Acta Ichthyologica et Piscatoria 51(2), 2021, 193–197 | DOI 10.3897/aiep.51.64220

PENSOFT.

ACTA ICHTHYOLOGICA ET PISCATORIA

Investigating otolith mass asymmetry in six benthic and pelagic fish species (Actinopterygii) from the Gulf of Tunis

Nawzet BOURIGA[1,2], Marwa MEJRI[2], Monia DEKHIL[2], Safa BEJAOUI[2], Jean-Pierre QUIGNARD[3], Monia TRABELSI[2]

1 *Institut Supérieur de Pêche et d'Aquaculture de Bizerte, Tunis, Tunisia*

2 *Laboratory of Ecology, Biology and Physiology of Aquatic Organisms, Faculty of Sciences of Tunis, University of Tunis El Manar, Tunis, Tunisia*

3 *Laboratoire d'Ichtyologie, Université Montpellier II, Montpellier, France*

http://zoobank.org/5FF09E48-D532-4958-B268-1FAEDD581DC4

Corresponding author: N. Bouriga (hanounawsset@live.fr)

Academic editor: Sanja Matić-Skoko ♦ **Received** 7 October 2020 ♦ **Accepted** 1 February 2021 ♦ **Published** 12 July 2021

Citation: Bouriga N, Mejri M, Dekhil M, Bejaoui S, Quignard J-P, Trabelsi M (2021) Investigating otolith mass asymmetry in six benthic and pelagic fish species (Actinopterygii) from the Gulf of Tunis. Acta Ichthyologica et Piscatoria 51(2): 193–197. https://doi.org/10.3897/aiep.51.64220

Abstract

Otolith mass asymmetry can significantly affect the vestibular system functionalities; usually, the *X* values of mass asymmetry vary between −0.2 and + 0.2 (−0.2 < X < + 0.2). These values can change during a fish life and therefore they are not related to the fish total length. We collected a total of 404 fish specimens from the Gulf of Tunis, including three pelagic species: *Sardina pilchardus* (Walbaum, 1792) (74 otolith pairs), *Trachurus mediterraneus* (Steindachner, 1868) (66 otolith pairs), and *Chelon auratus* (Risso, 1810) (60 otolith pairs) and three benthic species: *Gobius niger* Linnaeus, 1758 (77 otolith pairs), *Mullus barbatus* Linnaeus, 1758 (60 otolith pairs), and *Trachinus draco* Linnaeus, 1758 (67 otolith pairs). The relation between the total length and the otolith mass asymmetry was first calculated and compared, and then was evaluated. The comparison of the otolith mass asymmetry between benthic and pelagic species showed a significant difference ($P < 0.05$), where the absolute mean value of *X* does not exceed the critical value (0.2) for all the studied species. No relation has been found between the magnitude of the otolith mass asymmetry and the length in both benthic and pelagic specimens. Environmental factors have an indirect effect on somatic growth and otolith accretion. The significant difference found in this study can be due to the difference between the benthic and pelagic environments.

Keywords

benthic, Gulf of Tunis, Otolith, otolith mass asymmetry, pelagic

Introduction

Otoliths are calcified structures found in the inner ears of teleosts, in the vestibular system. Every fish has three otolith pairs mostly composed of calcium and carbonate layers precipitated in an organic matrix (Carlström 1963; Panfili et al. 2002; Pracheil et al. 2019). These paired structures are involved in acoustic and balance systems of teleosts (Paxton 2000). These calcified structures are metabolically inert, they grow by accretion and they cannot be used by the organism as a source of calcium (Campana and Neilson 1985). Thanks to these properties, otolith pairs and especially the sagittas are used to rebuild fish life cycle and their interactions with the environment (Vignon and Morat 2010).

Several authors have used otoliths simply to study fish age status, sexual dimorphism, and migration (Walther and Limburg 2012; Fatnassi et al. 2017). Also, otolith weight has been used as a surrogate method to estimate the age of fish (Francis and Campana 2004; Nazir and Khan 2019).

For many years, otolith shape has been routinely used to differentiate between fish stocks (Keating et al. 2014; Bailey et al. 2015; Ider et al. 2017; Mejri et al. 2018b; Nazir and Khan 2019; Avigliano et al. 2020). Otolith microchemistry has also been used to explore the environmental and food effects on fish metabolism (Mille et al. 2015; Perry et al. 2015). Furthermore, many presently cited studies indicate that the otolith mass asymmetry is also an important parameter because otoliths play a major role in acoustic functionalities.

