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Menba Kastamonu Üniversitesi Su Ürünleri Fakültesi Dergisi 2022; 8(2): 83-93 Araştırma Makalesi/Research Article

The Relationships Between Fish Size and Otolith Dimensions Between Sexes in the Grey Wrasse, *Symphodus cinereus* (Bonnaterre, 1788)

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Abstract

In this study, the morphology of the sagittal otoliths of *Symphodus cinereus*, which exhibits an alternative reproductive tactic, was compared between the sexes. Fish specimens were collected using trammel net during the period June 2015 and May 2016 from waters around Rize City at the Black Sea coasts. The otolith weights (OWe), eyed and blind side otolith lengths (OL) and otolith width (OW) of each specimen were measured at the scale of nearest 0.0001g and 0.001 mm, respectively. A total of 119 sample fish (48 males and 71 females) were collected to examine. It was recorded that total length and weight ranged from 10.2 to 14.4 cm and 18.51 to 68.23 g for females and 10 to 16.4 cm and 17.14 to 70.29 g for males. It was found that the coefficients of determination between otolith weight and total length is R^2 = 0.725 whereas otolith weight and the total length for sexes combined is R^2 = 0.715. It was also demonstrated a strong positive relationship between the total length- otolith dimensions (OL/TL, OW/TL, Owe/TL). We determined the differences in otolith length, width and weight between male and female sagittal otoliths. According to these results, female sagittal otoliths tend to be larger than males ($p<0.05$). The results of this study are important as they reveal the otolith differences of *Symphodus cinereus* between the sexes.

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INTRODUCTION

Teleost fish have three semicircular canals arranged perpendicular to each other. These channels open into nested chambers or otic sacs (Wright et al., 2002). Teleost fish have three couplesotoliths, three on each side; sagitta, asteriskus, lapillus (Ekingen 1983). Otoliths are consisting of calcium carbonate and are found in the inner ears of bony fish and also function in hearing and balance (Campana, 2004). When the sagittal otolith varies less within the species, it is a very important otolith because it shows significant morphological differences between species (Campana, 2004). Therefore, trait of the sagittal otolith such as shape and size are used to distinguish fish species and higher order taxonomic groups (Paxton, 2000; Tuset et al., 2003, 2015). Otolith morphology in many different fields of fish biology; anatomy of fish species, taxonomic revisions of fish taxa, determination of phylogenetic relationships, ecomorphology studies, determination of the relationships among fish growth and otolith growth and, determination of similarities between fish that are fossil and the growth of fish living today (Bostancı et al. 2012). It is significantly important to know relationship between fish length and sagittal otolith length to have a considerable information on determining fish length from otoliths in stomach of predators and understand prey predator relationships (Granadeiro & Silva 2000, Battaglia et al. 2010, Kasapoglu & Duzgunes 2013). The morphological variant of the sagittal otolith may be affected by age, environmental and genetic factors (Cardinale et al., 2004; Vignon & Morat, 2010), however, it may vary according to its somatic growth ratio, habitat and is also related to the individual's nutrition (Lombarte & Lleonart, 1993; Strelcheck et al., 2003; Gagliano & McCormick, 2004). A 20 species of Labridae (Wrasse), which has 504 species in the world (Parenti & Randall 2011), live on the coasts of Turkey (Bilecenoğlu et al. 2014) and 8 in the Black Sea (Keskin 2010). As a territorial marine fish, Labridae are described as small and inhabit in rocky and algal inshore areas (Costello 1991). Grey wrasse (*Symphodus cinereus* (Bonnaterre, 1788) is a species belonging to the genus *Symphodus.* It shows distribution in Mediterranean Sea and from Gibraltar to Arcachon basin in Eastern Atlantic. It lives in eel grass beds in coastal areas and sometimes in soft bottoms between 1-20 m. This species is usually found in lagoons and estuaries with plenty of cover. It feed on shrimps, amphipods, isopods, gastropods and bivalves (Quignard & Pras 1986). There are different types of otolith research in the literature. However, knowledge on some aspects of otolith morphology is still limited.

