UNIVERSIDAD DEL ZULIA
Serbiluz
Sistema de Servicios Bibliotecarios y de Información
Biblioteca Digital
Repositorio Académico

https://doi.org/10.52973/rcfcv-e35526 Revista Científica, FCV-LUZ / Vol. XXXV

Analysis of mineral and heavy metals in fish otoliths in theTigris River, Turkey

Análisis de minerales y metales pesados en otolitos de peces del río Tigris, Turquía

Muhammed Yaşar Dörtbudak[1], Hikmet Dinç[2], Serbest Bilici[3]*

[1]Harran University, Veterinary Faculty, Department of Fisheries and Diseases, Sanlıurfa, Turkey.
[2]Gaziantep Islamic Science and Technology University, School of Medicine, Pharmacology Department, Gaziantep, Turkey.
[3]Şırnak University, Faculty of Agriculture, Department of Animal Science, Şırnak, Turkey.
*Corresponding author: serbestbilici@hotmail.com.tr - serbestbilici@gmail.com

ABSTRACT

Rising pollution levels pose significant threats to fisheries. By analysing the different components of fish body structures, the interactions that occur in response to environmental changes can be better understood. Otoliths are structures in the inner ears of fish and record environmental changes that fish are exposed to throughout their lifes. Recent studies have shown that fish otoliths provide information on the accumulation of mineral and heavy metal in the environment. The accumulation of mineral and heavy metal in fish otoliths can be an important indicator for understanding environmental interactions and ultimately assessing the sustainability of fishery resources. In this study, 62 samples of *Acanthobrama marmid, Alburnus mossulensis, Paracapoeta trutta, Capoeta umbla, Carassius gibelio, Chondrostoma regium, Cyprinion kais, Cyprinion macrostomum, Luciobarbus mystaceus* and *Planiliza abu* were obtained from fishermen in the Tigris River. The presence of Co, Cr, Cu, Fe, Mn, Ni, Pb and Zn were analysed by inductively couple plasma optical emission apectrometry (ICP-OES) in the otoliths. The average levels of heavy metals residues in the otoliths were determined as Co>Cr>Cu>Fe>Mn>Ni>Pb and Zn. The minerals Ca, K, Mg, Na and P were found to be statistically significant among fish species (P<0.05). According to Tukey HSD multiple comparison test, the highest values of Ca, K and Na were found in P. abu and Mg in *C. gibelio*. The data can be used as a reference for the evaluation of the accumulation of mineral and heavy metal in fish otoliths in terms of fishery management and environmental protection, and can be compared with the data from studies in different fisheries.

Key words: Otolith; heavy metals; minerals in fish; Tigris river

RESUMEN

Los niveles crecientes de contaminación plantean amenazas significativas para la pesca. Al analizar los diferentes componentes de las estructuras corporales de los peces, se pueden comprender mejor las interacciones que ocurren en respuesta a los cambios ambientales. Los otolitos son estructuras en los oídos internos de los peces y registran los cambios ambientales a los que están expuestos los estos a lo largo de su vida. Estudios recientes han demostrado que los otolitos de los peces brindan información sobre la acumulación de minerales y metales pesados en el ambiente. La acumulación de minerales y metales pesados en los otolitos de los peces puede ser un indicador importante para comprender las interacciones ambientales y, en última instancia, evaluar la sostenibilidad de los recursos pesqueros. En este estudio, se obtuvieron 62 muestras de *Acanthobrama marmid, Alburnus mossulensis, Paracapoeta trutta, Capoeta umbla, Carassius gibelio, Chondrostoma regium, Cyprinion kais, Cyprinion macrostomum, Luciobarbus mystaceus* y *Planiliza abu* de pescadores en el río Tigris. La presencia de Co, Cr, Cu, Fe, Mn, Ni, Pb y Zn se analizó mediante espectrometría de emisión óptica de plasma inductivamente acoplado (ICP-OES) en los otolitos. Los niveles promedio de residuos de metales pesados en los otolitos se determinaron como Co>Cr>Cu>Fe>Mn>Ni>Pb y Zn. Se encontró que los minerales Ca, K, Mg, Na y P eran estadísticamente significativos entre especies de peces (P<0,05). Según la prueba de comparación múltiple de Tukey HSD, los valores más altos de Ca, K y Na se encontraron en P. abu y de Mg en *C. gibelio*. Los datos se pueden utilizar como referencia para la evaluación de la acumulación de minerales y metales pesados en los otolitos de peces en términos de gestión de la pesca y protección ambiental, y se pueden comparar con los datos de estudios de diferentes pesquerías.

