisheries management and for understanding the distributional range and migration behaviour of species. Neglecting stock identification in fisheries management can result in overfishing of non-targeted regional populations and the loss of genetic diversity, which may be essential for adaptability in a climate-changed ocean (Smith et al., 1991; Libungan et al., 2015). The investigation of the morphological and chemical properties of fish stock identification has been made easier with the use of otoliths (Ferguson et al., 2011). In relation to environmental conditions, otolith form is distinctly species-specific and frequently changes geographically within species (Cardinale et al., 2004; Stransky and MacLellan, 2005). Many stock identification studies of marine fish species, particularly small pelagic fish like anchovies (Mahmoud et al., 2014), blue jack mackerel (Moreira et al., 2019), and European sardine (Sharif et al., 2015), have successfully used otolith shape analysis to identify stocks. However, this study focuses solely on the morphometrics and shape analysis of *L. vitta* otoliths in Iligan Bay. It does not account for other variables like genetic variations or the influence of environmental factors that could alter otolith morphology.

Conclusion

The current findings underscore the value of otolith morphometric parameters and shape analysis for the stock identification of the brown-striped snapper (*Lutjanus vitta*). Although these phenotypic groups may be genetically similar, they are likely isolated and respond differently to environmental changes and human exploitation. The observed variations in otolith morphometrics and shape may be influenced, in part, by environmental factors. To better understand the stock structure of *L. vitta* in Iligan Bay, Mindanao, Philippines, further research is necessary to evaluate the relative contributions of genetic and environmental factors. As this study represents the first attempt to identify *L. vitta* stocks in Iligan Bay, future studies should incorporate otolith microchemistry, genetic analyses, spatial variations in age and growth, other life history-based approaches, and assessments of current exploitation levels. These efforts will provide a more comprehensive understanding of the stock structures of *L. vitta* within Philippine waters.

Acknowledgements

We are deeply grateful to the Accelerated Science and Technology Human Resources Development Program of the DOST Science Education Institute (DOST-SEI ASTHRDP) for the funding support. The authors are also grateful to the people who gave their assistance during sample collection and to the reviewers for their comments and suggestions, which significantly enhanced the manuscript.

Conflict of interest: The authors declare that they have no conflict of interest.

Author contributions: Maricel Tumampos Gumoloc: Conceptualisation, methodology, software, formal analysis, resources, investigation, data curation, writing – original draft, visualisation. Ivane R. Pedrosa-Gerasmio: Conceptualisation, software, validation, resources, writing - review and editing.

References

Allen, G. 1985. FAO species catalogue: Vol. 6. Snappers of the world. An annotated and illustrated catalogue of lutjanid species known to date. FAO fisheries synopsis. 208 pp.

Barnuevo, K.D.E., Morales, C.J.C., Calizo, J.K.S., Delloro, E.S., jr., Añasco, C.P., Babaran, R.P., Lumayno, S.D.P. 2023. Distinct stocks of the redtail scad *Decapterus kurroides* Bleeker, 1855 (Perciformes: Carangidae) from the Northern Sulu and Southern Sibuyan Seas, Philippines revealed from otolith morphometry and shape analysis. Fishes 8:12. https://doi.org/10.3390/fishes8010012

Begg, G.A, Brown, R.W. 2000 Stock identification of haddock *Melanogrammus aeglefinus* on Georges Bank based on otolith shape analysis. Transactions of the American Fisheries Society 129:935–145. https://doi.org/10.15488659(2000)129<0935:SIOHMA>2.3.CO;2

Begg, G.A., Cadrin, S.X. 2016. Stock identification. Fish reproductive biology: Implications for assessment and management. John Wiley & Sons Ltd., pp. 252–278. https://doi.org/10.1002/9781118752739.ch6

Begg, G.A., Waldman, J.R. 1999. An holistic approach to fish stock identification. Fisheries Research 43:35–44. https://doi.org/10.1016/S0165-7836(99)00065-X

Bose, A.P., McCallum, E.S., Raymond, K., Marentette, J.R., Balshine, S. 2018. Growth and otolith morphology vary with alternative reproductive tactics and contaminant exposure in the round goby *Neogobius melanostomus*. Journal of Fish Biology 93:674–684. https://doi.org/10.1111/jfb.13756

Bose, A.P.H., Adragna, J.B., Balshine, S. 2016. Otolith morphology varies between populations, sexes and male alternative reproductive tactics in a vocal toadfish *Porichthys notatus*. Journal of Fish Biology 90:311–325. https://doi.org/10.1111/jfb.13187