The inconsistency between the right and left otolith's movements inside the inner ear can cause balance problems and sound perception difficulties (Lychakov and Rebane 2005; Lychakov et al. 2006).

Several studies on otolith mass asymmetry have shown that the majority of the symmetric fish species have X values within the range of $-0.2 < X < +0.2$ (Lychakov and Rebane 2004, 2005; Jawad 2013). Thus, in theory, only an absolute value of X that exceeds 0.2 can alter the acoustic functionality of a fish (Lychakov et al. 2006). Moreover, the relation between otolith mass asymmetry and fish total length has been explored (Mejri et al. 2018a). Lychakov and Rebane (2004) have shown a relation between saccular otolith mass asymmetry and fish length exists only in littoral and bottom populations and not in the pelagic species. Also, some studies have shown that there is no relation between otolith mass asymmetry and fish total length (Jawad 2013; Yedier et al. 2018). The presently reported study aimed to compare the otolith mass asymmetry between pelagic and benthic species and to assess the relation between mass asymmetry and the fish total length. The following species were studied: three pelagic species: *Sardina pilchardus* (Walbaum, 1792), *Trachurus mediterraneus* (Steindachner, 1868), and *Chelon auratus* (Risso, 1810) and three benthic species: *Gobius niger* Linnaeus, 1758, *Mullus barbatus* Linnaeus, 1758, and *Trachinus draco* Linnaeus, 1758.

Materials and methods

Sample collection

A total of 204 benthic fish species and 200 pelagic fish samples were collected from the Gulf of Tunis, in the north of Tunisia (36°49′09″N, 10°18′22″E) from March to May 2017 (Fig. 1). In this study, we only used adult fishes to compare the otolith mass asymmetry between benthic and pelagic species without considering gender. Standard and total length (L_s, L_t) were measured for each specimen to the nearest mm. The mean total length of the benthic species ranged from 150.8 ± 14.34 for *Mullus barbatus* to 238.322 ± 16.59 for *Gobius niger* and from 150.375 ± 9.69 to 235.14 ± 11.62 for pelagic species (*Sardina pilchardus* and *Chelon auratus*, respectively) (Table 1).

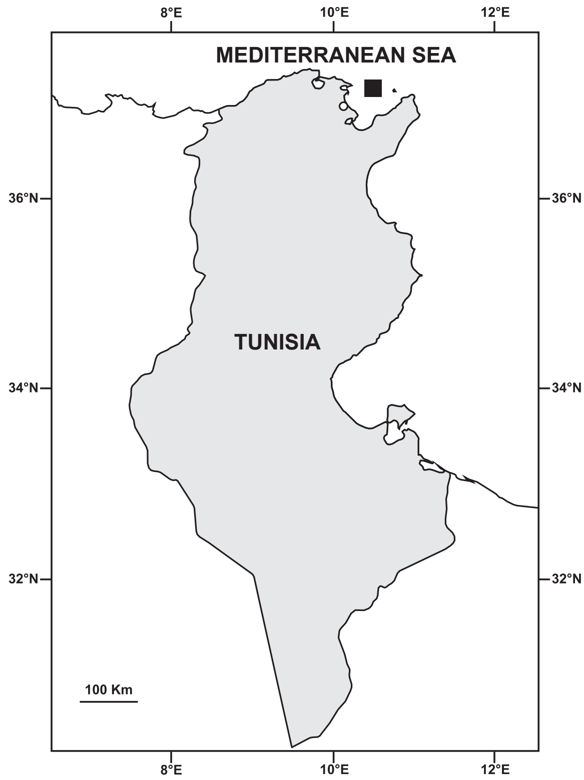

Figure 1. Sampling sites of the studied species in the Gulf of Tunis.

Otolith extraction

Sagittal otoliths pairs were manually removed by the dissection of the auditory capsules, washed with distilled water, and air-dried at room temperature. The weight of the right and left paired otoliths was also determined using a precision electronic balance (Mettler Toledo AL204) to an accuracy of 0.1 mg (Table 1).

Table 1. Descriptive statistics of the total length of benthic and pelagic fish species from the Gulf of Tunis.