Palabras clave: Otolitos; metales pesados; minerales en peces; río Tigris

Recibido: 03/10/2024 Aceptado: 11/01/2025 Publicado: 23/03/2025

INTRODUCTION

Otoliths are calcified structures located inside the inner ear cavity of teleost fish and form part of the organs that provide hearing and sense of balance [1 , 2]. Otolith is a type of calcium carbonate biomineral that grows in the inner ear of teleost fish. There are three pairs of otoliths: asteriscus, lapillus and sagitta. Asteriscus are the most commonly used otoliths in Cypriniformes fish species such as carp. For these species, the asteriscus is the largest of the three otolith pairs [3 , 4]. Compared to muscle and visceral data, otoliths stands out more by showing clear diurnal increments that can record accurate information over years, seasons, months and even down to a single day [5 , 6].

The elements deposited in otolith growth layers cannot be modified, absorbed or reconstituted [7 , 8]. Otoliths grow continuously throughout a fish's life as a result of the deposition of calcium carbonate crystals on a matrix of proteins. During this deposition, elements from the resident waters pass through the gills into the bloodstream and are contained in the calcium carbonate protein cage of the otolith. The chemical composition of the otolith is thought to depend primarily on the environmental conditions of the water body in which the fish live, although it is not directly related to ambient water chemistry, primarily due to the effects of physiological processes [9 , 10]. In particular, the uptake of metals is usually proportional to calcium (Ca), which inhibits uptake at high relative concentrations [1 , 11 , 12].

Otolith trace metals (i.e. metals other than Ca, Na and Sr) constitute only 1% of the total otolith mass [13]. The otolith mainly contains calcium and protein carbonates, which continue to be stored with increasing age but are not reused by physiological processes [14 , 15 , 16].

Otolith chemical composition can represent a reliable natural label of the environment in which the organism lives. The mechanism of otolith uptake of trace elements is not fully understood due to the complex behavior of different metals and the fact that incorporation between metals varies greatly [17]. Moreover, the bio-mineralization process is a function of different biological and environmental factors [9 , 18 , 19 , 20 , 21 , 22] and may be physiological differences or species-specific ontogenics [9 , 17 , 21 , 23 , 24 , 25 , 26 , 27]. Although the study of elements ranging from water to carbonate structure is complex, it is undeniable that the chemical analysis of otoliths is a valuable tool used to identify individuals from different regions and thus obtain information on habitat use [22 , 28 , 29].

The majority of metal contamination studies in fish have so far focused on soft tissues (such as liver, kidney, gills and muscle), which are known to be target organs for metal accumulation. Less data are available on calcareous tissues as an indicator of metal contamination. However, in these tissues, several metals are incorporated during calcification; some of them (e.g. Pb) are of high importance during acidification processes [30 , 31].

Fish samples in freshwater systems are considered one of the most decisive factors for the estimation of trace metals pollution potential [32 , 33 , 34]. Therefore, many studies on heavy metal accumulation in fish have been conducted and published [34 , 35 , 36 , 37 , 38].

Planiliza abu, *A. marmid*, *A. mossulensis*, *C. kais*, *C. macrosto*mum, *P. trutta*, *C. umbla*, *C. gibelio* and *L. mystaceus* fish are widely consumed as food by the people living in our study area [39]. In this study, otoliths in fish were used as the main research subject because they accumulate heavy metals that cause environmental pollution and are a marker to reveal the level of pollution.

MATERIAL AND METHODS

Heavy metal analysis in otoliths

62 fish of different species were collected from fishermen working in the Tigris River. The asterisk otoliths were removed, cleaned and stored dry in labeled boxes. Otoliths were removed from fish with plastic tweezers and stored in dry plastic bottles [40]. For analysis, otolith samples (right and left pairs) were weighed with the And Hr-250 Low Precision Balance and placed in 15 mL tubes. They were placed in a hot water bath for complete dissolution. When the samples were completely dissolved and no particles remained, they were topped up to 15 mL with distilled water. After dissolution of all samples was complete, Otolith samples were analyzed twice for Co, Cr, Cu, Fe, Mn, Ni, Pb and Zn using a inductively coupled plasma optical emission spectrometer Perkin Elmer Optima 5300 DV ICP/OES, a fast multi-element technique with dynamic linear range and moderate detection limits [41]. Heavy metal concentrations were expressed as µg g-1 as wet weight of tissue. Absorption wavelength: 267.716 nm for Cr; 228.616 nm for Co; 327.393 nm for Cu; 238.204 nm for Fe; 257.610 nm for Mn; 231.604 nm for Ni; 220.353 nm for Pb and 206.200 nm for Zn. Digestion and analytical procedures were controlled by analysis of standard reference material (DORM-2 and DOLT-3 National Research Council Canada, Ottawa, Ontario, Canada).