Cadrin S. X., Kerr, L. A., Mariani, S. 2014. Interdisciplinary evaluation of spatial population structure for definition of fishery management units. Stock identification methods: Applications in fishery science. Academic Press, USA, pp. 535–575. https://doi.org/10.1016/B978-0-12-397003-9.00022-9

Cadrin, S.X., Friedland, K.D. 1999. The utility of image processing techniques for morphometric analysis and stock identification. Fisheries Research 43:129–139. https://doi.org/10.1016/S0165-7836(99)00070-3

Cadrin, S.X., Goethel, D.R., Berger, A., Jardim, E. 2023. Best practices for defining spatial boundaries and spatial structure in stock assessment. Fisheries Research 262:106650. https://doi.org/10.1016/j.fishres.2023.106650

Campana, S.E., Casselman, J.M. 1993. Stock discrimination using otolith shape analysis. Canadian Journal of Fisheries and Aquatic Sciences 50:1062–1083. https://doi.org/10.1139/f93-123

Campana, S.E., Neilson, J.D. 1985. Microstructure of fish otoliths. Canadian Journal of Fisheries and Aquatic Sciences 42:1014–1032. https://doi.org/10.1139/F85-127

Cardinale, M., Doering-arjes, P., Kastowsky, M., Modegaard, H. 2004. Effects of sex, stock, and environment on the shape of Atlantic cod (*Gadus morhua*) otoliths. Canadian Journal of Fisheries and Aquatic Sciences 61:158–167. https://doi.org/10.1139/f03-151

Casselman, J.M., Collins, J.J., Grossman, E.J, Isheen, P.E, Spangler, G.R. 2011. Lake whitefish (*Coregonus clupeaformis*) stocks of the Ontario waters of Lake Huron. Canadian Journal of Fisheries and Aquatic Sciences 38:1772–1789. https://doi.org/10.1139/f81-225

Castonguay, M., Simard, P., Gagnon, P. 1991. Usefulness of fourier analysis of otolith shape for Atlantic mackerel (*Scomber scombrus*) stock discrimination. Canadian Journal of Fisheries and Aquatic Sciences 48:296–302. https://doi.org/10.1139/f91-041

Cruz, A., Lombarte, A. 2004. Otolith size and its relationship with colour patterns and sound production. Journal of Fish Biology 65:1512–1525. https://doi.org/10.1111/J.0022-1112.2004. 00558.X

Davis, T.L.O., West, G.J. 1992. Growth and mortality of *Lutjanus vittus* (Quoy and Gaimard) from the North West Shelf of Australia. Fishery Bulletin 90:395–404.

De Vries, D.A, Grimes, C.B, Prager, M.H. 2002. Using otolith shape analysis to distinguish eastern Gulf of Mexico and Atlantic Ocean stocks of king mackerel. Fisheries Research 57:51–62. https://doi.org/10.1016/S0165-7836(01)00332-0

Deepa, K., Kumar, K.A., Kottnis, O., Nikee, R., Bineesh, K., Hashim, M., Saravanane, N., Sudhakar, M. 2018. Population variations of Opal fish, *Bembrops caudimacula* Steindachner, 1876 from Arabian Sea and Andaman Sea: Evidence from otolith morphometry. Regional Studies in Marine Science 25:100466. https://doi.org/10.1016/j.rsma.2018.100466

Ferguson, G.J., Ward, T.M., Gillanders, B.M. 2011. Otolith shape and elemental composition: complementary tools for stock discrimination of mulloway (*Argyrosomus japonicus*) in southern Australia. Fisheries Research 110:75–83. https://doi.org/10.1016/j.fishres.2011.03.014

Freitas, T.M.S., Almeida, V.H.C., Valente, R.M., Montag, L.F.A. 2011. Feeding ecology of *Auchenipterichthys longimanus* (Siluriformes: Auchenipteridae) in a riparian flooded forest of Eastern Amazonia. Brazil. Neotropical Ichthyology 9:629–636. https://doi.org/10.1590/S1679-62252011005000032

Galli, O., Norbis, W. 2013. Morphometric and meristic spatial differences and mixed groups of the Whitemouth croaker (*Micropogonias furnieri* (Demearest, 1823)) during the spawning season: implications for management. Journal of Applied Ichthyology 29:782–788. https://doi:10.1111/jai.12114

Golden, C.D., Allison, E.H., Cheung, W.W.L., Dey, M.M., Halpern, B.S., Mccauley, D.J., Smith, M., Vaitla, B., Zeller, D., Myers, S.S. 2016. Fall in fish catch threatens human health. Nature 534:317–320. http://doi.org./10.1038/534317a

