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Biometric relation between body size and otolith size of *Cyprinion kais* and *C. macrostomum* collected from Tigris River, Şırnak Province, Türkiye

# by

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#### Abstract

One hundred and twenty asterisci were obtained from 30 specimens each of Cyprinion kais and Cyprinion macrostomum. Significant relationships between otolith mass and the three fish lengths (TL, SL, and FL) were attained for C. kais. Similar associations were obtained for OL and the three fish lengths of C. macrostomum. For C. kais, the highest in the relation between otolith mass, hereafter referred to as OM, and fish fork length, henceforth known as FL (0.5894). The lowest coefficient of determination was observed between otolith width, hereafter called OW, and standard fish length, hereafter referred to as SL (0.2861). For C. macrostomum, the highest coefficient of determination was attained for the relation between OL and FL (0.7280) and the lowest for the relationship between OM and TL (0.1717). The analysis shows that evaluations of the body size of the two fish species investigated through biometric studies of otoliths are trustworthy.

**Key words:** linear relationship, Cyprinidae, otolith morphometry, asteriscus, body length, freshwater

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## 1. Introduction

The Kais kingfish, Cyprinion kais, is a freshwater species in the benthopelagic habitat (Froese & Pauly, 2022). It is endemic to the Tigris-Euphrates Rivers basin, where it is reported from Iran, Syria, Turkey, and Iraq (Coad 2012). It reaches a maximum total length of 159 mm and a maximum registered weight of 49.90 g (Esmaeili et al. 2014a). The male-to-female ratio was 1:1, and males reached the age group three years and females four years (Esmaeili et al. 2014b). Individuals of this species feed on filamentous algae, aguatic insects, and detritus (Al-Rudainy 2008). This species reaches maturity at age two, with females spawning over sand, stones, and gravel in May-June (Ünlü 2006). The fecundity of this species can range between 295.1 and 1,252.2 eggs, with the meaning of the oocyte ranging from 0.14 to 0.86 mm (Alkan Uçkun & Gökçe 2015).

Freyhof (2014a) evaluated this species as the Least Concern on the Red List of the IUCN based on the following reasons. The species is confined to the areas that are characterised by flowing water. Individuals of this species usually move from this moving water region to canals and other aquatic habitats for feeding. It is widespread in the Tigris-Euphrates Rivers basin, representing several separate populations.

The Tigris kingfish, Cyprinion macrostomum, is a freshwater species that prefers to live in a benthopelagic environment (Froese & Pauly 2022). It reaches a maximum total length of 204 mm and a maximum reported weight of 91.20 g (Esmaeili et al. 2014b). Mature males differ from females in having large tubercles on the snout in a broad band below the nostril level, extending under the eye and breaking into a few tubercles on the operculum. Also, there is a prominent tubercle between the nostril and the eye, and fine tubercles are scattered over the top of the head (Coad 2012). Individuals of this species are distributed in the Orontes River (also known as the Asi River) and Quwayg River and the Tigris-Euphrates basins. In Iran, it is found in the Tigris River basin (Coad 2012). This species inhabits rivers, streams, lakes, dams, lagoons, ponds, springs, marshes, canals, jubes (irrigation ditches), and gravel pits (Pirani et al. 2013). It can live up to five years (Faghani Langroudi & Mousavi Sabet 2018). Individuals of this species reach maturity at 100-111 mm (Allouse et al. 1986). Male: female ratio was not considerably different from 1:1, and males and females attained age group 4, with age group 1 being the most abundant (Alkan Uçkun & Gökçe 2015). As main items, this species feeds on periphyton, including Navicula, Cymbellla, Diatoma, and Nitzschia (Marammazi et al. 2014). Zooplankton was believed to be unintentional food items. Occasionally, the intestine

included mud and sand, proof of a bottom-feeding habit (Khan 1988). Gonad indices revealed that spawning occurred from late May to mid-August when the water temperature was 16–24°C (Faghani Langroudi & Mousavi Sabet 2018).