Statistical analysis

SPSS v.23 (IBM Corp, USA) package program was used for all analyses [42]. Shapiro-Wilk and Levene's test were used for normal distribution and homogeneity assumptions, respectively ($P>0.05$). Differences between sexes according to the related element within the species were analyzed by independent t-test. One-way analysis of variance was applied to investigate whether there was a difference between the species in terms of the elements examined, and Tukey HSD multiple comparison test was used for species with significant differences between groups ($P<0.05$). For each species, a Pearson correlation matrix was created between the elements analyzed. Data were presented as minimum, maximum, mean, standard error and analyzed at $P<0.05$ level. The results obtained were statistically expressed as mean, standard deviation and P values were calculated using ANOVA analysis of variance [43].

RESULTS AND DISCUSION

For the Brond-snout *Chondrostoma regium*, there was a statistically significant difference between the sexes in terms of Ca, Mg and P ($P<0.05$), while no significant difference was found between the other elements ($P>0.05$). In all three elements, the values of male individuals were significantly higher than female individuals.

For the Tigris scraper *Capoeta umbla*, there was a statistically significant difference between the sexes in terms of Ca, Mg and Na ($P<0.05$), while there was no significant difference between the other elements (P>0.05). In all three elements, the values of female individuals were significantly higher than male individuals (P<0.05). There was no significant difference between the sexes for the elements examined in other species (P>0.05).

There was a statistically significant difference between fish species for Ca, K, Mg, Na and P (P<0.05) (TABLE I) (FIG. 1). When the highest and lowest differences between species were evaluated according to Tukey HSD multiple comparison test.

In terms of Ca, the Abu mullet (*Planiliza abu)* (16,690.00 ± 726,97 a) had the highest value, and the Tigris bream (*Acanthobrama marmid)* (1,873.2 ± 190.61 d), the Mossul bleak (*Alburnus mossulensis)* (1,334.48 ± 216.36 d), the Kais kingfish (*Cyprinion kais)* (1,063.56 ± 139.61 d) and the Tigris kingfish (*Cyprinion macrostomum)* (1,463.42 ± 195.68 d) they did not have significant statistical differences and showed the lowest values.

In terms of K, the Abu mullet (*Planiliza abu)* (27.29 ± 1.393 a) had the highest value, while the Tigris bream (*Acanthobrama marmid)* (2.15 ± 0.19 c), the Mossul bleak (*Alburnus mossulensis)* (2.36 ± 0.49 c), the Kais kingfish (*Cyprinion kais)* (1.04 ± 0.24 c), the Tigris kingfish (*Cyprinion macrostomum)* (2.02 ± 0.35 c), the Long thorn scratcher (*Paracapoeta trutta)* (3.53 ± 0.38 c) and the Tigris scraper (*Capoeta umbla)* (3.94 ± 0.87 c) they didn't show statistically significant differences and had the lowest values. In terms of Mg, the Prussian carp (*Carassius gibelio)* (9.46 ± 1.66 a) had the highest value, while the Kais kingfish (*Cyprinion kais)* (0.47 ± 0.14 c) and the Tigris kingfish (*Cyprinion macrostomum)* (0.86 ± 0.17 c) had the lowest values and without significant statical differences between them.

In terms of Na, the Abu mullet (*Planiliza abu)* (80.58 ± 3.10 a) had the highest value, while the Tigris bream (*Acanthobrama marmid)* (3.39 ± 0.30 c), the Mossul bleak (*Alburnus mossulensis)* (3.66 ± 0.84 c), the Kais kingfish (*Cyprinion kais)* (1.39 ± 0.33 c), the Tigris kingfish (*Cyprinion macrostomum)* (2.56 ± 0.44 c) and the Long thorn scratcher (*Paracapoeta trutta)* (7.03 ± 0.84 c) had the lowest values and they did not show significant statistical differences between them. In terms of P, the Abu mullet (*Planiliza abu)* (3.06 ± 0.08 a) had the highest value and the Kais kingfish (*Cyprinion kais)* (0.83 ± 0.11 d) had the lowest value.