Halim, L.J., Rahim, I., Mahboob, S., Al-ghanim, K.A., Naim, A.D. 2021. Phylogenetic relationships of the commercial red snapper (*Lutjanidae* sp.) from three marine regions. Journal of King Saud University–Science 34:101756. https://doi.org/10.1016/j.jksus.2021.101756

Hilborn, R., Walters, C.J. 2004. Quantitative fisheries stock assessment: Choice, dynamics and uncertainty. Reviews in Fish Biology and Fisheries 2:177–178. https://doi:10.1007/BF00042883

Hoff, G.R., Fuiman, L.A. 1993. Morphometry and composition of red drum otoliths: Changes associated with temperature, somatic growth rate, and age. Comparative Biochemistry and Physiology Part A: Physiology 106:209–219. https://doi.org/10.1016/0300-9629(93)90502-U

Hüssy, K., Mosegaard, H., Albertsen, C.M., Nielsen, E.E., Hemmer-Hansen, J., Eero, M. 2016. Evaluation of otolith shape as a tool for stock discrimination in marine fishes using Baltic Sea cod as a case study. Fisheries Research 174:210–218. https://doi.org/10.1016/j.fishres.2015.10.010

Kallio-Nyberg, I., Saura, A., Ahlfors, P. 2002. Sea migration pattern of two sea trout (*Salmo trutta*) stocks released into the Gulf of Finland. Annales Zoologici Fennici 39:221–235. https://www.jstor.org/stable/23736710

Kerr, L.A., Hintzen, N.T., Cadrin, S.X., Clausen, L., Worsøe, D., Collas, M., Goethel, D.R., Hatfield, E.M.C., Kritzer, J.P., Nash, R.D.M. 2017. Lessons learned from practical approaches to reconcile mismatches between biological population structure and stock units of marine fish. ICES Journal of Marine Science 74:1708–1722. https://doi:10.1093/icesjms/fsw188

Koolkalya S., Trueman, C., Sawusdee, A., Jutagate, T. 2020. Preliminary determination of the stocks of short mackerel *Rastrelliger brachysoma* in the Gulf of Thailand. Asian Fisheries Science 33:249–257. https://doi.org/10.33997/j.afs.2020.33.3.006

Larkin, P.A. 1972. The stock concept and management of Pacific salmon. University of British Columbia, HR MacMillan Lectures in Fisheries, Vancouver, pp. 11–15.

Lecomte-Finiger, R. 1992. The crystalline ultrastructure of otolith of the eel (*A. anguilla* L. 1758). Journal of Fish Biology 40:181–190. https://doi.org/10.1111/j.1095-8649.1992.tb02565.x

Lestrel, P.E. 1997. Fourier Descriptors and their applications in biology. Cambridge University Press, Cambridge. 466 pp. https://doi.org/10.1017/CBO9780511529870

Libungan, L.A., Óskarsson, G.J., Slotte, A., Jacobsen, J.A., Pal, S. 2015. Otolith shape: a population marker for Atlantic herring *Clupea harengus*. Journal of Fish Biology 86:1377–1395. https://doi.org/10.1111/jfb.12647

Libungan, L.A., Pálsson, S. 2015. ShapeR: an R package to study otolith shape variation among fish populations. PLoS One 10:e0121102. https://doi.org/10.1371/journal.pone.0121102

Lin, C. H., Chang, C. W. 2012. 'Otolith Atlas of Taiwan Fishes.'. National Museum of Marine Biology and Aquarium. pp. 415. https://doi.org/10.1111/jfb.12313

Lin, Y.J, Al-Abdulkader, K. 2019. Identification of fish families and species from the western Arabian Gulf by otolith shape analysis and factors affecting the identification process. Marine and Freshwater Research 70:1818–1827. https://doi.org/10.1071/MF18282

Longmore, C., Fogarty, K., Neat, F. 2010. A comparison of otolith microchemistry and otolith shape analysis for the study of spatial variation in a deep-sea teleost, *Coryphaenoides rupestris*. Environmental Biology of Fishes 89:591–605. https://doi.org/10.1007/s10641-010-9674-1

Lord, C., Morat, F., Lecomte-Finiger, R., Keith, P. 2012. Otolith shape analysis for three *Sicyopterus* (Teleostei: Gobioidei: Sicydiinae) species from New Caledonia and Vanuatu. Environmental Biology of Fishes 93:209–222. https://doi.org/10.1007/S10641-011-9907Y