Species	Domain	*n*	Mean ± SE [mm]
Mullus barbatus	Benthic	60	150.80 ± 14.34
Trachinus draco	Benthic	67	202.57 ± 17.99
Gobius niger	Benthic	77	238.32 ± 16.59
Chelon auratus	Pelagic	60	235.14 ± 11.62
Sardina pilchardus	Pelagic	74	150.38 ± 9.69
Trachurus mediterraneus	Pelagic	66	187.15 ± 17.32

Data calculation

The otolith mass asymmetry (X) was computed using the following formula:

$$X = (M_R - M_L)\, M_M^{-1}$$

where M_R and M_L are the otolith masses of the right and left paired otoliths, and M_M is the mean mass of the right and left paired otoliths.

Theoretically, X values vary from −2 to +2. These limit values indicate maximal asymmetry while the '0' value refers to the absence of asymmetry between right and left otoliths of the same fish. A negative value of X means that the left otolith is heavier than the right one ($M_L > M_R$), whereas a positive value of X means the opposite.

The relation between absolute otolith mass asymmetry ($|X|$) and the total fish length was calculated using the following formula:

$$|X| = a \cdot L_t + b$$

where a is the coefficient characterizing the growth rate of the otolith and b is a constant for a given species.

Statistical analyses

The comparison of the otolith mass asymmetry between benthic and pelagic specimens was performed using Student's t-test. To assess the existence of a relation between the total length and the absolute otolith mass asymmetry, a regression analysis was used to calculate correlation coefficients and regression equations for each species. XLSTAT (2007) software was used for all statistical analyses.

Results

It is known that the absolute symmetry between left and right otolith ($X = 0$) is rare, usually, X values fluctuate around 0. In this study, the percentage of asymmetry exceeded 50% in all the studied fish species. *Trachinus draco*, which is a benthic species, has shown an asymmetry rate equal to 100%. This means that all the otolith pairs of the 67 used fish samples are asymmetric in terms of weight, unlike the results observed in *Sardina pilchardus* population which have shown the lowest asymmetry rate with only 53%.

The otolith mass asymmetry was within the range of $-0.496 \leq X \leq 0.3379$ for all the studied species. It varied between −0.0513 and 0.1531 for benthic species and between −0.496 and 0.3379 for pelagic species (Table 2). The mean values of $|X|$ were calculated for all investigated species. The results have shown a mean value of $|X|$ equal to 0.0251 ± 0.0021 for benthic species and 0.0383 ± 0.0046 for pelagic species. Moreover, a significant difference was found between benthic and pelagic fish species ($P < 0.05$) (Table 3).

Table 2. Descriptive statistics results of otolith mass asymmetry (X) and absolute otolith mass asymmetry ($|X|$) for benthic (*Mullus barbatus*, *Trachinus draco* and *Gobius niger*) and pelagic fish species (*Chelon auratus*, *Sardina pilchardus* and *Trachurus mediterraneus*) from the Gulf of Tunis.

Species	$\|X\|$ Mean ± SD	X Minimum	X Maximum	Asymmetry rate [%]
Mullus barbatus	0.0304 ± 0.0424	–0.0258	0.0952	91
Trachinus draco	0.0399 ± 0.0648	–0.0347	0.0771	100
Gobius niger	0.0264 ± 0.0265	–0.0513	0.1531	54
Chelon auratus	0.0264 ± 0.0632	–0.4960	0.0508	53
Sardina pilchardus	0.0493 ± 0.0683	–0.3636	0.1538	93
Trachurus mediterraneus	0.0186 ± 0.0151	–0.3665	0.3379	89

Table 3. Descriptive statistics of absolute otolith mass asymmetry ($|X|$) for benthic (*Mullus barbatus*, *Trachinus draco*, *Gobius niger*) and pelagic fish species (*Chelon auratus*, *Sardina pilchardus*, *Trachurus mediterraneus*) from the Gulf of Tunis.

Parameter	Species	
	Benthic	Pelagic
n	204	200
Mean	0.0251	0.0383
Minimum	0.0000	0.0000
Maximum	0.2588	0.4962
SD	0.0021	0.0046
t-values	1.9660	
P-values	0.0102	

The correlation coefficients R^2 and regression equations were calculated for all of the six studied species each apart. The results reject the hypothesis since no significant relation between absolute otolith mass asymmetry and the total fish length was found ($0.0008 \leq R^2 \leq 0.0356$) (Table 4).

Table 4. Correlation coefficient and P-value of benthic (*Mullus barbatus*, *Trachinus draco*, *Gobius niger*) and pelagic fish species (*Chelon auratus*, *Sardina pilchardus*, *Trachurus mediterraneus*) from the G ulf of Tunis.