TABLE I.
Average mineral concentrations according to fish species in theTigris River

Species	n	Ca (Mean ± Sh) mg kg-1	K (Mean ± Sh) mg kg-1	Mg (Mean ± Sh) mg kg-1	Na (Mean ± Sh) mg kg-1	P (Mean ± Sh) mg kg-1
Acanthobrama marmid	5	1873.20±190.61 d	2.15±0.19c	1.81 ± 0.18 bc	3.39 ± 0.30 c	1.07 ± 0.09 cd
Alburnus mossulensis	4	1334.48±216.36d	2.36 ± 0.49 c	1.33 ± 0.33 bc	3.66 ± 0.84 c	1.07 ± 0.14 cd
Carassius gibelio	7	5560.43±983.44 b	9.29 ± 1.83 b	9.46 ± 1.66 a	16.61 ± 2.95 b	2.88 ± 0.38 ab
Cyprinion kais	5	1063.56± 139.61 d	1.04 ± 0.24 c	0.47 ± 0.14 c	1.39 ± 0.33 c	0.825 ± 0.11 d
Cyprinion macrostomum	6	1463.42± 195.68 d	2.02 ± 0.35 c	0.86 ± 0.17 c	2.56 ± 0.44 c	1.17 ± 0.18 cd
Chondrostoma regium	6	4146.67±1207.58 bc	4.86 ± 1.45 bc	2.53 ± 0.69 bc	7.28 ± 2.23 bc	1.88±0.48 abcd
Paracapoeta trutta	11	2940.09±325.34 bc	3.53 ± 0.38 c	3.03 ± 0.37 bc	7.03 ± 0.84 c	1.51±0.13 bcd
Capoeta umbla	6	2959.50±602.67 bc	3.94 ± 0.87 c	2.54 ± 0.68 bc	8.19 ± 2.09 bc	1.76±0.30 abcd
Planiliza abu	5	16690.00±726.97 a	27.29 ± 1.39 a	-	80.58 ± 3.10 a	3.06 ± 0.08 a
Luciobarbus mystaceus	7	3739.29±566.25 bc	5.65 ± 1.45 bc	5.27 ± 1.41 ab	10.15±2.55 bc	2.52 ± 0.51 abc
P-value	62	0.000***	0.000***	0.000***	0.000***	0.000***

n: number of individuals, Ca: Calcium, K: Potassium, Mg: Magnesium Na: Sodium P: Phosphate.

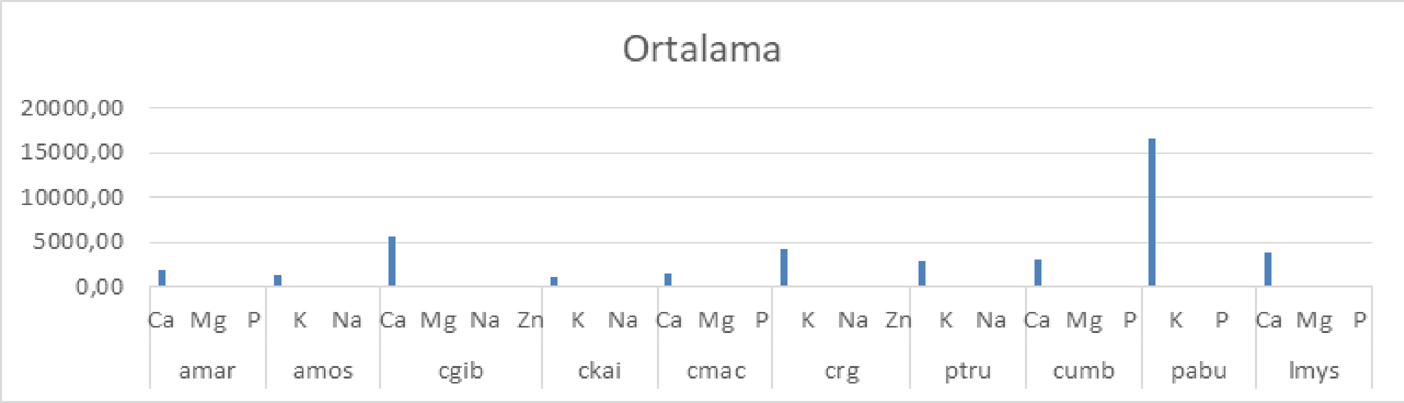

FIGURE 1. Average mineral values by fish species in theTigris River

For the Brond-snout *Chondrostoma regium* according to the results of Pearson correlation analysis, Ca, K, Mg, Na and P elements had statistically strong correlation with each other (lowest correlation 0.94) (P<0.05), while these elements had low correlation with Zn (P>0.05) (TABLE II).

TABLE II.
Correlation table of the Brond-snout Chondrostoma Regium.

