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Research Article

Age and Some Ring Characteristics and Morphometric Analyses of Mediterranean Horse Mackerel's Otoliths (*Trachurus mediterraneus* Steindachner, 1868) in The Black Sea

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Öz

Karadeniz'de İstavrit (*Trachurus mediterraneus* Steindachner, 1868) Balığı Otolitlerinin Yaş, Bazı Halka Karakterleri ve Morfometrik Analizleri

Bu çalışmada Karadeniz için önemli bir tür olan istavrit (*Trachurus mediterraneus* Steindachner, 1868) balığında farklı yaşlara sahip (0-5 yaş) otolitlere örnekler verilerek, türün otolit morfolojisi ve bazı halka yapısı özellikleri irdelenmiştir. Ordu-Rize arası ticari avcılığın yoğun olarak yapıldığı bölgelerden elde edilen örneklerden sağlanan 4265 adet otolit üzerinde çalışma gerçekleştirilmiştir. Otolitlerde şekil analizleri yapılarak F_F, R_D, A_R değerleri hesaplanmıştır. Morfolojik özelliklerine göre 4 farklı otolit tipi kaydedilmiştir. Nispeten yorumlanması güç bir otolit yapısına sahip olan istavrit de yaş tahmini çalışmalarında dikkat edilmesi gereken yalancı halka, larval dönem halkaları ve benzer oluşumlar belirlenmiştir. Bunun yanı sıra farklı tipte otolitler ve yapısal bozukluk gösteren bazı otolitler fotoğraflarla sunulmuştur.

Anahtar Kelimeler: İstavrit, otolit, *Trachurus mediterraneus*, Karadeniz.

Abstract

Mediterranean Horse Mackerel (*Trachurus mediterraneus*), one of the important species for Black Sea, was investigated in order to illustrate different aged otoliths' (0-5) morphology and some ring characters. This research was carried out with 4265 otoliths which were sampled from the regions have an intensive commercial fishing between Ordu and Rize. It was conducted otolith shape analyses F_F, R_D, A_R. Four different otolith types according to their morphological features were fixed. Mediterranean Horse Mackerel has a complicated otolith structure that is relatively difficult to interpret the age, for this reason false ring, larval stage rings and similar formations were considered. Besides, different type of otoliths and some structural defected ones were presented with photographs.

Keywords: Mediterranean horse mackerel, otolith, *Trachurus mediterraneus*, Black Sea.

Introduction

The Black Sea horse mackerel is the subspecies of the Mediterranean horse mackerel, *Trachurus mediterraneus*. (Aleev, 1957; 1959) considers that in Black Sea, the species is represented by four local subpopulations: the south side of Crimean, the northern side of Crimean, the eastern (Caucasian) and the southern (Anatolian) each one with its own biological characteristics. Mackerel is one of the most economical caught species after anchovy when the species belong to Black Sea are criticized. To get the greatest yield from important species for the region, fishable population of this improving appropriate management for growing and regeneration of the population is required without decreasing the productivity. Therefore, determination of growth characteristics of the species and age determination must be made as close as possible to reality. Age and growth studies is important for the solution of common problems of fisheries management. For this reason, determination of the growth properties and age determination for the species are requiered to be done in an accurated way (Polat, 2000). The age reading difficulties for a commercial species such as horse mackerel lead to the organization of several international otolith exchange programmes and workshops during the last decade (Eltink, 1985; ICSEAF, 1986; Marecos, 1986; Borges, 1989; Eltink and Kuiter, 1989; ICES, 1991). Erkoyuncu (1995) reported that determining age of an economical species small or large in comparison with the real age causes direct economic losses. Age data provides information about height and weight measurements of the age values, the stock composition, age at first sexual maturity, reproductive life, productivity, stock participation age, growth, death and the amount of product.

It is known that the otoliths are different size and shape. In most species sagitta otolith is the biggest one and mostly used in during the ageing of fish. Although the otoliths inside the inner ear have similar functions, their form, size, weight, growth, consistency and chemical composition vary considerably among species (Labeelund, 1988; Gauldie, 1994; Karlou-Riga, 2000; Yosshinaga et all., 2000). Otoliths are generally considered as taxonomical and biological archives as they reflect growth and development of species (Cotrell et all., 1996).