Maciel, T.R., Vianna, M., de Carvalho, B.M., Miller, N., Avigliano, E. 2021. Integrated use of otolith shape and microchemistry to assess *Genidens barbus* fish stock structure. Estuarine, Coastal and Shelf Science 261:107560. https://doi.org/10.1016/j.ecss.2021.107560

Mahe, K., Oudard, C., Mille, T., Keating, J., Gonçalves, P., Worsøe Clausen, L., Petursdottir, G., Rasmussen, H., Meland, E., Mullins, E., Pinnegar, J.K., Hoines, Å., Trenkel, V.M. 2016. Identifying blue whiting (*Micromesistius poutassou*) stock structure in the Northeast Atlantic by otolith shape analysis. Canadian Journal of Fisheries and Aquatic Sciences 73:1363–1371. https://doi.org/10.1139/cjfas-2015-0332

Mahmoud, B., Sherif, J., Azzedine, H., Khalef, R., Rachid, A. 2014. Population structure of the European anchovy, *Engraulis encrasicolus*, in the SW Mediterranean Sea, and the Atlantic Ocean: evidence from otolith shape analysis. ICES Journal of Marine Science 71:2429–2435. https://doi.org/10.1093/icesjms/fsu097

Marini, M., Pedrosa-Gerasmio, I.R., Santos, M.D., Shibuno, T., Daryani, A., Romana-Eguia, M.R.R., Wibowo, A. 2021. Genetic diversity, population structure and demographic history of the tropical eel *Anguilla bicolor* pacifica in Southeast Asia using mitochondrial DNA control region sequences. Global Ecology and Conservation 26:e01493. https://doi.org/10.1016/j.gecco.2021.e01493

Mérigot, B., Letourneur, Y., Lecomte-Finiger, R. 2007. Characterization of local populations of the common sole *Solea solea* (Pisces, Soleidae) in the NW Mediterranean through otolith morphometrics and shape analysis. Marine Biology 151:997-1008. https://doi.org/10.1007/s00227-006-0549-0

Mille, T., Mahe, K., Villanueva, M.C., De Pontual, H., Ernande, B. 2015. Sagittal otolith morphogenesis asymmetry in marine fishes. Journal of Fish Biology 87:646–663. https://doi.org/10.1111/jfb.12746

Moreira, C., Froufe, E., Vaz-Pires, P., Correia, A.T. 2019. Otolith shape analysis as a tool to infer the population structure of the blue jack mackerel, *Trachurus picturatus*, in the NE Atlantic. Fisheries Research 209:40–48. https://doi.org/10.1016/J.FISHRES.2018.09.010

Morris, A.V., Callum, M.R., Julie, P.H. 2000. The threatened status of groupers (Epinephelinae). Biodiversity Conservation 9:919–942. https://doi.org/10.1023/A:1008996002822

Oksanen, J., Gavin, S., Guillaume Blanchet, F., Kindt, R., Legendre, P., Weedon, J. 2022. vegan: Community Ecology Package. R package version 2.6-4.

Palla, H.P., Gonzales, B.J., Sotto, F.B., Ilano, A.S., Tachihara, K. 2016. Age, growth and mortality of brown stripe snapper *Lutjanus vitta* (Quoy and Gaimard 1824) from West Sulu Sea, Philippines. Asian Fisheries Science 29:28–42. https://doi.org/10.33997/j.afs.2016.29.1.003

Pedrosa-Gerasmio, I.R., Agmata, A.B., Santos, M.D. 2015. Genetic diversity, population genetic structure, and demographic history of *Auxis thazard* (Perciformes), *Selar crumenophthalmus* (Perciformes), *Rastrelliger kanagurta* (Perciformes) and *Sardinella lemuru* (Clupeiformes) in Sulu-Celebes Sea inferred by mitochondrial DNA sequences. Fisheries Research 162:64–74. https://doi.org/10.1016/j.fishres.2014.10.006

Popper, A.N., Coombs, S. 1982. The morphology and evolution of the ear actinopterygian fishes. American Zoologist 22:311–328. https://doi.org/10.1093/icb/22.2.311

Pothin, K., Gonzales-Salas, C., Chabanet, P, Lecomte-Finiger, R. 2006. Distinction between *Mulloidichthys flavolineatus* juveniles from reunion Island and Mauritius Island (southwest Indian Ocean) based on otolith morphometrics. Journal of Fish Biology 69:38–53. https://doi.org/10.1111/J.1095-8649.2006.01047.X