Elements		Ca	K	Mg	Na	P
K	r	0.986				
	p	0.000***				
	n	6				
Mg	r	0.994	0.995			
	p	0.000***	0.000***			
	n	6	6			
Na	r	0.942	0.983	0.962		
	p	0.004**	0.000***	0.002**		
	n	6	6	6		
P	r	0.989	0.991	0.993	0.971	
	p	0.000***	0.000***	0.000***	0.001**	
	n	6	6	6	6	
Zn	r	-0.306	-0.154	-0.242	0.023	-0.206
	p	0.556	0.771	0.643	0.965	0.696
	n	6	6	6	6	6

For the Tigris kingfish *Cyprinion macrostomum* according to the results of Pearson correlation analysis, Ca, K, Mg, Na and P elements were statistically strongly correlated with each other (except Ca-K correlation) (lowest correlation 0.742) (P<0.05) (TABLE III).

TABLE III.
Correlation table of the Tigris kingfish Cyprinion macrostomum.

		Ca	K	Mg	Na
K	r	0.742			
	p	0.091			
	n	6			
Mg	r	0.826	0.95		
	p	0.043*	0.003**		
	n	6	6		
Na	r	0.848	0.968	0.994	
	p	0.032*	0.001**	0.000***	
	n	6	6	6	
P	r	0.837	0.903	0.979	0.972
	p	0.037*	0.013*	0.000***	0.001**
	n	6	6	6	6

For the Kais kingfish *Cyprinion kais*; according to the results of Pearson correlation analysis, Ca, K, Mg, Na and P elements had a statistically strong relationship with each other (the lowest correlation was 0.927 for Na-P pair) (P<0.05) (TABLE IV).

Tablo IV. Correlation table of the Kais kingfish Cyprinion Kais.

		Ca	K	Mg	Na
K	r	0.966			
	p	0.007**			
	n	5			
Mg	r	0.976	0.994		
	p	0.004**	0.000***		
	n	5	5		
Na	r	0.966	0.995	0.988	
	p	0.007**	0.000***	0.001**	
	n	5	5	5	
P	r	0.986	0.94	0.962	0.927
	p	0.002*	0.017*	0.008**	0.023*
	n	5	5	5	5

Many international studies have recognized otoliths as a new option for monitoring toxic element accumulation in fish [44 , 45] Many international studies have been conducted to detect toxic elements in these calcareous structures of fish [17 , 40 , 46].

Campana [1] conducted research to confirm that otolith chemistry can detect differences between freshwater and marine fishes, to determine the scale of variation in trace element concentrations between habitats, to examine links with temperature, salinity, growth and freshwater flow volumes, and to assess the role of freshwater flows for freshwater fishes that may have returned to the estuary after a major flood event. Previous otolith studies have shown that salinity, temperature and growth rate can affect the otolith chemistry of some marine species, but the pattern varies [1]. Partial correlations with linear, nonlinear and inverse effects could not explain a significant proportion of the variance in otolith chemistry [47 , 48]. The relationship between otolith and water concentrations will be strongly influenced by the residence time of each fish in different parts of the estuary at different times. Despite these potential confounding effects, otolith Sr/Ca has proven to be an unambiguous determinant of fish movement between freshwater and estuary.

Ranaldi and Gagnon [17] found differences in trace element concentrations in otoliths of fish between different sampling locations. In our study, different species of fish from the same location were examined. They concluded that non-essential metals such as Cd and Pb represent good markers of dietary/waterborne exposure, while essential metals such as Zn may be predominantly associated with dietary intake and therefore exposure history should be taken into account when interpreting trace metal compositions in otoliths of wild fish and subsequent susceptibility to metal pollution. The use of inductively Coupled Plasma Mass Spectrometry (ICP-MS) provides sensitive spatial information that has the potential to help accurate reconstruction of the fish's habitat. Since we used Inductively Coupled Plasma-Optical Emission Spectrophotometer (ICP-OES) in our study, we could not obtain some sensitive information.

Herrera-Reveles *et al.* [40] analyzed the otoliths of fish in Venezuela for heavy metals using Energy Dispersive X-ray spectroscopy (EDS) stabilized by scanning electron microscopy (SEM) and found that five heavy metals (Cd, Cu, Hg, Pb and Zn) were detected mostly in the outer layers of otoliths in fish from all regions. The highest values of Pb/Ca and Hg/Ca weights were observed. These results showed significant spatial variation in otoliths, providing evidence of different Cd, Hg and Pb concentrations in the water and/or sediments of these locations.

CONCLUSION

In light of the data obtained in our study through the inductively couple plasma optical emission apectrometry (ICP-OES), the amount of minerals detected in the otoliths of the fish summarized the situation due to diet/water exposure in fish species living in the Tigris River and is expected to be a source for future studies.