Species	R^2	P-value
Gobius niger	0.0008	0.993
Chelon auratus	0.0302	0.184
Mullus barbatus	0.0014	0.777
Sardina pilchardus	0.0040	0.958
Trachurus mediterraneus	0.0048	0.579
Trachinus draco	0.0356	0.138

Discussion

In the presently reported study, all benthic and pelagic species had a mean value of otolith mass asymmetry varying from −0.2 to 0.2 similar to previous studies (Lychakov et al. 2008; Jawad 2013; Jawad and Sadighzadeh 2013). Only pelagic species showed X values exceeding the critical limits, in particular, *T. mediterraneus* with an otolith mass asymmetry $-0.3665 < X < 0.3379$. This is probably related to the physiological state of this species, its habitat, and environmental factors (abiotic and biotic) as previously reported by Grønkjær (2016) and (Izzo et al. 2018) since the fish studied survived at different latitudes and longitudes. In the same context, the variations of environmental factors as well as anthropogenic ones have remarkable effects on the development of otoliths (Munday et al. 2011). In fact, in their study on the physicochemical parameters of the Gulf of Tunis, Ben Lamine et al. (2011) showed that this area suffers from several problems, such as metal pollution and urban discharges. Likewise, other studies have shown that pollution can affect the growth of otoliths (Elsdon and Gillanders 2002; Munday et al. 2011; Perry et al. 2015). It has been shown that metals impact otolith growth when fish inhabiting affected areas are likely to accumulate metals, including anthropogenic metals from surrounding environments, and transfer / transport them to the upper links in the food chain (Wang 2002). This is reflected in the continued incorporation of the metals into the crystal matrix of the otolith since they are metabolically inert and their increments can undergo resorption. Our results are consistent with other studies conducted on the effect of metal accumulation on otolith growth (Vrdoljak et al. 2020).

Morphological variability of sagittae is impacted by a dual regulation: genetic and environmental factors (L'Abée-Lund 1988; Lombarte et al. 2010; Vignon and Morat 2010; Annabi et al. 2013). Also, the increase or decrease in otolith mass asymmetry can negatively affect other factors that are necessary for the life of the fish, especially the sense of hearing and balance. The otolith mass asymmetry has been used as a bioindicator to test the condition in different aquatic habitats (Grønkjær 2016) and to test different environmental effects on fish populations. In our study, we have shown that the relative size of sagittae is larger in benthic species than in pelagic ones, which could be related to the different ecological niches of the studied fish (Lombarte and Cruz 2007).

The relation between the otolith mass asymmetry and the total length has been investigated in several studies (Mille et al. 2015; Yedier et al. 2018). We used the absolute value of X to prove whether there is any relation between the absolute otolith mass asymmetry and fish total length. According to the presently reported results, the absolute value of the otolith mass asymmetry does not depend on the total length in any studied species. Our results agree with the previous findings which investigated roundfish and flatfish (Jawad 2013; Yedier et al. 2018).

The otolith mass and shape asymmetry was explored in many studies all over the world. In Tunisia, otolith research is limited to fish stock identification and the assessment of sexual dimorphism. We need more research studying the impact of the environment on otolith mass asymmetry and on fish behavior. Therefore, it is important to use a large number of specimens and a wide range of body sizes in future studies to fully understand the relation between the asymmetry in otolith mass and the fish length.

Conclusion

Environmental factors have an indirect effect on somatic growth and otolith accretion. The significant difference found in this study can be related to the difference between the benthic and pelagic environments. Food and genetic variability can also be used to explain the presently reported results.

Acknowledgments

This work was supported by the laboratory of Ecology, Biology, and Physiology of Aquatic Organisms, Faculty of Sciences, University of Tunis El Manar. We thank Miss Hajer ZARROUK for her English Language correction. In the memory of my brother Hanou who passed away on 6 January 2021.

References

Annabi A, Said K, Reichenbacher B (2013) Interpopulation differences in otolith morphology are genetically encoded in the killifish *Aphanius fasciatus* (Cyprinodontiformes). Scientia Marina 77(2): 269–279. https://doi.org/10.3989/scimar.03763.02A

Avigliano E, Martinez G, Stoessel L, Mendez A, Bordel N, Pisonero J, Volpedo A (2020) Otoliths as indicators for fish behaviour and procurement strategies of hunter-gatherers in North Patagonia. Heliyon 6(3): e03438. https://doi.org/10.1016/j.heliyon.2020.e03438

Bailey DS, Fairchild EA, Kalnejais LH (2015) Microchemical signatures in juvenile winter flounder otoliths provide identification of natal nurseries. Transactions of the American Fisheries Society 144(1): 173–183. https://doi.org/10.1080/00028487.2014.982259

Ben Lamine Y, Yahia-Kefi OD, Daly Yahia NM (2011) Caracterisation Physico-Chimique de la partie sud oust de la baie de Tunis sous l'influence des apports de l'oued Meliane. Bulletin de l'Institut National des Sciences et Technologies de la Mer de Salammbô 38: 0330–0080.