The studies of intra-sex and between-sex variations in otolith morphology in species with alternative reproductive tactics are limited in the literature. Alternative reproductive tactics (ART) occur in case of the individuals of any sex species, refers at least two different reproductive morphs, as well as each morph try to achieve reproduction by alternative means. (Gross, 1996; Taborsky et al., 2008; Neff & Svensson, 2013). In most species with ARTs, males do not show a continuity for distribution of phenotypic traits, which brings about two or more reproductive morphs together within a population that are behaviorally, physiologically and morphologically distinct from one another. It is known that otolith morphology differs in species, adopting alternative reproduction (Bose et al. 2017). Accordingly, ARTs could be a critical source of intraspecific variation in otolith morphology that could provide further investigation. *S. cinereus* is one of these different alternative reproductive species. Males of *S. cinereus* was reported to nest with algae on sandy-muddy base on the coast of France (Thau lagoon) (Quignard, 1962). Subsequently, males were reported to guard and ventilate the nest (Lejeune, 1985). *S. cinereus* of males have three different phenotypes regarding their reproductive behavior: territorial male, satellite males and sneakers (Lejeune and Voss, 1980; Michel and Voss, 1982; Lejeune, 1985). So, there is the possibility of an ART for males (Lejeune, 1985). Bose et al (2017) suggested that examining the otoliths of fish species exhibiting different reproductive tactics may provide two benefits. First, it provides an opportunity to use study systems that identify differences in physiology, behavior, and life history that cause phenotypic changes in sagittal otolith morphology, which is a species-specific and limited feature Second, it tests the importance of considering alternative reproduction tactics when examining the population structure of species with economic value (Bose et al., 2017). In this study, the relationships between fish length and otolith length, width and weight of *S. cinereus* species were investigated between sagittal otoliths for female, male and all individuals. Considering the different reproductive tactics in *S. cinereus* males, it was hypothesized that the sagitta morphology might be different.

MATERIAL AND METHODS

Fish specimens (N=119) were collected by using trammel net during the period of June 2015 and May 2016 from waters around Rize City at the Black Sea coasts (41° 3'58.23"N 40°38'20.93"E) (Figure 1). All sampled grey wrasse was taken to the laboratory for other analysis. For each fish, the weight (W) (±0.0001 g) and total length (TL) (±0.1 mm) were determined. After that, sagittal otolith pairs were carefully removed and then properly cleaned. Otolith pairs were stored dry for further examination. After all otoliths were measured without distinguishing right and left otoliths. It was determined the sex was through macroscopic examination of the gonads (71♀, 48♂). The width and length of the otoliths were measured with the Nikon SMZ1000 stereomicroscope, with Nikon SMZ1000 digital camera was connected (magnification from x0.8 to x8.0) imaging system (Figure 2). The description of otolith length was the longest axis between posterior and anterior otolith edge. The width of otolith was described as the distance from dorsal to ventral edge taken perpendicular to the length through the otolith focus. The weights of the otoliths were weighed in Shimadzu ATX-224 brand precision scales with 0.0001 mg sensitivity. Relationships between otolith dimensions and fish length and weight were calculated according to the equation y=ax + b (Le Cren, 1951, Froese, 2006; (a=point of the y-axis of the line, b=slope of the line). The deviations from the expected 1:1 sex ratio were determined using chi-square (χ^2) test. Total lengths of male and female fish were compared with the t-test. In addition, statistical test (ANOVA) was applied to determine whether the regression was statistically significant. The ratio of otolith length to total length was calculated for each fish. Thus, the effect of different length frequencies of male and female otoliths was eliminated. Using these data, the Mann-Whitney U test was performed to test the statistical difference in otolith lengths between the sexes.