Some ring characters and morphological structures were examined to determine age of mackerel in this study. In addition otolith samples of different ages and otolith shape analysis are presented. It is expected from this study to be shed light on age and stock separation studies about Mediterranean horse mackerel.

Although the age interpretation for horse mackerel has been much improved, for Mediterranean horse mackerel the ageing appears to have many problems. The present work examined the structure of Mediterranean horse mackerel otoliths, as well as age, otolith morphology, otolith shape analysis and some ring characteristics.

Materials and Methods

This study was conducted in Black Sea coast of Turkey between Ordu and Rize (Figure 1).

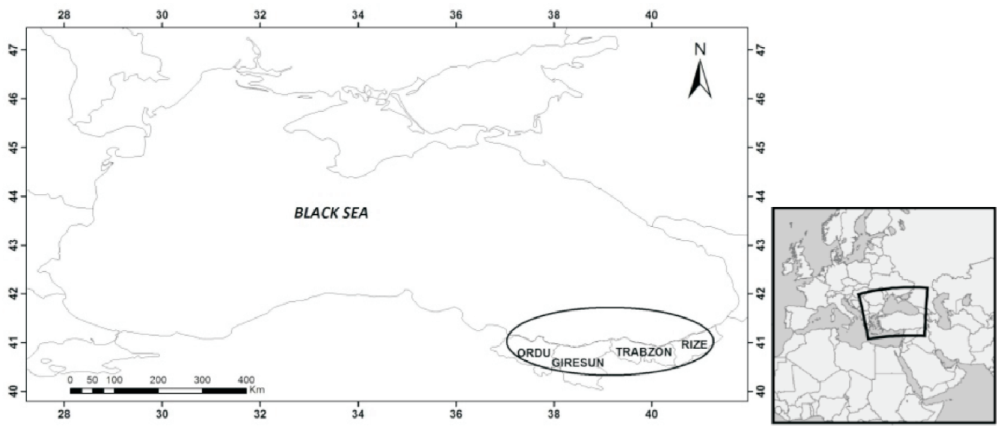

Figure 1. Map of the research area.

Samples of *Trachurus mediterraneus* were collected in this study from commercial purse-seiners and gill-nets and samples were caught monthly from January 2010 to August 2011. Samples of Mediterranean horse mackerel were brought to the laboratory by providing appropriate conditions. Gill gap of fish is opened with a fine-tipped tweezers and otoliths are removed without damaging then they are placed in to dish containig 90% alcohol in order to remove the remains of tissues finally they are placed in to Eliza petri dishes. Readings were realized of the distal surface (Figure 2).

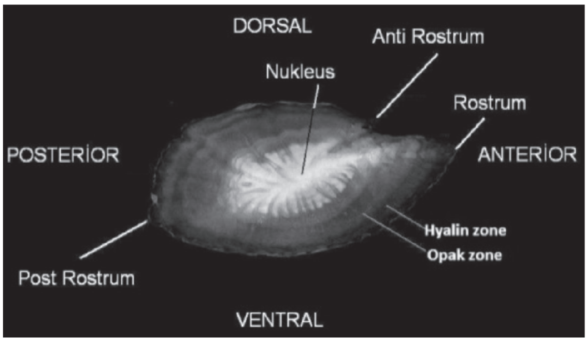

Figure 2. Distal surface of Mediterranean horse mackerel right otolith.

Photographs taken with 10x2,5 Leica MZ 75 magnification imaging system. When otoliths age group was determined Karlou-Riga (2000)'s criteria about *Trachurus mediterraneus* was considered and last age ring was regarded (Table 1).

Table 1. Age determining criteria in *Trachurus mediterraneus*

Date of capture	Otolith Edge	Age
1 January-30 June	Hyaline	n
	Opaque	n+1
1 July-31 December	Hyaline	n
	Opaque	n

n: Number of complete annulus.

