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RESEARCH ARTICLE **ARAŞTIRMA MAKALESİ**

Spatial variation in relationships of otolith measurements with body length of Prussian carp, *Carassius gibelio* (Bloch, 1782) collected from four lentic habitats in Samsun Province, Türkiye

Samsun İli (Türkiye)'ndeki dört lentik habitattan örneklenen gümüşi havuz balığının (*Carassius gibelio* (Bloch, 1782)) vücut uzunluğu ile otolit ölçümlerinin ilişkilerinde alansal varyasyon

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Abstract: This study aimed to disclose the spatial variability in the lagenar otolith (asteriscus) dimensions-total length relationships generated for four Prussian carp, *Carassius gibelio* (Bloch, 1782) populations (Altınkaya Dam Lake, Bafra Fish Lakes, Lake Ladik and Lake Simenit) in Samsun Province, Türkiye. Relationship between length and weight of the fish was also described for each population. Samples were obtained from commercial fishermen in different periods between February 2017 and March 2018. The length, height, and weight of each otolith were determined. A non-linear function was used to define allometric relationships between asteriscus measurements and body size. The slopes of the regressions acquired for the right and left otoliths at each sampling site did not show any significant difference. All of the relationships were found to be statistically significant (P<0.001). The r^2 values ranged from 0.61 to 0.95, and the mean values of the percent prediction error varied from 3.10 to 7.45. The ANCOVA test determined significant spatial differences in regression slopes for the three otolith variables. Our findings revealed that otolith development reflected somatic growth, but this varied by sampling area.

Keywords: Otolith biometry, asteriscus, length-weight relationship, *Carassius gibelio*

Öz: Bu çalışma, Samsun İli (Türkiye)'ndeki dört gümüşi havuz balığı, *Carassius gibelio* (Bloch, 1782) popülasyonu (Altınkaya Baraj Gölü, Bafra Balık Gölleri, Ladik Gölü ve Simenit Gölü) için elde edilen lagenar otolit (asteriskus) boyutları-toplam uzunluk ilişkilerindeki alansal değişkenliği ortaya koymayı amaçlamıştır. Her popülasyon için boy ve ağırlık ilişkisi de tanımlanmıştır. Örnekler, Şubat 2017 ile Mart 2018 arasında farklı dönemlerde ticari balıkçılardan temin edilmiştir. Her bir otolitin uzunluğu, yüksekliği ve ağırlığı belirlenmiştir. Otolit ve balık büyümesi arasındaki allometrik ilişkileri tanımlamak için üssel bir fonksiyon kullanılmıştır. Her bir örnekleme alanında sağ ve sol otolitler için elde edilen regresyonların eğimleri önemli bir farklılık göstermemiştir. Tüm ilişkiler istatistiksel olarak önemli bulunmuştur (P<0,001). Belirleme katsayıları (r^2) 0,61 ile 0,95 ve yüzde tahmin hatasının ortalama değerleri 3,10 ile 7,45 arasında değişmiştir. ANCOVA testi, üç otolit değişkeni için regresyon eğimlerinde önemli alansal farklılıklar tespit etmiştir. Bulgularımız otolit gelişiminin somatik büyümeyi yansıttığını, ancak bu durumun örnekleme alanına göre değişiklik gösterdiğini ortaya koymuştur.

Anahtar kelimeler: Otolit biyometrisi, asteriskus, boy-ağırlık ilişkisi, *Carassius gibelio*

