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Comparable Age Determination in Different Bony Structures of *Pleuronectes flesus luscus* Pallas, 1811 Inhabiting the Black Sea

Nazmi POLAT, Derya BOSTANCI, Savaş YILMAZ
Ondokuz Mayıs University, Faculty of Science and Arts, Department of Biology, Samsun - TURKEY

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Abstract: The aim of this study is to determine the most reliable bony structure for ageing of *Pleuronectes flesus luscus* inhabiting the Black Sea. The age determination method and its accuracy are the most important subjects for the evaluation of age-weight-length relationships in fisheries biology, since the estimates of survival, growth and mortality rates, and the analysis of year classes and recruitment all depend on age structure. In this study, scales, vertebrae and otoliths removed from each fish were examined by two readers three times each without reference to any data except the date of capture. The precision of readers and structures were estimated and the ageing error was determined by analysis of variance. The results are shown in tables.

Key Words: *Pleuronectes flesus luscus*, Black Sea, Bony Structure, Age

Karadeniz'de Yaşayan *Pleuronectes flesus luscus* Pallas, 1811'un Farklı Kemiksi Yapılarında Karşılaştırmalı Yaş Tayini

Özet: Bu çalışmada Karadeniz'de yaşayan *Pleuronectes flesus luscus*'un yaşının tespitinde kullanılacak en uygun kemiksi yapının belirlenmesi amaçlanmıştır. Yaş tayini metodu ve doğruluğu balıkçılık biyolojisinde yaş-uzunluk-ağırlık ilişkilerinin belirlenmesinde en önemli konulardan biridir. Dolayısıyla yaşama, büyüme ve ölüm oranları, yıl-sınıfı analizleri ve katılım ile ilgili hesaplamalar yaş kompozisyonuna dayandırılır. İncelenen örneklerin herbirinden alınan pul, omur ve otolit balığın yakalandığı tarih dışında hiçbir bilgi kullanılmadan iki farklı okuyucu tarafından, üçer kez incelenmiştir. Okuyucu ve yapı uyumu belirlenmiş ve yaş tayini hata payı değerleri varyans analiziyle hesaplanmıştır. Sonuçlar tablolar halinde gösterilmiştir.

Anahtar Sözcükler: *Pleuronectes flesus luscus*, Karadeniz, Kemiksi Yapı, Yaş

Introduction

Age determination is one of the most important stages in studies on subjects involving age-length keys, rate of survival, growth and mortality, age composition and reproduction rate of stock (1). Age and growth studies are important for problems associated with management of fisheries. Determination of age in fish helps in determining the age at first maturity, studying the population dynamics, estimating the growth and optimising the harvesting time (2).

Different methods are used in determining the age of fish. The most accurate method of age determination under natural conditions is the mark-release-recapture method; its application is limited in fisheries due to a number of constraints, such as time and money (2). The other method is the analysis of length-frequency data (3). This method is useful for fish which breed only once a year, and it cannot be used to determine the age of an individual fish (2). The third method is the anatomical approach. It is possible to determine the age of fish by evaluation of the growth rings forming on bony structures such as scales, otoliths, the opercular bone, the vertebra and the cross section of dorsal or pectoral fin rays. Ages of fish are estimated by comparison of the readings from various bony structures and different readers (4,5,6). The most reliable ageing method may vary among the species. Thus, the evaluation of the precision of bony structure by readers should be studied (7,8,9,10,11). Furthermore, ageing errors must be considered before deciding on the most reliable bony structure for the ageing of fish (8,12, 13, 14).

We have attempted to age *Pleuronectes flesus luscus*, since no study on this subject has been carried out before.

Materials and Methods

The study material consisted of 112 *Pleuronectes flesus luscus* specimens (Figure 1,2) provided from commercial fishery trawlers in the Black Sea between October 1998 and July 1999.

The weight, total length and standard length of each sample were recorded. The otoliths, vertebrae and scales were removed from the fish and prepared for ageing (15,16,17,18). Vertebrae and otoliths were examined by a binocular microscope with 10x magnification and top lighting. Scales were also examined under a binocular microscope with 10x magnification with transmitted light. Each bony structure was examined by two readers three times each. A total of 2016 readings were made (112 samples x 3 structures x 2 readers x 3 replicates). Two readers did not have reference to any information such as fish length, weight and sex, except the collection date of the sample. Readers examined each bony structure independently.

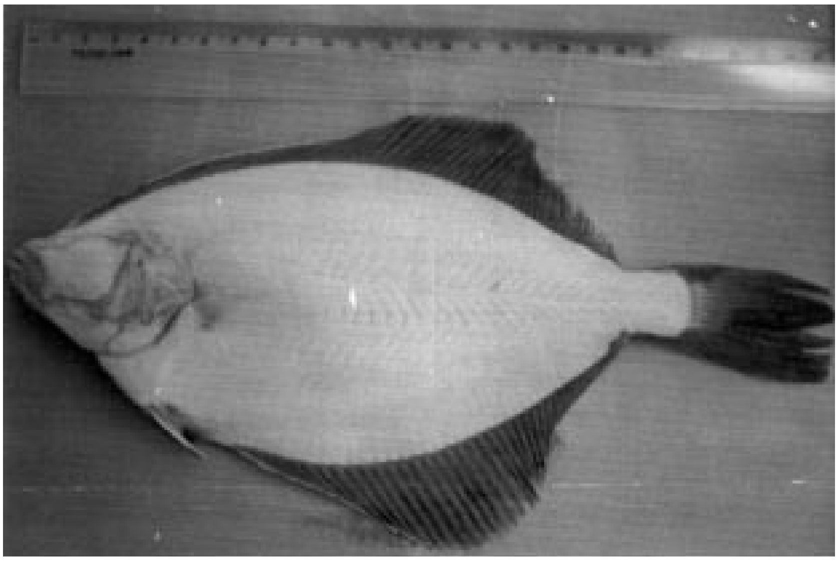

Figure 1. *Pleuronectes flesus luscus* from the blind side.