Some of the criteria used to identify annuli in horse mackerel (Karlou-Riga and Sinis, 1997) were also followed for Mediterranean horse mackerel otoliths. These criteria were: (a) the annuli are deposited at progressively increasing distances from the nucleus, (b) the completion of the first annulus is detected on the rostrum, (c) a new opaque ring is sometimes accompanied by a change of otolith surface level, while the orientation of calcium layers does not always follow the post-rostrum axis and (d) when the opaque ring is deposited, a protrusion is sometimes formed above the surface of the otolith edge on the rostrum.

According to Ponton (2006), maximum otolith length (LO, mm), otolith width (W, mm), area (A, mm^2), perimeter (P, mm) measurements are taken for morphometric analyses which are included in the otolith shape definition. It is reported the width, height and weight measurements were not significant differences, as a result of analysis of variance (ANOVA) between left and right otoliths (Atılgan et all., 2012). Therefore, it is preferred right otolith in the calculations. Form factor, roundness and length ratio (Zorica et all., 2010) were calculated for otolith shape definition using the data acquired via measurements. LAS Image Analysis 3.8 software in the MZ75 Leica Imaging System was used for biometric measurements and shape analyses carried out on otoliths. The measurements acquired from the otoliths have been given in Figure 3.

Results

4265 otoliths are examined and was carried out on 3872 otoliths for age determination. The remaining 393 otoliths for various reasons (broken structure, crystallization, vateritic accumulation) were excluded from the study. Different aged otoliths are shown in Figure 4-5.

$$(FF)Form factor = 4\pi * Area \div Perimeter^2$$

$$(RD)Roundness = 4 * Area \div \pi Max\ R^2$$

$$(AR)Aspect\ Ratio = Length \div width$$

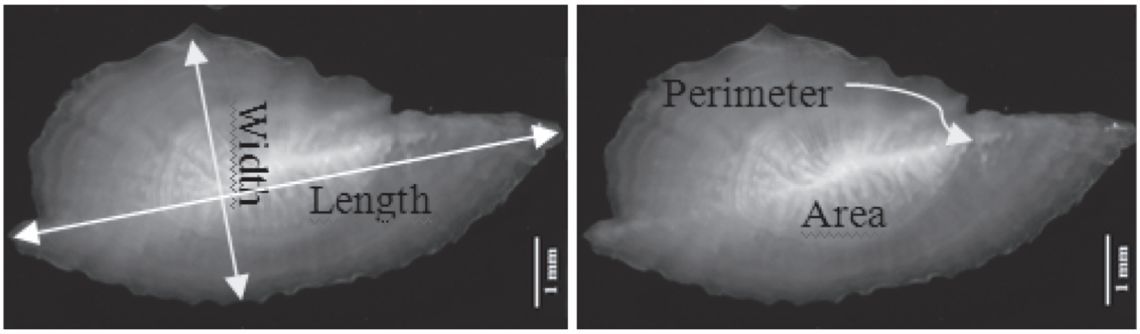

Figure 3. Measurements of the *Trachurus mediterraneus* otolith.

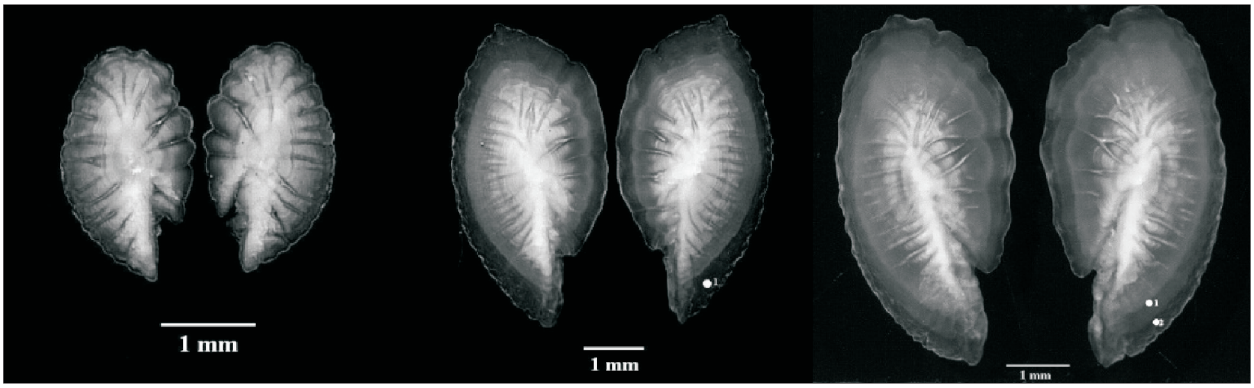

Figure 4. 0 age otolith. Catch date: 07.12.2010. Fish length: 9,3 cm.
1 age otolith. Catch date: 05.04.2011. Fish length: 14 cm.
2 age otolith. Catch date: 14.06.2011. Fish length: 15,8 cm.