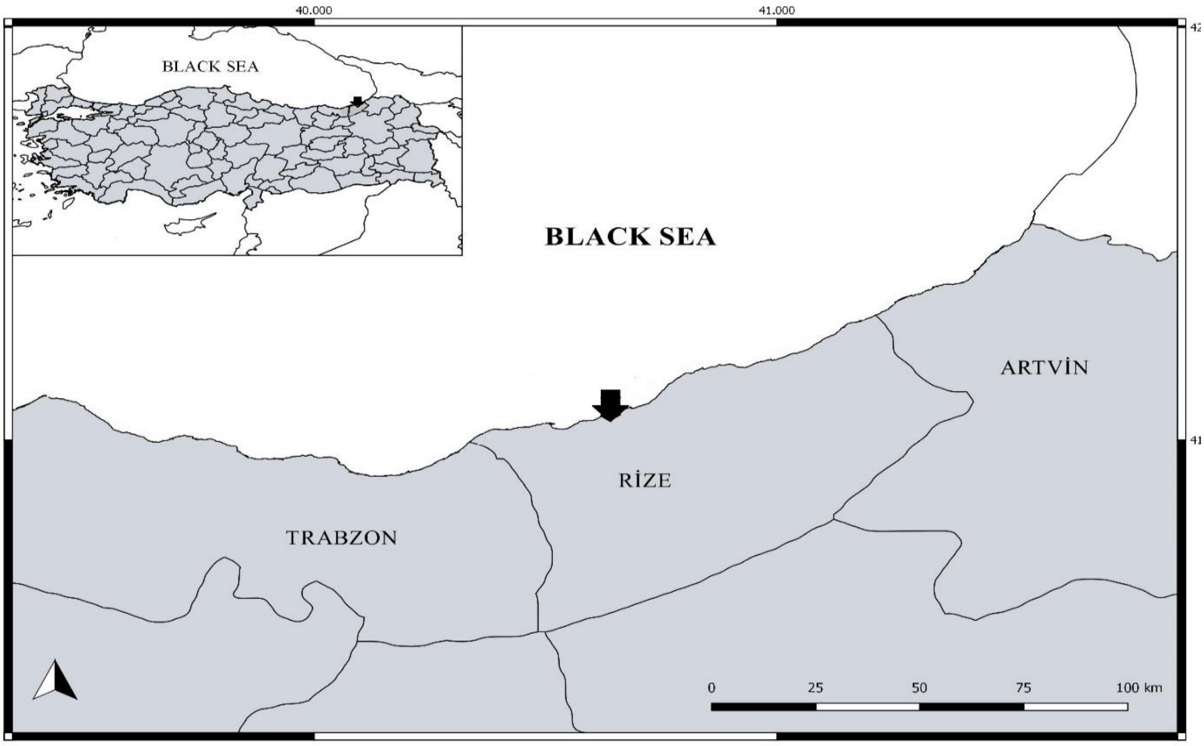

Figure 1. Study area in the Black Sea.

Figure 2. Typical images of *Symphodus cinereus* otoliths and the position of fix-points used to measure length and width under the microscope (OL was the longest axis between posterior and anterior otolith edge. OW was described as the distance from dorsal to ventral edge).

RESULTS

A total of 119 (71♀, 48♂) fish otoliths were examined throughout the study. The morphological data on weight and length for females, males, and both sexes combined are given in Table 1. The ratio of females and males of the entire sample was 1/1.47. This ratio was statistically differing significantly form the expected 1:1 ratio between sexes ($\chi 2$=8.036; $p < 0.05$). The difference in the values of the total length of female and male is statistically significant. ($t_{d.f.}$:118 =2,79, p < 0.05). The mean OL/TL value of female individuals (2.58±0.215) is greater than the OL/TL value of male individuals (2.47±0.266). The difference in the OL/TL values between the sex groups is greater than would be expected by chance, thus there is a statistically significant difference (T=5219.5, p<0.001). Regression relationship parameters and coefficients between fish length and weight with otolith dimensions and statistical test results by sex are given in Table 2. In the linear regression model, the coefficients of (R^2) were determined between 0.587 and 0.831. The regressions between total length and otolith width in both sex and total weight and otolith weight in males were not statistically significant (Table 2). The determined coefficient was R^2 = 0.72 between total length and otolith length and for total length and otolith weight, it was found as R^2 = 0.72 for female (Figure 3). The determined coefficient was R^2 = 0.731 between total length and otolith length, whereas it was R^2 = 0.747 between total length and otolith weight for male (Figure 4). The correlation between total length and otolith width was weak for both sexes (Figure 3, 4). The results obtained from this study indicated that otolith dimensions increase when the total length increase and it can be made correlation between otolith growth and growth of *S. cinereus*. The determined coefficient between fish weight and otolith weight was found R^2 = 0.664 whereas, the coefficient between total length and otolith weight was R^2 = 0.715 for overall (Figure 5). It was seen a moderate relationship between otolith weight and fish weight, and also between total length and otolith weight. When the male and female sagittal otoliths of *S. cinereus* were compared, the difference between the length, width and weights of the otoliths was found to be significant according to the Mann-Whitney U test results (Table 3).