INTRODUCTION

Otoliths are white stones located in the inner ears of fishes (Tuset et al., 2008). They provide a sense of balance to fish and also aid in hearing (Popper and Fay, 1993). Teleosts have three pairs of otoliths, known as the sagitta, lapillus, and asteriscus. These calcified structures differ markedly in size. The sagittae are the largest pair in most teleost fishes, whereas the asterisci are bigger than other otolith types in Cypriniform fishes (Assis, 2003; Schulz-Mirbach and Reichenbacher, 2006). Otoliths also exhibit significant intra- and interspecific variability in shape (Wright et al., 2002; Vignon and Morat, 2010; Yedier and Bostancı, 2022). There is always a strong correlation between otolith development and fish length, as otoliths tend to grow as the fish grows. In this case, it is possible to estimate the length of the fish from which the otolith was acquired based on a measurement of otolith size (Campana, 2004). These estimates can be a reliable tool in studies on length-frequency distributions of fish populations, as well as in predator-prey studies with the aim to calculate the original size of consumed prey (Belfethi and Moulaï, 2022). The otolith-fish size relationships and their importance may vary between species or between different populations of the same species, as well as between different sizes of individuals of the same species (Bostancı et al., 2017; Saygın et al., 2017; Zengin Özpiçak et al., 2018; Emre, 2019; Saygın et al., 2020).

The Prussian carp, belongs to the Cyprinidae family, is regarded to be native from central-eastern Europe to Siberia (Perdikaris et al., 2012). It has been introduced outside of its natural range, into Europe, Asia and North America (Kottelat and Freyhof, 2007; Elgin et al., 2014). In Türkiye, the Prussian carp was first reported from Lake Gala in 1986 (Baran and Ongan, 1988). It has since spread to nearly all of Türkiye's lotic and lentic water bodies (Yerli et al., 2014; Tarkan et al., 2015; Çiçek et al., 2022). This species has become one of the most successful invasive fish beyond its natural distribution area due to its unique biological characteristics and extreme ecological tolerance (Tarkan et al., 2012; Fuad et al., 2021). The relationships between otolith parameters and body size of the Prussian carp were investigated in the Topçam Dam Lake (Bostancı, 2009) and Lake Ladik (Yılmaz et al., 2015) in Türkiye. However, no study has previously been carried out on otolith-fish size relationships of the Prussian carp inhabiting many freshwater environments in Samsun Province, northern Anatolia. Here, we examined whether there was a difference in otolith measurements-fish size relationships among the Prussian carp populations of four geographically distant lakes in Samsun Province, Türkiye. Additionally, the length-weight relationship of each population was also determined.

MATERIALS AND METHODS

The Prussian carp specimens were collected from four lentic ecosystems (Altınkaya Dam Lake, Bafra Fish Lakes, Lake Ladik, and Lake Simenit) in Samsun Province, Türkiye (Figure 1). The Altınkaya Dam Lake is 27 kilometers southwest of the Bafra district, on the Kızılırmak River. It has a surface area of 11830 hectares and a length of 70 kilometers (Yılmaz et al., 2007). The eastern part of the Kızılırmak Delta, which is located in the north of the Bafra district, is known as the Bafra Fish Lakes. It is made up of a series of different-sized lakes with lagoon characteristics (Gumus et al., 2007). Lake Ladik is 10 kilometers away from Ladik district. Its formation is tectonic, with a surface area of 1000 hectares and a maximum depth of 6 meters (Yılmaz et al., 2012). Lake Simenit is located within the Terme district's Gölyazı town. This lake with a surface area of 80 hectares and a maximum depth of 2 meters is the lagoon characteristic. It has a waterway on its south end that connects it to Lake Akgöl (Ersanlı and Gönülol, 2003).

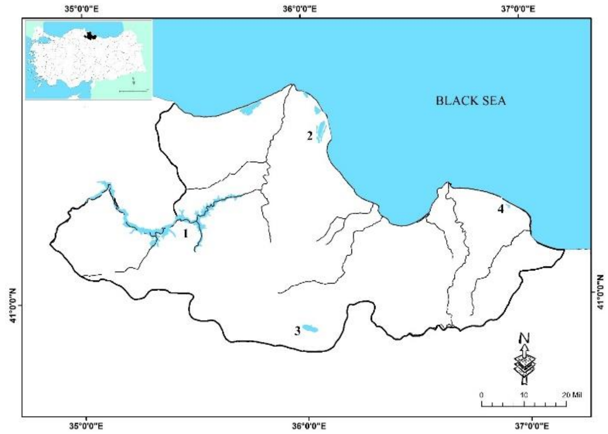

Figure 1. The sampling sites (1-Altınkaya Dam Lake, 2-Bafra Fish Lakes, 3-Lake Ladik, and 4-Lake Simenit)