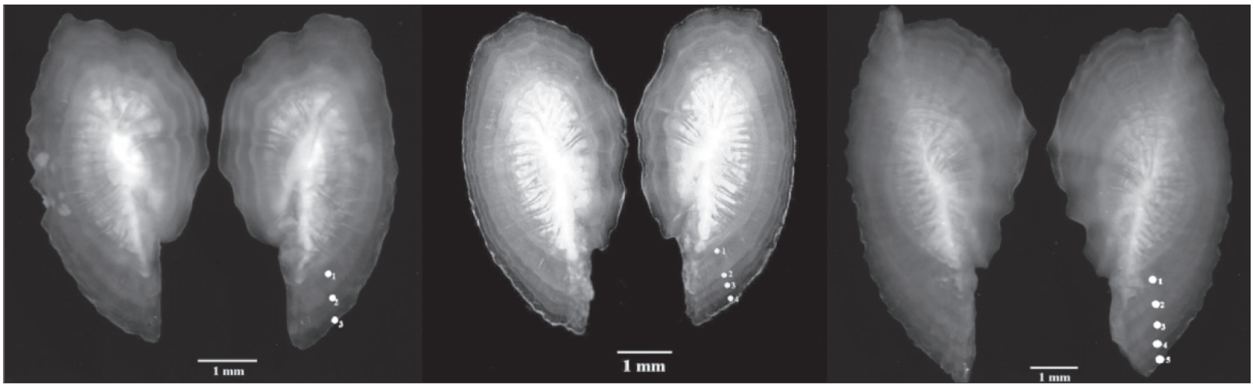

Figure 5. 3 age otolith. Catch date: 02.06.2010. Fish length: 15,6 cm.
4 age otolith. Catch date: 18.01.2011. Fish length: 17,2 cm.
5 age otolith. Catch date: 12.03.2011. Fish length: 21,4 cm.

Too thick or broken otoliths and vaterite accumulated ones were left out of assessment. Otoliths have vaterite structure are shown in Figure 6. Otolith readings are usually done considering anterior (rostrum) as part of the hyaline zones. However, to distinguish the real annuli from false rings continuation of the ring should be observed.

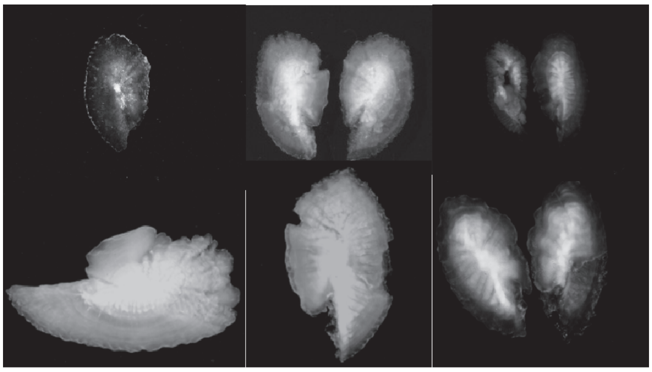

Figure 6. Undetermined Mediterranean horse mackerel otoliths because of vaterite accumulation.