Table 1. Morphometric measurements of otoliths by sex.

	OL (mm)	OW (mm)	Owe (g)	TL (cm)	W (g)
		♀			
Min	2.428	1.413	0.0007	10.2	18.51
Max	4.461	2.526	0.004	15.4	68.23
Mean	3.342	1.895	0.002	12.8-	37.92
SD	0.498	0.237	0.001	0.14	1.39
		♂			
Min	1.966	1.347	0.0007	10.0	17.40
Max	4.554	2.318	0.0033	16.4	70.29
Mean	3.122	1.746	0.002	12.5-	35.71
SD	0.595	0.231	0.001	0.15	1.51
		♀+♂			
Min	1.966	1.347	0.0007	10.0	17.14
Max	4.554	2.526	0.004	16.4	70.29
Mean	3.254	1.836	0.002	12.7	37.03
SD	0.548	0.245	0.001	0.12	1.15

OL: otolith length, OW: otolith width, Owe: otolith weight, TL: total length, W: total weight

Table 2. Regression relationship parameters and coefficients between fish length and weight with otolith dimensions and statistical test results by sex.

		Coefficient				Linear regression		
Sex	Variable	*a*	P(0.001)	*b*	P<0.001	R^2	F	P(0.001)
Female	TL&OL	-0.873	<	0.327	<	0.720	270.4	<
	TL&OW	-0.0003	>	0.147	<	0.645	190.8	<
	TL&OWe	-0.0037	<	0.000444	<	0.722	273.1	<
	TW&OWe	0.00042	<	0.00004	<	0.646	191.2	<
	OL&OWe	-0.00204	<	0.00123	<	0.819	475.1	<
Male	TL&OL	-1.276	<	0.350	<	0.731	187.8	<
	TL&OW	0.0626	>	0.134	<	0.711	170.1	<
	TL&OWe	-0.00308	<	0.000379	<	0.747	203.7	<
	TW&OWe	0.0002	>	0.000041	<	0.739	195.5	<
	OL&OWe	-0.00137	<	0.000975	<	0.831	339.2	<
Overall	TL&OL	-1.111	<	0.342	<	0.726	464.8	<
	TL&OW	-0.0644	>	0.149	<	0.587	249.9	<
	TL&OWe	-0.0035	<	0.000425	<	0.715	442.6	<
	TW&We	0.000297	<	0.000043	<	0.664	348.1	<
	OL&OWe	-0.00177	<	0.00113	<	0.817	785.7	<

OL: otolith length, OW: otolith width, Owe: otolith weight, TL: total length, W: total weight, TW: total weight

Table 3. Mann-Whitney U test results of otolith length relationships between sexes

Sex	♀		♂		Statistical test
	Mean	SD	Mean	SD	
TL/OL	3.892061	0.317712	4.089408	0.439954	*U*=2681.5 p<0.001
TL/W	6.768921	0.842388	7.228125	0.534509	*U*=2163.5 p<0.001
TL/Owe	6846.02	2033.619	8259.489	2254.166	*U*=2296.5 p<0.001

Table 4. *a,b* and R^2 values of Grey wrasse individuals compared with other studies.