Campana SE, Neilson JD (1985) Microstructure of fish otoliths. Canadian Journal of Fisheries and Aquatic Sciences 42(5): 1014–1032. https://doi.org/10.1139/f85-127

Carlström D (1963) A crystallographic study of vertebrate otoliths. Biological Bulletin 125(3): 441–463. https://doi.org/10.2307/1539358

Elsdon TS, Gillanders BM (2002) Interactive effects of temperature and salinity on otolith chemistry: Challenges for determining environmental histories of fish. Canadian Journal of Fisheries and Aquatic Sciences 59(11): 1796–1808. https://doi.org/10.1139/f02-154

Fatnassi M, Khedher M, Trojette M, Mahouachi NEH, Chalh A, Quignard JP, Trabelsi M (2017) Biometric data and contour shape to assess sexual dimorphism and symmetry of the otolith pairs of *Trachinus draco* from North Tunisia. Cahiers de Biologie Marine 58(3): 261–268. https://doi.org/10.21411/CBM.A.8EB74E07

Francis RIC, Campana SE (2004) Inferring age from otolith measurements: a review and a new approach. Canadian Journal of Fisheries and Aquatic Sciences 61: 1269–1284. https://doi.org/10.1139/f04-063

Grønkjær P (2016) Otoliths as individual indicators: A reappraisal of the link between fish physiology and otolith characteristics. Marine and Freshwater Research 67(7): 881–888. https://doi.org/10.1071/MF15155

Ider D, Ramdane Z, Kelig M, Jean-Louis D, Mahmoud B, Rachid A (2017) Use of otolith-shape analysis for stock discrimination of *Boops boops* along the Algerian coast (southwestern Mediterranean Sea). African Journal of Marine Science 39(3): 251–258. https://doi.org/10.2989/1814232X.2017.1363817

Izzo C, Reis-Santos P, Gillanders BM (2018) Otolith chemistry does not just reflect environmental conditions: A meta-analytic evaluation. Fish and Fisheries 19(3): 441–454. https://doi.org/10.1111/faf.12264

Jawad LA (2013) Otolith mass asymmetry in *Carangoides caerulepinnatus* (Rüppell, 1830) (family: Carangidae) collected from the sea of Oman. Ribarstvo 71(1): 37–41. https://doi.org/10.14798/71.1.622

Jawad LA, Sadighzadeh Z (2013) Otolith mass asymmetry in the mugilid fish, *Liza klunzingeri* (Day, 1888) collected from Persian Gulf near Bandar Abbas. Anales de Biología 35: 105–107. https://doi.org/10.6018/analesbio.0.35.16

Keating JP, Brophy D, Officier RA, Mullins E (2014) Otolith shape analysis of blue whiting suggests a complex stock structure at their spawning grounds in the Northeast Atlantic. Fisheries Research 157: 1–6. https://doi.org/10.1016/j.fishres.2014.03.009

L'Abée-Lund JH (1988) Otolith shape discriminates between juvenile Atlantic salmon, *Salmo salar L.*, and brown trout, *Salmo trutta L.* Journal of Fish Biology 33(6): 899–903. https://doi.org/10.1111/j.1095-8649.1988.tb05538.x

Lombarte A, Cruz A (2007) Otolith size trends in marine fish communities from different depth strata. Journal of Fish Biology 71(1): 53–76. https://doi.org/10.1111/j.1095-8649.2007.01465.x

Lombarte A, Palmer M, Matallanas J, Gómez-Zurita J, Morales-Nin B (2010) Ecomorphological trends and phylogenetic inertia of otolith sagittae in Nototheniidae. Environmental Biology of Fishes 89(3–4): 607–618. https://doi.org/10.1007/s10641-010-9673-2

Lychakov DV, Rebane YT (2004) Otolith mass asymmetry in 18 species of fish and pigeon. Journal of Gravitational Physiology 11: 17–34.