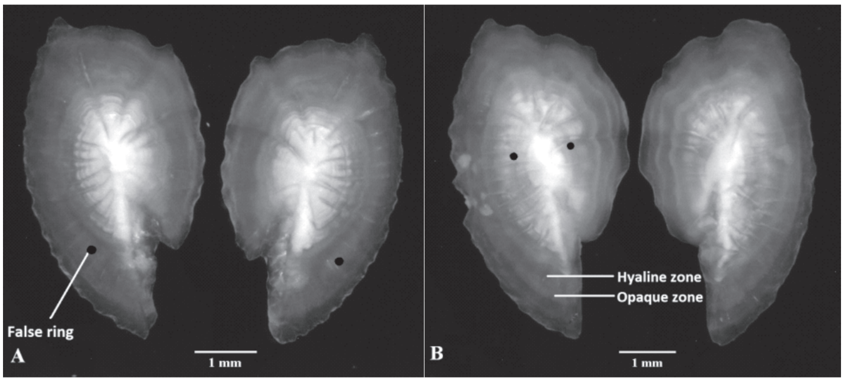

Figure 7. A) False ring formation in ventral (●). Catch date: 13.10.2010. Fish length: 14,3 cm.
B) Otolith growth zones and juvenil ring in the first opaque zone (●).
Catch date: 02.06.2010. Fish length: 15,6 cm.

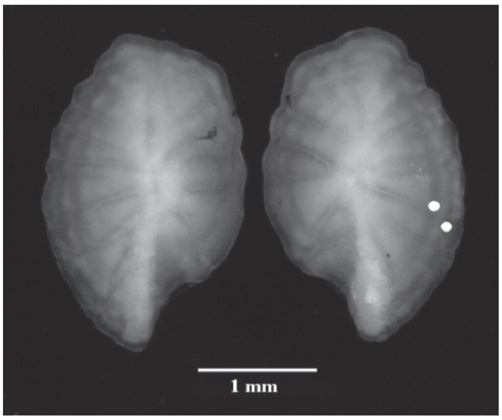

Figure 8. False rings in the first opaque zone (●). Catch date: 09.02.2010. Fish length: 7,4 cm.

Analyzed otolith structure of *T. Mediterraneus,* continuation of the otolith center (primordium) showing growth of the first year is the presence of a large opaque thin hyaline ring (juvenile ring) in the region. It is thought that this ring is a sign reflecting partial hunger when the fish drops its yolk sac to start free swim or reflecting an adaptation process rather than being an age ring (Figure 7.B).

The formation of false-rings in the otoliths of the studied species is generally observed after the first age ring or inside the first opaque increment (Figure 7.A, Figure 8).

This can be explained by the fact that there is more change in the life cycle of the horse-mackerel in terms of adaptation to the environment until its first age and the fact that it starts reproducing again after age 1. The fat and energy capacity of the species drops to a minimum level after spawning in the summer following the rapid increase in energy capacity in August and September. Irregularities in the formation of increments on the otolith during this period when the energy is directed to spawning causes the formation of false rings. These rings that are generally observed on the otolith surface ventral and less frequently at the postrostrum region can be differentiated from real age rings by lacking continuity in all aspects. Since accepting the hyaline structure observed in the first opaque zone along with the false rings as age rings will affect the age composition, age-length averages of the samples, it can cause substantial errors in the formation of the growth model as well as fishery planning. The formation of false rings in horse-mackerel otoliths can develop right before the first true annulus sometimes as a weak band and sometimes very thick formations that make it difficult to interpret the first age ring (Figure 9).

As can be seen in Figure 9, false annulus formation is not observed in picture A and the opaque region has continued until the annulus. Whereas in picture B, the formation of false annulus is partially observed. In picture C this formation is separated by a more distinct line while in picture D hyaline formation in the shape of a thick band is observed. 4 different otoliths according to morphological properties of horse-mackerel were recorded during the study and these 4 different otoliths have been shown in Figure 10.