Samplings were performed in different periods, from February 2017 to March 2018. Fish samples were obtained from commercial fishermen in these lakes. At the laboratory, fish body size (TL in cm), weight (W in g), and gender were determined and recorded. The lagenar otolith (asteriscus) pairs were removed, washed, dried, and stored in labeled plastic vials. Due to the low number of male individuals in all sampling areas, only female individuals were used in this study. The weight of each otolith (OW) was weighed with an accuracy of 0.01 mg. After that, the right and left otoliths were viewed under a binocular microscope (Leica S8APO) at 10x magnification. Two-dimensional digital images of the otoliths were taken with a digital camera (Leica DFC295). Otolith length (OL) and otolith height (OH) were measured with an accuracy of 0.001 mm using image analysis software (Leica Application Suite ver. 3.8). The OL and OH (Figure 2) were defined as the maximum distance between the anterior and posterior edges and between the dorsal and ventral borders, respectively (Yılmaz et al., 2015).

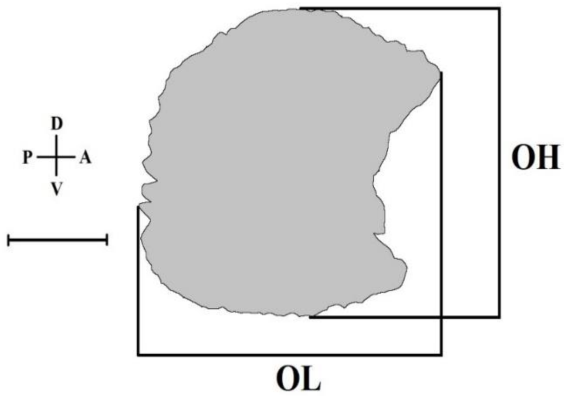

Figure 2. Distal view and measurement axes of the right asteriscus of Prussian carp. (OL-otolith length, OH-otolith height, D-dorsal, V-ventral, P-posterior, A-anterior, scale bar=2 mm)

The relationships of OL, OH, and OW versus TL were calculated using the simple non-linear regression model: $Y = aX^b$, where the coefficients *a* and *b* are constants of the regression (Lleonart et al., 2000). Three regressions were generated separately for the right and left otolith of each sampling site. Then, the slopes of the regressions were compared using the ANCOVA test (Zar, 1999). When no considerable differences were found, the H_0 hypothesis ($b_{right} = b_{left}$) was accepted and only one regression was presented for each variable by preferring the right otolith data. The *F*-test was used to confirm the regressions' significance. The deflection of each regression slope from isometry was controlled by a t-test (Zar, 1999). The statistical differences in regression slopes among the sampling areas were checked with the ANCOVA test (Zar, 1999).

The r^2 and the mean percent prediction error (PE%) were both used to evaluate the strength of each relationship. The mean PE% for a correlation is the average of the PE% values obtained for entire samples. The calculation of the PE% for an individual was performed by using the equation proposed by Smith (1980):

$$PE\% = \frac{|x_{observed} - x_{predicted}|}{x_{predicted}} \times 100$$

The dissimilarity between predicted and observed TL was analyzed for each measurement of the otolith by using a t-test or Mann-Whitney U test. The ANOVA test or Kruskal-Wallis test was used to compare the variations between mean PE% values of three parameters of the otolith at each sampling site. The variability in the mean PE% values of each otolith parameter among the sampling localities was also examined with the Kruskal-Wallis test (Zar, 1999).

The length-weight relationship (LWR) of female Prussian carp specimens from each sampling site was estimated through the formula $W = a\,TL^b$, where W is the body mass, TL is the body size, *a* and *b* are coefficients of the equation (Bagenal and Tesch, 1978). The *t*-test was used to see if the parameter *b* differed significantly from 3, indicating whether the growth was isometric ($b = 3$) or allometric ($b \neq 3$) (Zar, 1999).