Freyhof (2014b) evaluated this species as the Least Concern in the Red List of the IUCN based on the following reasons. This species is common, usually very abundant, and not eligible for a threat category.

Otoliths are heavy crystalline formations located in the inner ear of fish, which are created by the deposition of concentric layers of calcium carbonate in the structure of aragonite and the protein otoline and have annual growth rings. There are three pairs of otoliths in teleost: sagittae, lapilli, and asterisci (Campana & Casselman 1993, Green et al. 2009). The sagitta is frequently the largest otolith in most fishes (Tuset et al. 2008); nevertheless, the asteriscus is bigger in ostariophysian fishes (Harvey et al. 2000, Campana & Casselman 1993). Sagitta and asterisci otoliths vary in shape among species, while the shape of lapilli is more consistent (Campana 2004). The otoliths are considered chemically inert configurations because, after deposition of the layers, they remain meta- biologically inactive during the individual's life (Green et al. 2009). As valuable tools in taxonomy, the surface features of the otoliths can change during the growth of the individuals of the species and between different species of fish (de Assis et al. 2018). Regarding such features, a link was discovered between body size and otolith size (Altin & Ayyildiz 2017, Zan 2015). This biometric relationship is imperative in food investigations since otoliths are very resistant to processes of digestion, making it possible to recognise species and resolve prey size, which in turn permits improved scrutiny of predator-prey associations and feeding habitats of piscivorous species (Granadeiro & Silva 2000, Jobling & Breiby 1986). The biometric link could as well benefit from the resumption of paleontological research of modern bony fish, as otoliths are well-conserved configurations in fossils of marine species (Reichenbacher et al. 2007). The link between fish length and otolith size has been effectively applied in numerous studies for marine and freshwater fish species (Zan et al. 2015, Viva et al. 2015, Yilmaz et al. 2015, Jawad et al. 2017, Saygin et al. 2020, Jawad & Adams 2021).

With the significance and the practicability of these biometric relationships, the present work intended to confirm the presence of links between the biometric measurements of the asteriscus (length, width, mass) and the body size (total, standard, and fork length) of the Tigris kingfish, *C. macrostomum*, and the Kais kingfish, *C. kais* collected from the Tigris River, Şırnak

region, Türkiye and to develop equations to assess the body size of these species through otolith biometric features.

## 2. Materials and methods

The province of Şırnak is one of the important regions of Türkiye, located in southeastern Anatolia. It has a population of 403,607 (Baz 2016) and is rich in mountains in the west and the south. Still, the more significant part of the region comprises elevations created by the many rivers that cross it. The chief of these rivers is the Tigris River and its tributaries, the Hezil, the Kızılsu, and the Çağlayan (Baz 2016). The members of the family Cyprinidae dominate the freshwater system of Türkiye (ÇiÇek et al. 2020), and the two members of the genus Cyprinion are among the most common cyprinid species in the Tigris River basin (Bilecenoglu et al. 2014). Sixty specimens (30 specimens for each of C. kais and C. macrostomum) ranging in total length of 115 – 143 mm and 170 – 175 mm for C. kais and C. macrostomum, respectively were obtained from a local fisherman using a seine net (5 mm mesh; small and medium sizes). They fished in the Tigris River in the Şırnak region between Güçlükonak and Cizre from September 2015 to December 2015 (Fig. 1). Fish specimens of both species examined (Fig. 2) were kept on ice until reaching the laboratory. They were classified following Coad (2012). In the laboratory,