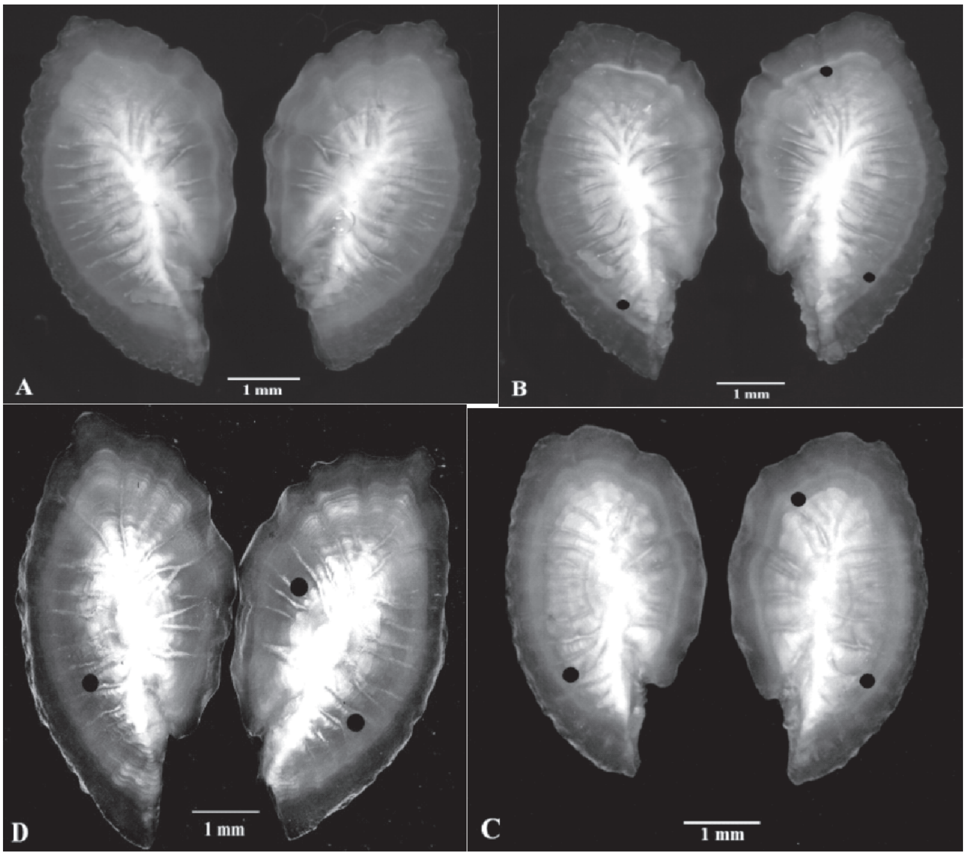

Figure 9. Prior to the first annual ring otolith samples with and without the formation of a false annulus.

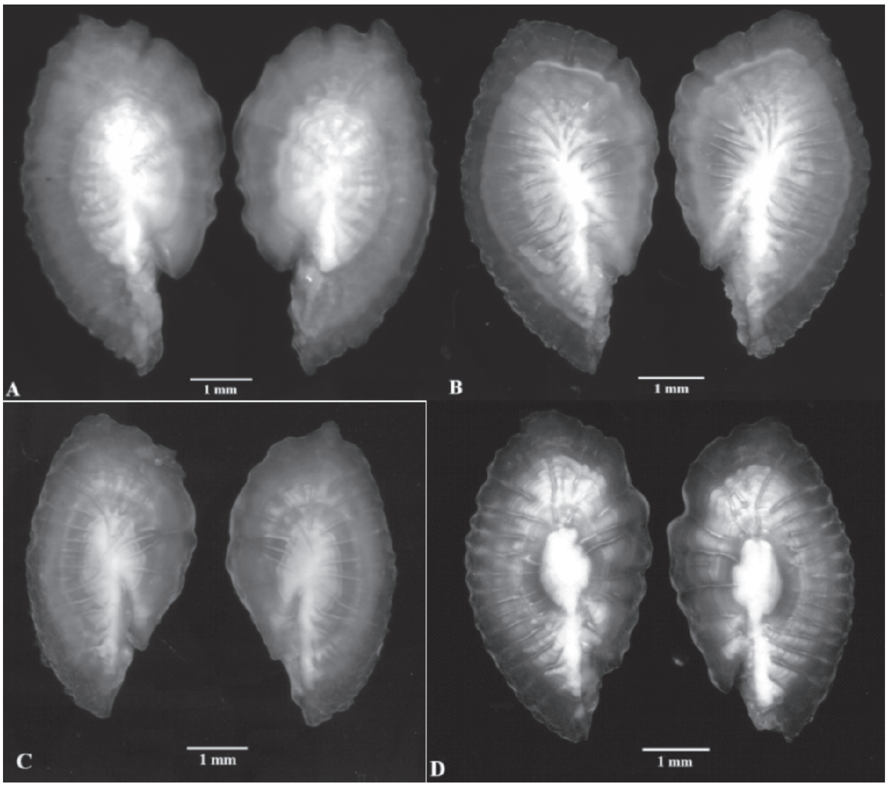

Figure 10. Mediterranean horse mackerel otoliths of different morphological structure.
A) Type 1: Catch date: 16.05.2011. Fish length: 15,5 cm.
B) Type 2: Catch date: 14.06.2011. Fish length: 15,8 cm.
C) Type 3: Catch date: 12.03.2011. Fish length: 14,8 cm.
D) Type 4: Catch date: 05.04.2011. Fish length: 13,7 cm.