S. cinereus	Location	N	Fish total length (cm)	Fish length			
				Otolith measurements	*a*	*b*	R^2
Škeljo and Ferri, 2011	Adriatic Sea	127	6.0-10.3	Length	1.83	0.66	57.3%
				Width	1.97	0.61	32.4%
				Weight	2.68	0.26	52.4%
This study	Black Sea	119	10-16.4	Length	0.34	1.11	0.72
				Width	0.14	0.06	0.58
				Weight	0.00	0.00	0.71

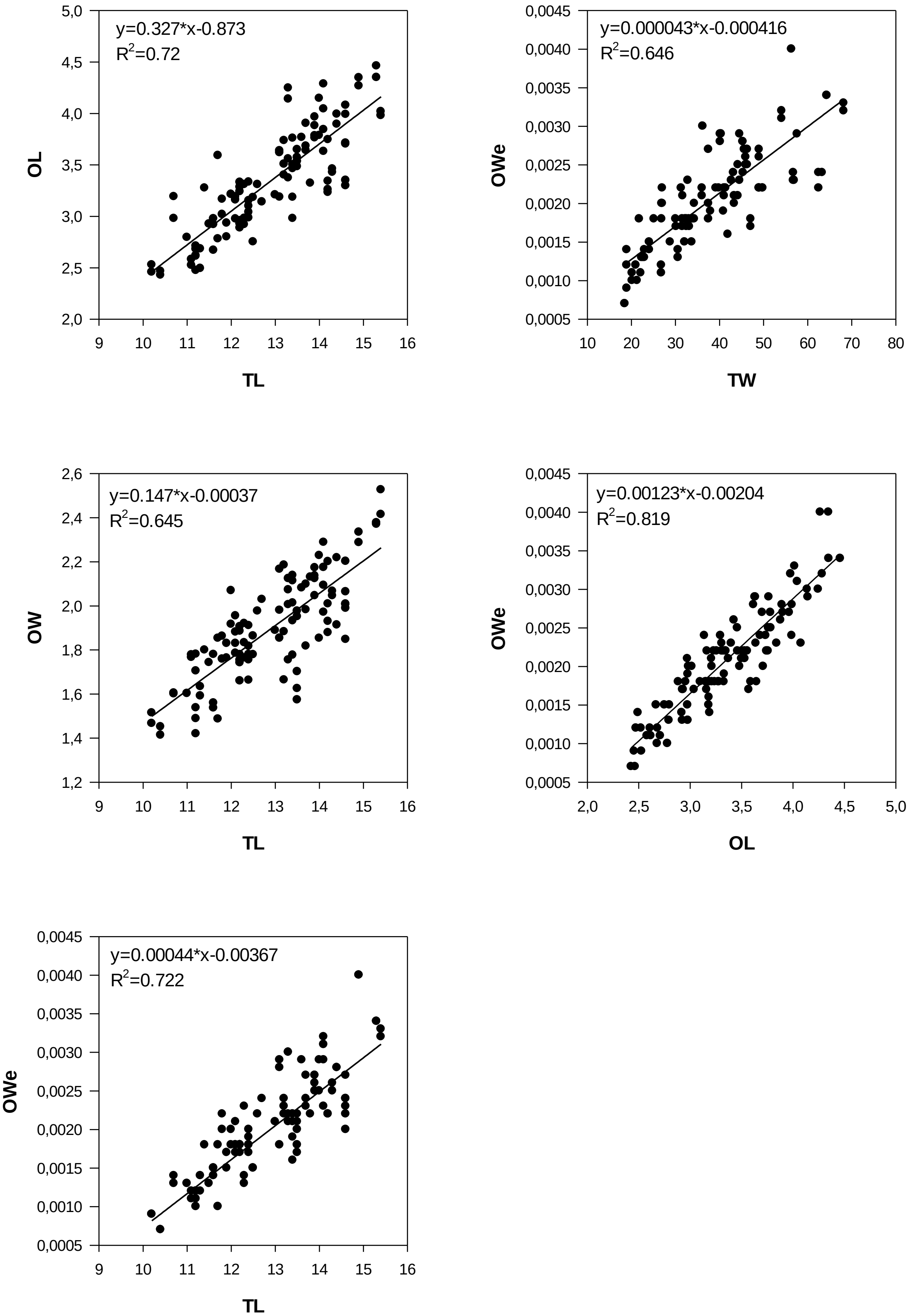

Figure 3. Relationships of otolith dimensions and weight with total length and body weight of female fish.