Lychakov DV, Rebane YT (2005) Fish otolith mass asymmetry: Morphometry and influence on acoustic functionality. Hearing Research 201(1–2): 55–69. https://doi.org/10.1016/j.heares.2004.08.017

Lychakov DV, Rebane YT, Lombarte A, Fuiman LA, Takabayashi T (2006) Fish otolith asymmetry: Morphometry and modelling. Hearing Research 219(1–2): 1–11. https://doi.org/10.1016/j.heares.2006.03.019

Lychakov DV, Rebane YT, Lombate A, Demestre M, Fuiman L (2008) Saccular otolith mass asymmetry in adult flatfishes. Journal of Fish Biology 72(10): 2579–2594. https://doi.org/10.1111/j.1095-8649.2008.01869.x

Mejri M, Trojette M, Allaya H, Ben Faleh A, Jmil Chalh A, Quignard JP, Trabelsi M (2018a) Stock Discrimination of two local populations of *Pagelluserythrinus* (Actinopterygii, Sparidae, Perciformes) in Tunisian waters by Analysis of Otolith Shape. Cahiers de Biologie Marine 59(6): 579–587. https://doi.org/10.21411/CBM.A.1DF06B15

Mejri M, Trojette M, Allaya H, Ben Faleh A, Jmil I, Chalah A, Quignard JP, Trabelsi M (2018b) Use of otolith shape to differentiate two lagoon populations of *Pagellus Erythrinus* (Achtinopterygii: Perciformes: Sparidae) in Tunisian waters. Acta Ichthyologica et Piscatoria 48(2): 153–161. https://doi.org/10.3750/AIEP/02376

Mille T, Mahe K, Villanueva MC, De Pontual H, Ernande B (2015) Sagittal otolith morphogenesis asymmetry in marine fishes. Journal of Fish Biology 87(3): 646–663. https://doi.org/10.1111/jfb.12746

Munday PL, Hernaman V, Dixson DL, Thorrold SR (2011) Effect of ocean acidification on otolith development in larvae of a tropical marine fish. Biogeosciences Discussions 8: 2329–2356. https://doi.org/10.5194/bgd-8-2329-2011

Nazir A, Khan MA (2019) Spatial and temporal variation in otolith chemistry and its relationship with water chemistry: Stock discrimination of *Sperata aor*. Ecology Freshwater Fish 28(3): 499–511. https://doi.org/10.1111/eff.12471

Panfili J, De Pontual H, Troadec H, Wright PJ (2002) Manuel de sclérochronologie des poisons. IFREMER-IRD, Brest, 463 pp. https://doi.org/10.4000/books.irdeditions.20901

Paxton JR (2000) Fish otoliths: Do sizes correlate with taxonomic group, habitat and/or luminescence? Philosophical Transactions of the Royal Society of London – Series B, Biological Sciences 355(1401): 1299–1303. https://doi.org/10.1098/rstb.2000.0688

Perry DM, Redman DH, Widman Jr JC, Meseck S, King A, Pereira JJ (2015) Effect of ocean acidification on growth and otolith condition of juvenile scup, *Stenotomus chrysops*. Ecology and Evolution 5(18): 4187–4196. https://doi.org/10.1002/ece3.1678

Pracheil BM, George R, Chakoumakos BC (2019) Significance of otolith calcium carbonate crystal structure diversity to microchemistry studies. Reviews in Fish Biology and Fisheries 29(3): 569–588. https://doi.org/10.1007/s11160-019-09561-3

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

Vrdoljak D, Matic-Skoko S, Peharda M, Uvanovic H, Markulin K, Mertz-Kraus R (2020) Otolith fingerprints reveals potential pollution exposure of newly settled juvenile *Sparus aurata*. Marine Pollution Bulletin 160: 111695. https://doi.org/10.1016/j.marpolbul.2020.111695

Walther BD, Limburg KE (2012) The use of otolith chemistry to characterize diadromous migrations. Journal of Fish Biology 81(2): 796–825. https://doi.org/10.1111/j.1095-8649.2012.03371.x

Wang WX (2002) Interactions of trace metals and different marine food chains. Marine Ecology Progress Series 243: 295–309. https://doi.org/10.3354/meps243295

XLSTAT (2007) XLSTAT Version 2007.1.02. Microsoft Excel. Copyright Addinsoft 1995–2007.

Yedier S, Bostancı D, Kontaş S, Kurucu G, Polat N (2018) Comparison of Otolith Mass Asymmetry in Two Different *Solea solea* Populations in Mediterranean Sea. Ordu University Journal of Science and Technology 8(1): 125–133.