Table 2. Morphometric measurements of otoliths

	Mean±SE	Min-Max	N
OL(mm)	4,19±0,111	2,45-6,32	92
OW(mm)	2,33±0,050	1,57-3,27	92
A(mm²)	7,17±0,323	2,77-13,53	92
P(mm)	12,21±0,339	6,99-18,38	92
R_D	1,61±0,021	1,25-2,20	92
A_R	1,78±0,014	1,46-2,10	92
F_F	7,05±0,141	4,76-9,59	92

(otolith length: OL, otolith width: OW, area: A, perimeter: P and shape defining factors (form factor: FF, roundness: RD, length-width ratio: AR), min-max, average and SE values)

It is believed these morphological differences to be related feding, ecological conditions and nutrient levels and oceanographic changes.

In this study, it was observed that average otolith length was 4,19 ± 0,111 mm. Average otolith width was 2,33 ± 0,050 mm, otolith area varied between 2,77 – 13,53 mm². Form factor acquired from otolith shape analyses was $4,76 \leqslant F_F \leqslant 9,59$ roundness factor: $1,25 \leqslant R_D \leqslant 2,20$, the length/width ratio was $1,46 \leqslant A_R \leqslant 2,10$ (Table 2).

Table 3. Correlation coeffecient between otolith length and shape factor (F_F, R_D, A_R)

F_F		R_D		A_R	
r^2	P	r^2	P	r^2	P
0,91	<0,001	0,42	<0,001	0,59	<0,001

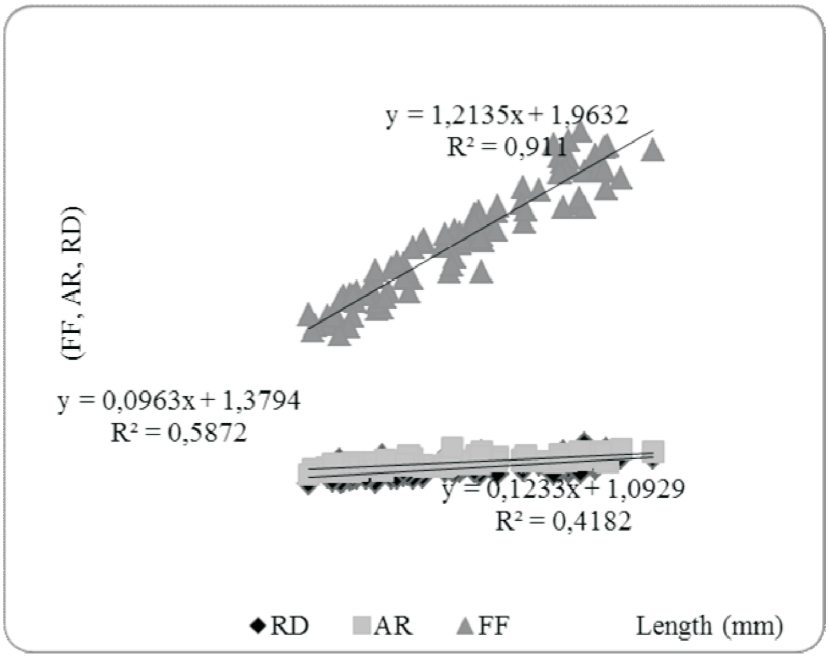

Figure 11. Relationship between otolith length and shape factor (F_F, R_D, A_R).

Relationship between otolith length and shape factor (F_F, R_D, A_R) is conducted respectively y= 1,213x+1,9632; y=0,1233x+1.0929; y= 0,0963x+1,3794. Correlation coeffecient between otolith length and shape factor is showed Table 3.