ACKNOWLEDGMENTS

We are highly thankful to the HUBAP (Harran University Scientific Research Projects Unit) for their support in this project. Project number: 18118

Conflict of interests

The authors declare that they have no conflict of interests in publishing this manuscript.

BIBLIOGRAPHIC REFERENCES

[1] Campana SE. Chemistry and composition of fish otoliths: pathways, mechanisms and applications. Mar. Ecol. Prog. Ser. [Internet]. 1999; 188:263-297. doi: https://doi.org/c2mc46

[2] Campana SE, Thorrold SR. Otoliths, increment and elements: keys to a comprehensive understanding of fish populations? Can. J. Fish. Aquat. Sci. [Internet]. 2001; 58(1):30–38. doi: https://doi.org/cs4ckn

[3] Phelps QE, Edwards KR, Willis DW. Precision of five structures for estimating age of common carp. N. Am. J. Fish. Manag. [Internet]. 2007; 27(1): 103-105. doi: https://doi.org/cdfgj9

[4] Jawad L, Mahé K. Fluctuating asymmetry in asteriscii otoliths of common carp (*Cyprinus carpio*) collected from three localities in Iraqi rivers linked to environmental factors. Fishes. [Internet]. 2022; 7(2):91. doi: https://doi.org/n96q

[5] Tanabe T, Kayama S, Ogura M, Tanaka S. Daily increment formation in otoliths of juvenile skipjack tuna *Katsuwonus pelamis*. Fish. Sci. [Internet]. 2003; 69(4):731-737. doi: https://doi.org/b3vkbc

[6] Morales-Nin B, Panfili J. Seasonality in the deep sea and tropics revisited: what can otoliths tell us? Mar. Freshw. Res. [Internet]. 2005; 56(5):585-598. doi: https://doi.org/cdrvgk

[7] Gao YW. Stable isotope analyses in otoliths of cod (*Gadus morhua* L., 1758): implication for long-term environmental changes in the Canadian Atlantic. Hamilton: McMaster University, 1997; p. 96. Available in: https://goo.su/B9030R

[8] Ren D, Yonghua G, Qingling F. Enrichment of Pb, Hg and Cr in cultured carp otolith. Afr. J. Biotechnol. [Internet]. 2012; 11(8):1939-1947. doi: https://doi.org/n96r

[9] Kalish JM. Otolith microchemistry: Validation of the effects of physiology, age and environment on otolith composition. J. Exp. Mar. Biol. Ecol. [internet]. 1989; 132(3):151-178. doi: https://doi.org/bjsq5s

[10] Fowler AJ, Campana SE, Thorrold SR, Jones CM. Experimental assessment of the effect of temperature and salinity on elemental composition of otoliths using laser ablation ICPMS. Can. J. Fish. Aquat. Sci. [Internet]. 1995; 52(7):1431-1441. doi: https://doi.org/cwwjnh

[11] Bath GE, Thorrold SR, Jones CM, Campana SE, Mclaren JM, Lam JWH. Strontium and barium uptake in aragonitic otoliths of marine fish. Geochim. Cosmochim. Acta. 2000; 64(10):1705-1714. doi: https://doi.org/c28c5p

[12] Kraus RT, Secor DH. Incorporation of strontium into otoliths of an estuarine fish. J. Exp. Mar. Biol. Ecol. [Internet]. 2004; 302(1):85-106. doi: https://doi.org/dbxzsp

[13] Thresher RE. Elemental composition of otoliths as a stock delineator in fishes. Fish. Res. [Internet]. 1999; 43(1-3):165-204. doi: https://doi.org/d763f2

[14] Degens ET, Deuser WG, Haedrich RL. Molecular structure and composition of fish otoliths. Mar. Biol. [Internet]. 1969 [cited Jul. 22 2024]; 2:105-113. Available in: https://goo.su/U7XA6l

[15] Mugiya Y, Hakomori T; Hatsutori K. Trace metal incorporation into otoliths and scales in the goldfish, *Carassius auratus.* Comp. Biochem. Physiol. C. Comp. Toxicol. [Internet]. 1991; 99(3):327-331. doi: https://doi.org/d2mhrt

[16] Pannela G. Fish otoliths: daily growth layer and periodical patterns. Science [Internet]. 1971; 173(4002):1124–1127. doi: https://doi.org/dz22zx

[17] Ranaldi MM, Gagnon MM. Trace metal incorporation in otoliths of pink snapper (*Pagrus auratus*) as an environmental monitor. Comp. Biochem. Physiol. C. Toxicol. Pharmacol. [Internet]. 2010; 152(3):248–255. doi: https://doi.org/fmthmm