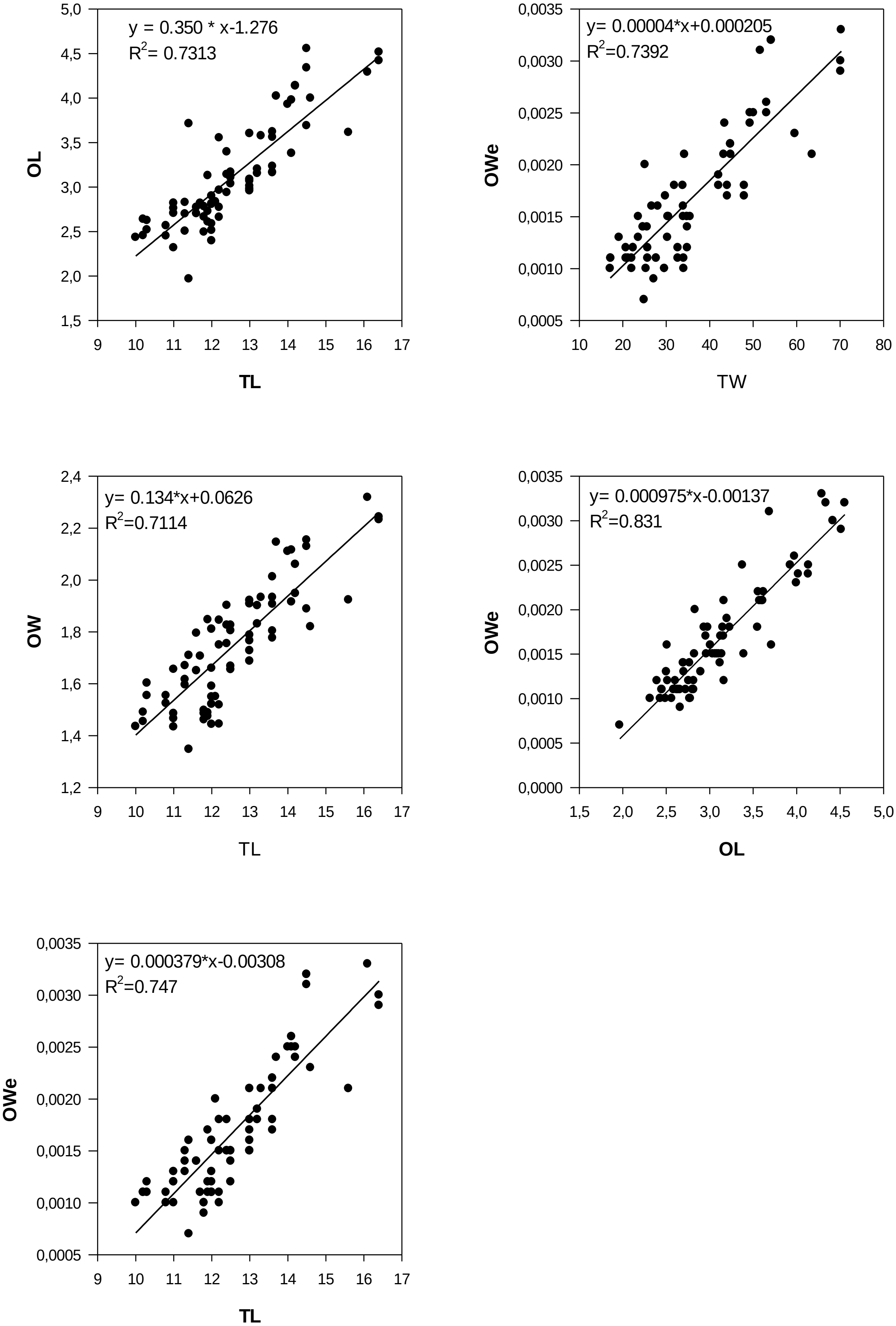

Figure 4. Relationships of otolith dimensions and weight with total length and body weight of male fish.