RESULTS

A total of 288 female Prussian carp (61 from Altınkaya Dam Lake, 80 from Bafra Fish Lakes, 82 from Lake Ladik, and 65 from Lake Simenit) was used in this study. In the analysis of asteriscus dimensions against TL, no considerable differences were detected between the slopes of the relationships acquired for the right and left asteriscii in each sampling locality (ANCOVA test, TL-OL, F=0.289, P=0.592; TL-OH, F=0.717, P=0.399; TL-OW, F=0.133, P=0.716 for Altınkaya Dam Lake; TL-OL, F=0.302, P=0.584; TL-OH, F=0.006, P=0.937; TL-OW, F=0.004, P=0.948 for Bafra Fish Lakes; TL-OL, F=0.076, P=0.783; TL-OH, F=0.101, P=0.751; TL-OW, F=0.030, P=0.863 for Lake Ladik; TL-OL, F=0.270, P=0.604; TL-OH, F=0.358, P=0.551; TL-OW, F=0.004, P=0.952 for Lake Simenit). Therefore, the regressions generated from the right otoliths were chosen for the next analyses. The details of the dataset are summarized in Table 1.

Table 1. Descriptive statistics of Prussian carp specimens and their otoliths collected from four lentic habitats in Samsun Province, Türkiye (n, sample size; SD, standard deviation)

Locality	Measurement	*n*	Mean±SD	Range
	Fish Length (cm)	61	33.04±5.06	22.4–40.3
	Fish Weight (g)	61	703.66±288.95	193.02–1191.30
Altınkaya Dam Lake	Otolith Length (mm)	61	5.80±0.71	4.34–7.05
	Otolith Height (mm)	61	5.25±0.68	3.69–6.36
	Otolith Weight (mg)	61	36.04±12.26	12.98–58.08
	Fish Length (cm)	80	20.28±4.11	11.7–30.7
	Fish Weight (g)	80	179.64±105.83	16.69–520.51
Bafra Fish Lakes	Otolith Length (mm)	80	4.20±0.64	2.56–5.46
	Otolith Height (mm)	80	3.83±0.60	2.35–5.11
	Otolith Weight (mg)	80	14.36±5.87	3.61–14.36
	Fish Length (cm)	82	26.09±2.58	19.1–35.1
	Fish Weight (g)	82	379.56±131.46	136.77–859.77
Lake Ladik	Otolith Length (mm)	82	4.66±0.32	3.78–5.89
	Otolith Height (mm)	82	3.92±0.27	3.28–5.17
	Otolith Weight (mg)	82	16.11±3.20	9.82–33.24
	Fish Length (cm)	65	18.76±2.63	14.8–27.4
	Fish Weight (g)	65	131.42±58.52	67.01–342.24
Lake Simenit	Otolith Length (mm)	65	4.15±0.42	3.46–5.22
	Otolith Height (mm)	65	3.69±0.39	3.04–4.62
	Otolith Weight (mg)	65	13.74±3.68	7.62–22.84

All relationships were highly significant (P<0.001). The r^2 values varied from 0.61 to 0.95 and the regressions explained more than 70% of the variance in most cases (Table 2). The r^2 values of the regressions generated for the Altınkaya Dam Lake and Bafra Fish Lakes were higher than those of the other lakes. The otolith height, which accounted for 95% of the variability in Altnkaya Dam Lake, and the otolith weight, which accounted for 94% in Bafra Fish Lakes, were the measurements most strongly related to fish size. The otolith length was the best indicator of fish length for Lake Ladik and Lake Simenit, with 75% and 73% of the variability, respectively. The results of the ANCOVA test indicated significant differences among slopes of four sampling sites for TL-OL (F=5.017, P=0.002), TL-OH (F=8.124, P=0.000), and TL-OW (F=10.721, P=0.000) relationships. The slopes of three regressions were higher for the Altınkaya Dam Lake and Bafra Fish Lakes. Negative allometric growth was observed for two relationships in Altınkaya Dam Lake and Bafra Balık Lakes (*t*-test, TL-OL and TL-OH, P<0.001), and for three in Lake Ladik and Lake Simenit (*t*-test, TL-OL, TL-OH and TL-OW, P<0.001). However, the TL-OW relationship was positive allometric for the Altınkaya Dam Lake (*t*-test, P<0.005), and isometric for the Bafra Fish Lakes (*t*-test, P>0.05).