Figure 1 Map showing the location of sampling



## A) Cyprinion kais, 179 mm TL; B) C. macrostomum, 190 mm TL

the fish specimens were measured for total length (TL), standard length (SL), and fork length (FL), and were then dissected to remove the asteriscus otolith from both sides of the fish head. Once the otoliths were released, they were washed in 70% ethanol and dried at room temperature. Measurements were immediately taken of the otolith length (OL), width (OW), and otolith mass (OM) (Fig. 3) using a digital caliper with an accuracy of 0.01 mm. Total otolith mass (OM) was weighed using a standard analytical scale (HR-250AZ, A&D Company Ltd) to the nearest 0.0001 mg. The relationship between the otoliths' biometric features and the fish body's three lengths (TL, SL, and FL) was analysed through linear regression. The analysis was performed with the free Minitab 17 Statistical Software (2010), and the significance level adopted was p < 0.05. The SEM images of the otolith of C. kais and C. macrostomum were taken using the SEM microscope Zeiss Evo 50 available at Harran University Central Laboratory.

#### 3. Results

The biostatistics of the otolith biometric features are shown in Table 1. For C. kais, significant (p < 0.001) correlations were shown for OM – FL ( $R^2 = 0.5894$ ), OM – TL ( $R^2 = 0.4059$ ), and OM – SL ( $R^2 = 0.5360$ ). The lowest correlation was observed in the relationship OW – FL ( $R^2 = 0.3891$ ). For C. macrostomum, significant (p < 0.001) correlations were noticed in OL – FL (R<sup>2</sup> = 0.7280), OL – TL ( $R^2 = 0.7216$ ), and OL – SL ( $R^2 = 0.7001$ ). The lowest correlation ( $R^2 = 0.1717$ ) was obtained Muhammed Yaşar Dörtbudak, Laith A. Jawad, Hamza Yalçin, Joo Myun Park



Asteriscus otolith of **A**) *Cyprinion kais*, 179 mm TL; **B**) *C. macrostomum*, 190 mm TL. OL = otolith; OW = otolith width

for the relationship OM – TL. These results obtained from the current study provide the first relationships between otolith morphometric and total fish length for the Kais kingfish, *C. kais*, and the Tigris kingfish, *C. macrostomum*.

#### 4. Discussion

There needs to be more data on the relationship between otolith biometrics and the size of the fish of Cyprinid fish from Turkish inland waters. In the present investigation, the relationships between OL, OW, and OM with the total fish length for *C. kais* and *C. macrostomum* were performed for the first time (Table 1). Such an outcome provides standard data for trophic studies to determine their size.

The significant relation between the otolith biometrics and body size explains the effect of body growth on the otoliths of the fish examined (Munk 2012). This linear relation is dependent relative to the growth rate of each species (Campana & Casselman 1993, Mugiya & Tanaka 1992). Most investigators have shown close relationships between somatic and otolith growth (Giménez et al. 2016, Jawad et al. 2017, See et al. 2016). In addition, the linear relation can be adjusted for different size groups (Hare & Cowen 1995). Nevertheless, in the present work, using the three fish lengths of both species studied, the linear pattern best fits the relationship between body size and the otoliths in some biometrics (see results). This showed to have a very significant correlation (p < 0.001). Such meaningful relationships agree with the other investigations (Jawad & Al-Mamry 2012, Jawad et al. 2011). The coefficient of determination (R<sup>2</sup>) had higher values for the OM – FL ( $R^2 = 0.5894$ ), OM – TL ( $R^2 =$ 0.5843), and OM – SL (R<sup>2</sup> = 0.5360) in the case of C. kais and for the OL - FL (R<sup>2</sup> = 0.7280), OL - TL (R<sup>2</sup> = 0.7216), and OL - SL ( $R^2 = 0.7001$ ) in case of C. macrostomum. Consequently, we propose utilising otolith mass and otolith length to calculate the body size of C. kais and C. macrostomum, respectively. Nevertheless, the straight assessment of fish weight using otolith length or otolith mass prevents the introduction of additional errors due to using more than one regression model.