[18] Secor DH, Ohta T, Nakayama K, Tanaka M. Use of otolith microanalysis to determine estuarine migrations of Japanese sea bass *Lateolabrax japonicus* distributed in Ariake Sea. Fish. Sci. [Internet]. 1998; 64(5):740–743. doi: https://doi.org/n96t

[19] Limburg KE. Otolith strontium traces environmental history of subyearling American shad *Alosa sapidissima.* Mar. Ecol. Prog. Ser. [Internet]. 1995 [cited Jul. 19 2024]; 119(1-3):25–35. Available in: https://goo.su/16Hk9Fj

[20] Sadovy Y, Severin KP. Trace elements in biogenic aragonite: correlation of body growth rate and strontium levels in the otoliths of the white grunt, *Haemulon plumier* (Pisces: Haemulidae). Bull. Mar. Sci. 1992; 50:237-257. Available in: https://goo.su/VkFkdyx

[21] Gillanders BM, Kingsford MJ. Spatial variation in elemental composition of otoliths of three species of fish (family Sparidae). Estuar. Coast. Shelf. Sci. [Internet]. 2003; 57(5-6):1049–1064. doi: https://doi.org/b8x6n4

[22] Pontual H, Bertignac M, Battaglia A, Bavouzet G, Moguedet P; Groison AL. A pilot tagging experiment on European hake (*Merluccius merluccius*): methodology and preliminary results. ICES J. Mar. Sci. [Internet]. 2003; 60(6):1318–1327. doi: https://doi.org/fs46hs

[23] Elsdon TS, Gillanders BM. Interactive effects of temperature and salinity on otolith chemistry: challenges for determining environmental histories of fish. Can. J. Fish. Aquat. Sci: 2002; 59(11):1796–1808. doi: https://doi.org/cfh5kp

[24] Swearer SE, Forrester GE, Steele MA, Brooks AJ, Lea DW. Spatio-temporal and interspecific variation in otolith trace-elemental fingerprints in a temperate estuarine fish assemblage. Estuar. Coast. Shelf Sci. [Internet]. 2003; 56(5-6):1111–1123. doi: https://doi.org/c2w9b4

[25] Martin GB, Thorrold SR. Temperature and salinity effects on magnesium, manganese, and barium incorporation in otoliths of larval and early juvenile spot *Leiostomus xanthurus*. Mar. Ecol. Prog. Ser. [Internet]. 2005; 293:223–232. doi: https://doi.org/bzvd7c

[26] Vasconcelos RP, Reis-Santos P, Tanner S, Fonseca V, Latkoczy C, Günther D, Costa MJ, Cabral H. Discriminating estuarine nurseries for five fish species through otolith elemental fingerprints. Mar. Ecol. Prog. Ser. [Internet]. 2007; 350:117–126. doi: https://doi.org/frnrv6

[27] Reis Santos P, Vasconcelos R, Ruano M, Latkoczy C, Günther D, Costa MJ, Cabral H. Interspecific variation of otolith chemistry in estuarine fish nurseries. J. Fish Biol. [Internet]. 2008; 72(10):2595–2614. doi: https://doi.org/c792cf

[28] Thorrold SR, Jones CM, Campana SE. Response of otolith microchemistry to environmental variations experienced by larval and juvenile Atlantic croaker (*Micropogonius undulatus*). Limnol. Oceanogr. [Internet]. 1997; 42(1):102–111. doi: https://doi.org/frnnsm

[29] Rooker JR, Secor DH, DeMetrio G, Schloesser R, Block BA, Neilson J.D. Natal homing and connectivity in Atlantic bluefin tuna populations. Sci. [Internet]. 2008; 322(5902):742–744. doi: https://doi.org/c966md

[30] Moreau G, Barbeau C, Frenette JJ, Saint-Onge J, Simoneau M. Zinc, manganese, and strontium in opercula and scales of brook trout (*Salvelinus fontinalis*) as indicators of lake acidification. Can. J. Fish. Aquat. Sci. [Internet]. 1983; 40(10):1685-1691. doi: https://doi.org/cj694q

[31] Gauldie R W, Fournier DA, Dunlop DE, Coote G. Atomic emission and proton microprobe studies of the ion content of otoliths of chinook salmon aimed at recovering the temperature life history of individuals. Comp. Biochem. Physiol. A, Physiol. [Internet]. 1986; 84(4):507-515. doi: https://doi.org/b8vxr9

[32] Evans DW, Dodoo DK and Hanson DJ. Trace elements concentrations in fish livers: Implications of variations with fish size in pollution monitoring. Mar. Pollut. Bull. [Internet]. 1993; 26(6):329–334. doi: https://doi.org/bzdr3x