Correlation coeffecient between otolith length and FF is highest (r^2= 0,91; P<0,001). Correlation coeffecient between otolith length and R_D is weak (r^2=0,42;P<0,001) (Figure 11, Table 3).

Discussion

A criterion that strikes the eye when trying to determine the first age ring is that the first age ring generally borders the rostrum on the anterior. However, the case when the first annulus appears in the rostrum is not always helpful. If it is thought that the determination of the first age ring is an important criterion in the determination of the age of the fish, it is important for the decision regarding the central and the first age ring that the ring character and structure is known. Rostrum has a structure that elongates with increasing age.

When the otolith structure of the species is examined, a thin hyaline structure is observed in the wide opaque region until the first hyaline ring (Figure 7.B). These are false rings that are different in appearance from the real annulus. Waldron and Kerstan (2001) state that there is a false ring that forms in the first age ring of the juvenile ring in the *T. trachurus* otoliths. Bostancı and Polat (2008) report that rings that are similar to hyaline rings can form in periods when the fish growth has temporarily slowed due to reasons such as hunger or diseases along with sudden decreases or increases in water temperature which indicate that the growth of osteoid structures has decreased. These rings that are generally seen on the otolith surface can be separated from the real age rings since they are not continuous in all otolith directions. The results obtained are in accordance with relevant literature.

In general, horse mackerel otoliths are very difficult to be read in older fish because they become thick with age (Kerstan 1985; Eltink and Kuiter 1989; Karlou-Riga and Sinis 1997), usually produce a distinct preannual ring which can be identifiable where the anti-rostrum and rostrum have not been yet formed (Wysokinski 1985; ICSEAF 1986). In the current study, false ring formation in the species is generally observed in the ventral following the first age ring and less in the postrostrum region or in the first opaque accumulation (Figure 7.A, Figure 8). Various researchers indicate that there is an unformed false ring formation generally in the rostrum and antirostrum region of horse-mackerel otoliths (Wysokinski 1985; ICSEAF 1986). Karlou Riga (2000) has also reported that false ring formation generally occurs in the first year. In this sense, the current results are in accordance with those of other researchers.

The main reason for difficulty in ageing was the presence of sometimes very distinct false annuli (those prior to the first true annulus) which often led to difficulty in detecting the first annulus (Karlou Riga, 2000). False ring formations have been observed during age reading in otoliths right before the first true annulus sometimes as a weak band or sometimes as a very thick band which makes it difficult to interpret the first age ring (Figure 9). Karlou Riga (2000) has examined the otolith structure along with its age and growth of *Trachurus mediterraneus* stating that even though age determination studies have advanced in Horse mackerel species, there are many problems for Mediterranean horse mackerel. These problems generally occur due to the wrong interpretation of the first true annulus. The researcher states the existence of false rings that make it difficult to interpret the first age ring which are sometimes very thick right before the first true annulus. Results are in accordance in this sense as well. Four different otoliths have been determined in the species during age readings (Figure 10). Karlou-Riga (2000) has also separated the otoliths of the species into 4 different types morphologically during the studies carried out with the same species in Eastern Mediterranean (Gulf of Saronikos, Greece).

In this study different shape indices that has given us indications about form factor, roundness, aspect ratio of otoliths have also been used (Tuset et al., 2003). End of the shape analyses Form factor was $4{,}76 \leqslant F_F \leqslant 9{,}59$ roundness factor: $1{,}25 \leqslant R_D \leqslant 2{,}20$, the length / width ratio was $1{,}46 \leqslant A_R \leqslant 2{,}10$.

It has not been seen a study on the otolith shape analysis on Mackerel for the Black Sea in the literature while this study constitutes the first. This part of the study will lead to other studies for otolith shape analysis. This study has once again shown that the age readings carried out at an annual level should be supported with daily age reading studies which is still insufficient in our country and that this is required especially for the correct interpretation of the first age ring thereby ensuring a healthy age reading.

In future studies, the determination of different otolith types if any in addition to the 4 different otoliths determined in the current study will be beneficial to the determination of the otolith structure and ring characteristic of the species.

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