Sala, I., Caldeira, R.M.A., Estrada-Allis, S.N., Froufe, E., Couvelard, X. 2013. Lagrangian transport pathways in the northeast Atlantic and their environmental impact. Limnology and Oceanography: Fluids & Environments 3:40–60. https://doi.org/10.1215/21573689-2152611

Sharif, J., Mahmoud, B., Gaby, K., David, D., Khalef, R., Rachid, A. 2015. What can otolith shape analysis tell us about population structure of the European sardine, *Sardina pilchardus* from Atlantic and Mediterranean waters. Journal of Sea Research 96:11–17. https://doi.org/10.1016/J.SEARES.2014.11.002

Simoneau, M., Casselman, J.M., Fortin, R. 2000. Determining the effect of negative allometry (length/height relationship) on variation in otolith shape in lake trout (*Salvelinus namaycush*), using Fourier-series analysis. Canadian Journal of Zoology 78:1597–1603. https://doi.org/10.1139/z00-093

Smith, P.J., Francis, R.I.C.C., Mcveagh, M. 1991. Loss of genetic diversity due to fishing pressure. Fisheries Research 10:309–316. https://doi.org/10.1016/0165-7836(91)90082-Q

Soeth, M., Spach, H.L., Daros, F.A., Adelir-Alves, J., de Almeida, A.C.O., Correia, A.T. 2019. Stock structure of Atlantic spadefish *Chaetodipterus faber* from Southwest Atlantic Ocean inferred from otolith elemental and shape signatures. Fisheries Research 211:81–90. https://doi.org/10.1016/j.fishres.2018.11.003

Stransky, C., MacLellan, S.E. 2005. Species separation and zoo geography of redfish and rockfish (genus *Sebastes*) by otolith shape analysis. Canadian Journal of Fisheries and Aquatic Sciences 62:2265–2276. https://doi.org/10.1139/f05-143

Strelcheck, A.J., Fitzhugh, G.R., Coleman, F.C., Koenig, C.C. 2003. Otolith-fish size relationship in juvenile gag (*Mycteroperca microlepis*) of the eastern Gulf of Mexico: a comparison of growth rates between laboratory and field populations. Fisheries Research 60:255–265. https://doi.org/10.1016/S0165-7836(02)00171-6

Teh, L.S.L., Witter, A., Cheung, W.W.L., Sumaila, U.R., Yin, X. 2017. What is at stake? Status and threats to South China Sea marine fisheries. Ambio 46:57–72. https://doi.org/10.1007/s13280-016-0819-0

Tuset, V.M., Imondi, R., Aguado, G., Otero-Ferrer, J.L., Santschi, L., Lombarte, A., Love, M. 2015. Otolith patterns of rockfishes from the

Northeastern Pacific Journal of Morphology 276:458–469. https://doi.org/10.1002/jmor.20353

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). Journal of Applied Ichthyology 19:88–93. https://doi.org/10.1046/j.14390426.2003.00344.x

Vandenbussche, P.S.P., Spennato, G., Pierson, P.M. 2019. Juvenile *Oblada melanura* (L. 1758) otolith shape variation as an early environmental indicator. Ecological Indicators 104:289–295. https://doi.org/10.1016/j.ecolind.2019.05.011

Vignon, M. 2012. Ontogenetic trajectories of otolith shape during shift in habitat use: Interaction between otolith growth and environment. Journal of Experimental Marine Biology and Ecology 420–421, 26–32. https://doi.org/10.1016/J.JEMBE.2012.03.021

Vignon, M., Morat, F. 2010. Environmental and genetic determinant of otolith shape revealed by a non-indigenous tropical fish. Marine Ecology Progress Series 411:231–241. https://doi.org/10.3354/meps08651

Waldman, J.R. 2005. Definition of stocks: an evolving concept. Academic Press. pp. 7–16. https://doi.org/10.1016/B978-012154351-8/50003-4

Wang, Y., Ye, Z., Liu, Q. 2011. Stock discrimination of spottedtail goby (*Synechogobius ommaturus*) in the Yellow Sea by analysis of otolith shape. Chinese Journal of Oceanology and Limnology 29:192–198. https://doi.org/10.1007/s00343-011-9087-9

Wickham, H. 2016. ggplot2: Elegant graphics for data analysis. Springer-Verlag, New York. https://ggplot2.tidyverse.org

Wujdi, A., Kim, H.J., Oh, C.W. 2022. Population structure of Indian mackerel (*Rastrelliger kanagurta*) in Java and Bali Island, Indonesia inferred from otolith shape. Sains Malaysiana 51:39–50. http://doi.org/10.17576/jsm-2022-5101-04