[33] Rashed MN. Monitoring of environmental heavy metals in fish from Nasser Lake. Environ. Int. [Internet]. 2001; 27(1):27–33. doi: https://doi.org/fwnrjd

[34] Papagiannis I, Kagalou I, Leonardos J, Petridis D, Kalfakakou V. Copper and zinc in four freshwater fish species from Lake Pamvotis (Greece), Environm. Int. [Internet]. 2004; 30(3):357–362. doi: https://doi.org/dvq4pz

[35] Guhathakurta H, Kaviraj A. Heavy metal concentration in water, sediment, shrimp (*Penaeus monodon*) and mullet (*Liza parsia*) in some brackish water ponds of Sunderban, India. 2000. Mar. Pollut. Bull. [Internet]. 2000; 40(11):914-920. doi: https://doi.org/b9tmgh

[36] Kargin F. Metal concentrations in tissues of the freshwater fish *Capoeta barroisi* from the Seyhan River (Turkey). Bull. Environ. Contam. Toxicol. [Internet]. 1998; 60(5):822–828. doi: https://doi.org/crmct4

[37] Canli M, Atli G. The relationships between heavy metal (Cd, Cr, Cu, Fe, Pb, Zn) levels and the size of six Mediterranean fish species. Environ. Pollut. [Internet]. 2003; 121(1):129–136. doi: https://doi.org/c6vkwh

[38] Fernandes C, Fontaínhas-Fernandes A, Peixoto F, Salgado MA. Bioaccumulation of heavy metals in *Liza saliens* from the Esmoriz-Paramos coastal lagoon, Portugal. Ecotoxicol. Environ. Saf. [Internet]. 2007; 66(3):426–431. doi: https://doi.org/cs99z5

[39] Karadede-Akin H, Ünlü E. Heavy metal concentrations in water, sediment, fish and some benthic organisms from Tigris River, Turkey. Environ. Monit. Assess. [Internet]. 2007; 131:323-337. doi: https://doi.org/bkcvvx

[40] Herrera-Reveles AT, Lemus M, Marín B, Prin JL. Trace metal incorporation in otoliths of a territorial coral reef fish (*Abudefduf saxatilis*) as an environmental monitoring tool. In: E3S Web of Conferences. Proceedings of the 16th Intrenational Conference on Heavy Metals in the Environment. EDP Sci. J. [Internet]. 2013; 1:34007. doi: https://doi.org/n983

[41] Sturgeon RE. Current practice and recent developments in analytical methodology for trace metal analysis of soils, plants and water. Commun. Soil Sci. Plant Anal. [Internet]. 2000; 31(11-14):1479-1512. doi: https://doi.org/b2sf35

[42] IBM Corp. Released. IBM SPSS Statistics for Windows, Version 23.0. Armonk: IBM Corp.2021 [cited Jul 22 2024] Available in: https://n9.cl/rc9b9

[43] Sümbüloğlu K, Sümbüloğlu V. Biyoistatistik. hatipoğlu basım ve yayım san. Tic. Ltd. Şti. Ankara, 2002.

[44] Gao Y, Feng Q, Ren D, Qiao L, Li S. The relationship between trace elements in fish otolith of wild carp and hydrochemical conditions. Fish Physiol. Biochem. [Internet]. 2010; 36(1):91-100. doi: https://doi.org/b3wkr6

[45] Fengqin D, Shengrong L, Lina Y, Wenjie L, Jing L, Wenyan S. Relationship of phosphorus content in carp otoliths with that in ambient water in Xiaoxi Port of the Taihu Lake, East China. Afr. J. Biotechnol. [Internet]. 2011; 10(54):11206-11213. doi: https://doi.org/n987

[46] Milton DA, Tenakanai CD, Chenery SR. Can the movements of barramundi in the Fly River region, Papua New Guinea be traced in their otoliths?. Estuar. Coast. Shelf Sci. [Internet]. 2000; 50(6):855–868. doi: https://doi.org/bpgq2m

[47] Tong SL, Ho CY, Pang FY. Monitoring of Ba, Mn, Cu and Ni during estuarine mixing. Anal. Sci. [Internet]. 1997; 13(Supplement):373-378. doi: https://doi.org/c4nnkm

[48] Elsdon TS, Gillanders BM. Temporal variability in strontium, calcium, barium, and manganese in estuaries: implications for reconstructing environmental histories of fish from chemicals in calcified structures. Estuar. Coast. Shelf Sci. [Internet]. 2006; 66(1-2):147-156. doi: https://doi.org/bdg7nj