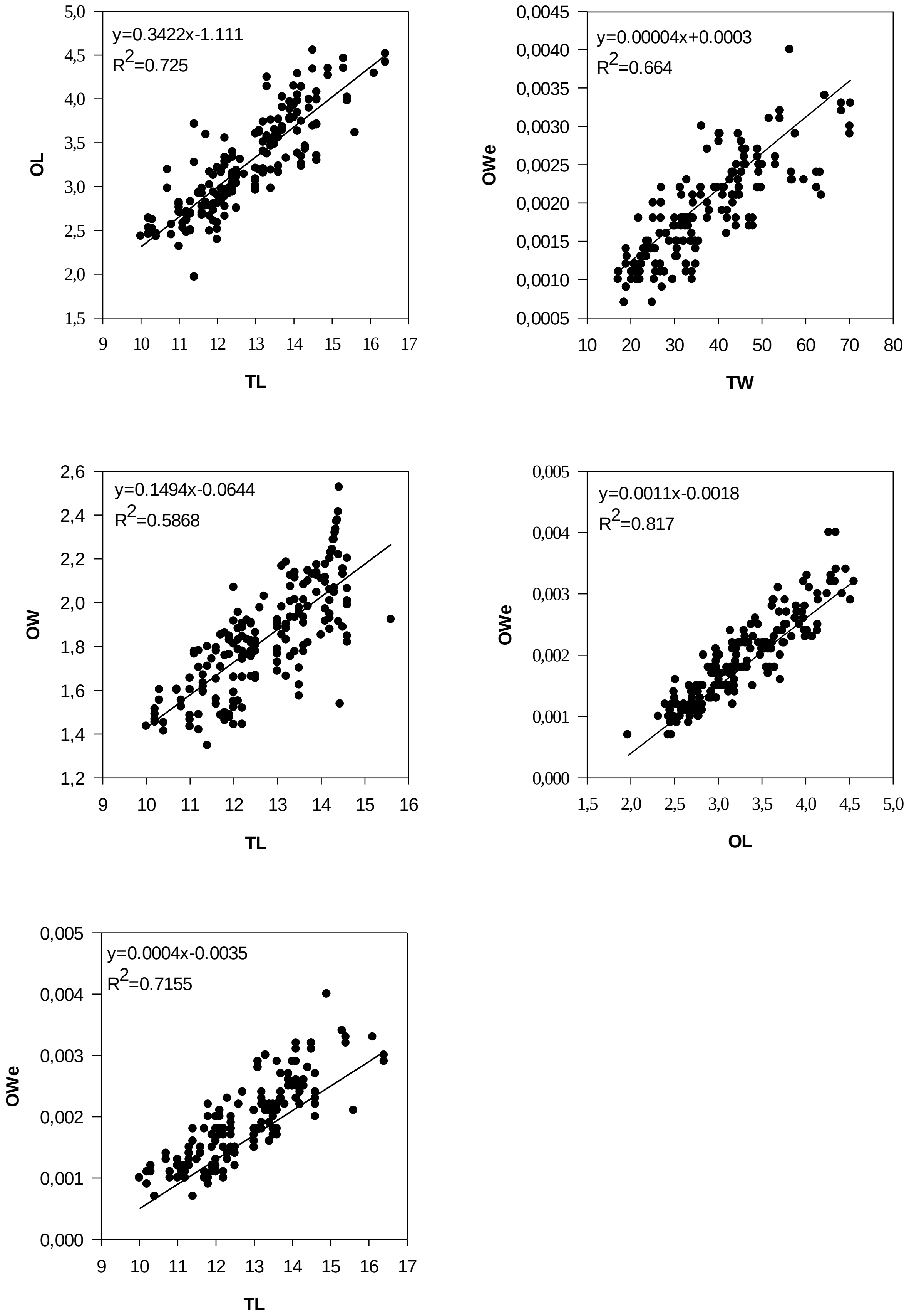

Figure 5. Relationships of otolith dimensions and weight with total length and body weight of overall individuals.