The mean PE% values ranged from 3.10 to 7.45 (Table 2). The values of the mean PE% of three otolith variables for the Altınkaya Dam Lake were lower than those of the other localities. No significant difference was determined between observed and predicted TL values in the Altınkaya Dam Lake (Mann-Whitney U test; OL, P=0.884; OH, P=0.965; OW, P=0.737), Bafra Fish Lakes (*t*-test; OL, P=0.961; OH, P=0.927; OW, P=0.886), Lake Ladik (Mann-Whitney U test; OL, P=0.914; OH, P=1.000; OW, P=0.908), and Lake Simenit (*t*-test; OL, P=0.949; OW, P=0.907; Mann-Whitney U test; OH, P=0.883). There were no significant differences in the mean PE% values of the otolith variables in all sampling sites (ANOVA test; F=1.05, P=0.352 for Altınkaya Dam Lake, Kruskal-Wallis test; H=0.629, P=0.730 for Bafra Fish Lake, and H=1.264, P=0.532 for Lake Simenit) except Lake Ladik (Kruskal-Wallis test; H=14.96, P=0.001). The Kruskal-Wallis test demonstrated that mean PE% values were different among sampling areas for OH (P=0.000) and OW (P=0.004), but not for OL (P=0.063).

Table 2. Estimated parameters of the relationships between otolith measurements and body length, and the mean percent prediction error value of each otolith variable of the Prussian carp collected from four sampling sites in Samsun Province, Türkiye (r^2, coefficient of determination; PE, prediction error)

Locality	Relationship	r^2	Observed TL (Mean±SD)	Predicted TL (Mean±SD)	PE% (Mean±SD)
	$OL=0.404TL^{0.762}$	0.93		33.15±5.30	3.72±2.53
Altınkaya Dam Lake	$OH=0.312TL^{0.808}$	0.95	34.04±5.06	33.03±5.32	3.10±2.28
	$OW=0.015TL^{2.210}$	0.94		33.35±5.39	3.35±2.27
	$OL=0.447TL^{0.745}$	0.93		20.31±4.14	4.39±3.78
Bafra Fish Lakes	$OH=0.402TL^{0.750}$	0.92	20.28±4.11	20.34±4.25	4.63±3.70
	$OW=0.033TL^{2.008}$	0.94		20.18±4.17	4.35±2.95
	$OL=0.667TL^{0.597}$	0.75		26.05±3.00	4.48±3.35
Lake Ladik	$OH=0.671TL^{0.542}$	0.61	26.09±2.58	26.12±3.46	6.74±4.11
	$OW=0.083TL^{1.611}$	0.74		26.21±3.10	4.81±3.05
	$OL=0.657TL^{0.630}$	0.73		18.78±3.05	6.31±5.21
Simenit Lake	$OH=0.591TL^{0.625}$	0.68	18.76±2.63	18.85±3.18	7.33±5.57
	$OW=0.151TL^{0.533}$	0.64		18.81±3.24	7.45±7.24

The length-weight relationship for female individuals of the Prussian carp was obtained as $W=0.012\ TL^{3.125}$ in the Altınkaya Dam Lake (*n*=61, SE (*b*)=0.048, r^2=0.97), $W=0.021\ TL^{2.975}$ in the Bafra Fish Lakes (*n*=80, SE (*b*)=0.065, r^2=0.96), $W=0.011\ TL^{3.200}$ in Lake Ladik (*n*=82, SE (*b*)=0.083, r^2=0.95), and $W=0.027\ TL^{2.876}$ in Lake Simenit (*n*=65, SE (*b*)=0.065, r^2=0.97). Positive allometric growth was detected in the Altınkaya Dam Lake (*t*-test, P=0.012) and Lake Ladik (*t*-test, P=0.018), whereas isometric growth was observed in the Bafra Fish Lakes (*t*-test, P=0.690) and Lake Simenit (*t*-test, P=0.061).