The reason behind developing more than one equation that links the fish's size with the otolith's biometric features is the occurrence of damage on the margins of the otoliths, as shown by several studies (de Assis et al. 2018). Even though the determination coefficient estimates were well represented in the results, the equations produced should be used cautiously. The shape and size of otoliths are affected by genetic, ontogenetic, and environmental characteristics (Campana & Casselman 1993). The shape is genetically affected, and the size relies on temperature and the existence of carbonate in the water (Checkley et al. 2009, Lombarte & Lleonart 1993), which can cause variations in the relation between otoliths and body size in populations from different locations. A further crucial aspect is the disclosure of chemicals in the digestive system of the predators, which can influence assessing prey size due to degradation to the surface of the otolith produced by stomach acids (Granadeiro & Silva 2000, Jobling & Breiby 1986). Contemplating these problems to lessen errors, we suggest using the equations generated in the present study in the Turkish waters, given that temperature influences the formation of otoliths, and to look for the existence of degradation from digestion. Nevertheless, the results show that

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#### Table 1

Data analysis of *Cyprinion kais* and *C. macrostomum* asteriscus otolith collected from Tigris River, Şırnak Province, Türkiye. R2, coefficient of determination; SD, Standard deviation

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Parameter/Species	Range	Mean (± SD)	Equation	R <sup>2</sup>
Cyprinion kais				
Otolith length (mm)	8.75 – 11.35	10.05 (1.23)	OL = 6 + 78.2 TL	0.4330
			OL = 180 + 81.0 SL	0.3406
			OL = 33 + 87.7 FL	0.3963
Otolith width (mm)	8.01 - 10.35	9.18 (1.02)	OW = 328 + 43.6 TL	0.4059
			OW = 450 + 42.7 SL	0.2861
			OW = 329 + 50.0 FL	0.3891
Otolith mass (g)	8-24	16 (0.98)	OM = -33.1 + 3.7 TL	0.4059
			OM = -28.1 + 4.1 SL	0.5360
			OM = -34.17 + 4.3 FL	0.5894
C. macrostomum				
Otolith length (mm)	9.68 - 14.69	12.19 (0.11)	OL = 260 + 63.4 TL	0.7216
			OL = 219 + 74.9 SL	0.7001
			OL = 156 + 72.9 FL	0.7280
Otolith width (mm)	8.61 - 12.35	10.48 (0.13)	OW = 316 + 43.5 TL	0.6461
			OW = 366 + 50.7 SL	0.6093
			OW = 305 + 50.5 FL	0.6663
Otolith mass (g)	11 - 33	22 (0.97)	OM = 1.45 + 1.9 TL	0.1717
			OM = 1.66 + 1.6 SL	0.1828
			OM = -1.75 + 1.7 FL	0.2257

estimates of the species' body size using biometric analyses of otoliths are reliable.

Nonetheless, the taxonomic value of the otolith of *C. kais and C. macrostomum* is not the subject of the present investigation; it is necessary to reveal that the calculation of the relationship between fish and otolith biometrics could be pondered as another taxonomic aid that can be added to those formerly set for fish otolith (Battaglia et al. 2010). Additionally, the relationship of fish size-otolith biometrics of these two species was not earlier investigated in Turkey. Consequently, this research increases the amount of data for these species and Turkish waters, which will be valuable in understanding marine trophodynamics in the area (Zan et al. 2015).

# 5. Conclusion

The investigation provides various provisions for the biometric relationships between asteriscus otolith biometrics and total fish length for Cyprinid fish caught in inland waters in Türkiye. Significant relationships between otolith mass and the three fish lengths (TL, SL, and FL) were attained for *C. kais*. Similar associations were obtained for OL and the three fish lengths of *C. macrostomum*. The outcome of the analysis shows that evaluations of the body size of the two fish species investigated through biometric studies of otoliths are trustworthy. This study will promote future dietary investigation, paleontological studies, and regional population dynamics.

#### **Conflicts of interest**

The authors declare that there is no conflict of interest.

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