DISCUSSION

It is little known about the biology and otolith morphology of *S. cinereus* living in Black Sea waters. The cause of difficulty in collecting data on this species is absence of a fishery targeted this fish because of the limited commercial value. That is why, the information is available only through bycatch from fishery and from scientific expeditions (Bal 2014). Śkeljo & Ferri (2012) investigated the otolith morphology of *S. cinereus* and five wrasse species in their study. According to their results in *S. cinereus* species, a strong correlation was found between fish length and fish weight and otolith length and otolith weight, while a weak correlation was found with otolith width. Although, different fish size ranges were examined in our study with this mentioned study, similar results were obtained (Table 4). Śkeljo & Ferri (2012) found that otolith length and otolith width values which were as a result of morphometric measurements in their study in the Eastern Adriatic, were lower than the otolith length and otolith width values obtained in this study. The environmental factors, especially temperature, are effective in otolith growth, so low temperatures slow down the otolith growth and affect the physiological process of material deposition on otoliths (Morales-Nin 1987). In this study, the length, weight and width of female and male otoliths were compared and it was determined that female otoliths tended to be larger than male otoliths. Bose et al. (2020) determined that the sagittal otoliths of female and male individuals in African cichlids are different and that the otoliths of females are longer than the otoliths of males. In the literature, there are studies examining the shape and size of the sagittal otolith between two stocks. For example, Bose et al. (2017) found no difference in shape between the sagittal otoliths of two different geographic populations of *Porichthys notatus*, while Campana and Casselman (1993) found differences in sagittal shape among *Gadus morhua* stocks and reported that this may be related to the growth rates between populations. The otolith variability shown is in accordance with genetic (population and stocks) and environmental (temperature) effects on otolith growth observed by Lombarte & Lleonart (1993) and Torres et al. (1999). In this study, TL/OL, TL/W, TL/OWe between the two sexes was significant. In the study revealing the difference of otoliths of *Porichthys notatus* species that show alternative reproductive tactics. Bose et al. (2017), reported that sagitta shape also differed between females and males of the conventional guarder tactic. Differences in the otolith shape between the sexes was reported in some studies in the literature (Campana & Casselman, 1993; Carvalho et al. 2020; Başusta and Khan, 2021). Differences in otolith shape between the sexes may be related to the physiology, somatic growth rate and physiology of the sexes. (Campana & Casselman, 1993; Cardinale et al., 2004). Additionally, some factors such as sex-specific hormone levels, unequal growth rates, and distinct habitat usage for male and female fish are also effective to facilitate regulation of consequent otolith shape which can be a result in significant measure of sexual dimorphism. (Tuset et al. 2015, Tuset et al. 2016, Parmentier et al. 2018, Vaux et al. 2019).

As a result, this study revealed the relationship between fish size and otolith size of *S. cinereus* and determined the difference in size of female and male otoliths. In this study, the length, width and weight relationships of sagittal otoliths are discussed. It is recommended to carry out studies comparing the sagittal otoliths of male phenotype forms of *S. cinereus*, which show alternative reproductive tactics in the future. Otolith shape and size provide an important tool to contribute to fisheries population studies and fish stock management (Bose et al., 2018). We believe that this study will make important contributions to future studies that examine the factors affecting otolith shape and size in more detail.

AUTHORS' CONTRIBUTIONS

HO designed the release and performed the Lab work. YC interpreted the data. Both authors prepared the article**.**

CONFLICT OF INTEREST

The author confirms that no conflicts of interest exist and the funders had no role in study design, data collection, analysis, and decisions.

ETHICAL APPROVAL

For this type of study, formal consent is not required.

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