DISCUSSION

The results of our study indicated no statistical difference between the biometric relationships generated separately for the right and left otoliths. Therefore, the size of the Prussian carp can be back calculated using only the right or left otolith measurements. The literature reports that remarkable differences between right and left asteriscus parameters are mostly not found for many members of the family Cyprinidae (Saygın et al., 2017; Zengin Özpiçak et al., 2018; Emre, 2019; Saygın et al., 2020).

In contrast, Kontas and Bostanci (2015) detected the asymmetry between right and left asteriscus measurements of *Barbus tauricus* captured from the lower basin of the Melet River, Türkiye. Also, Jawad and Mahé (2022) indicated that there was a significant effect of the head side on the relationship of each variable of the lagenar otolith with the body length of common carp (*Cyprinus carpio*) sampled from three Iraqi rivers.

In this study, we detected the spatial differences in relationships between asteriscus descriptors and body length of *C. gibelio*. Similar observations have also been reported for many fish species (Tombari et al., 2011; Bostancı et al., 2017; Jawad et al., 2017; Saygın et al., 2017; Nguyen and Dinh, 2020; Dinh et al., 2021). These variations may mainly be due to differences in the abiotic factors (e.g., temperature, pH, salinity, diet, etc.) of the sampling sites. According to Campana (1990), fish-otolith regressions often differ among populations or fish species with varying development rates.

The present study suggested that the allometric relationships between asteriscus variables and fish growth are reliable for the original somatic size estimates of the Prussian carp due to their high r^2 and low mean percent prediction error values. However, the best indicator of fish size differed among populations studied. The OL was the best predictor of fish length for Lake Ladik and Lake Simenit populations, whereas the OH and OW were the best predictors of fish somatic growth for Altınkaya Dam Lake and Bafra Fish Lakes populations, respectively. Similar results have also been obtained in previous works on otolith biometry of some other cyprinid fishes. Saygın et al. (2017) reported that the best indicator for estimating the body size of *Alburnus tarichi* is otolith length in Lake Erçek and Lake Aygır, and otolith height in Lake Van and Lake Nazik. Zengin Özpiçak et al. (2018) found that asteriscus otolith weight for Terme Stream and Yedikır Dam Lake populations, and asteriscus otolith length for the populations of Abdal and Akçay streams of *Squalius cephalus* are better indicators of the fish size than other measurements of otoliths. Similarly, when the relationships between lagenar otolith dimensions and body size of *Barbus tauricus* caught from five streams in the Black Sea region of Türkiye were analyzed, the best fit was found between total length and otolith height in Akçay, Engiz, Karadere, and Değirmenağzı streams, and between total length and otolith height in Terme Stream (Ozpicak, 2020). In previous works regarding the species studied, the otolith weight in Topçam Dam Lake (Bostancı, 2009) and the otolith height in Lake Ladik (Yılmaz et al., 2015) were shown to be the variables most closely associated with fish size.

From the otolith measurements-fish size regressions established in this study, the body length of the Prussian carp can be reconstructed but not its somatic weight. For this reason, the length-weight relationship is also given for each population of the fish in question. The relationship between size and body weight of Prussian carp was previously studied in Bafra Fish Lakes (Bostancı et al., 2007) and Lake Ladik (Yazıcıoğlu et al., 2013). This research provides the first information on the length-weight relationship of Altınkaya Dam Lake and Lake Simenit populations.

In conclusion, our study confirmed that the lagenar otolith growth of the Prussian carp reflected its somatic development, but that differed among populations. It is advised that all regressions from the current study be used within the fish size and weight limits presented in Table 1. The outcomes of this study can be used to provide the information about size and mass of this prey fish in future studies on the trophic ecology of the potential predators of Prussian carp.

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AUTHORSHIP CONTRIBUTIONS

All authors contributed equally to the idea, data collections, design and writing of the manuscript.

CONFLICTS OF INTEREST

The author declares that there is no conflict of interest on this manuscript.

ETHICS APPROVAL

No specific ethical approval was necessary for this study.

DATA AVAILABILITY

The data that support the findings of this study are available from the corresponding author upon reasonable request.

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