Figure 2. *Pleuronectes flesus luscus* from the ocular side.

Results

Age composition estimated from each structure is different (Figure 3-5). The vertebral age ranges from 0 to 6, the otolith from 0 to 5 and the scale from 1 to 6.

Mean age estimates were compared for structure-reader combinations, individual structures and readers (Table 1).

The lowest mean age was obtained in the otolith as 2.15 for the first reader and 2.07 for the second. Both of the readers recorded the highest mean age in vertebrae.

Precision of readers has been estimated from the percentage agreement of six readings (2 readers x3 times) (Table 2).

The highest agreement of two readers was 38.88%, in the vertebrae, and the lowest was 30.52% in the scales. Ageing errors for structure-reader, individual structure and individual reader were calculated. Both of the readers recorded the lowest ageing error on vertebrae at 0.59 and 0.67, respectively. So, the lowest mean ageing error has been obtained in this structure was 0.63. Data on ageing error is given in Table 3.

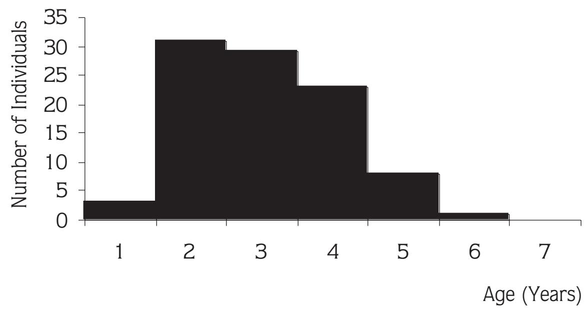

Figure 3. Age composition derived from reading the scale.

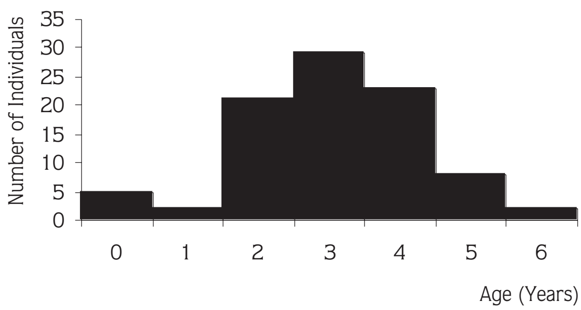

Figure 4. Age composition derived from reading the vertebra.

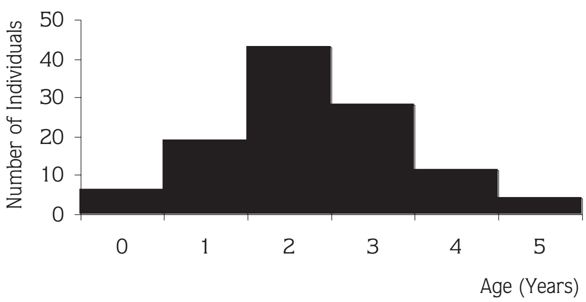

Figure 5. Age composition derived from reading the otolith.

Table 1. Mean ages for 112 samples.

Bony Structure Readers	Scale	Vertebra	Otolith	Mean	Bias
1	2.83	3.12	2.15	2.70	0.97
2	2.70	2.94	2.07	2.57	0.87
Mean	2.76	3.03	2.11		

Table 2. Precision of readers on different bony structures (Agreement of readers/number of total readings).

Bony Structure	Percentage Agreement of Age Estimations					Total
	6/6	5/6	4/6	3/6	2/6	
Scales	30.52	21.05	16.86	17.89	13.68	100
Vertebrae	38.88	24.44	17.88	15.55	3.25	100
Otoliths	34.23	25.22	15.31	15.31	9.93	100

Table 3. Ageing errors for the structure-reader combination.

Structure Reader	Scale	Vertebra	Otolith	Mean
1	0.69	0.59	0.63	0.63
2	0.72	0.67	0.73	0.70
Mean	0.70	0.63	0.68	

Discussion

Otoliths from the blind side are different in shape from those on the ocular side. Opaque and hyaline rings on the surface of both otoliths are arranged differently. The annuli are visible along the whole surface of the otolith on the blind side, but on the ocular side rings may be observed only on the anterior of the otolith (Figs. 6-7). This is mainly due to the varying position of focus on the otolith from two sides. The focus is central in the otolith of the blind side while the focus of the ocular side dislocates towards the posterior tip. A similar situation has been reported by Yabuki for *Tanakius kitaharai* (19).

Mean ages facilitate the elimination of the structure when an over- or underestimation exists. Table 1 shows that no over nor underestimation occurred in any of the bony structures. The difference between the two readers was limited to 0.13 years and this means that the criteria for age interpretation are relevant for readers.

Precision is related to reproducibility and is not a measure of accuracy. The degree of agreement among readers is a measure of the precision of the determinations and not the accuracy of the technique (1). Regarding the age groups, both readers obtained the highest agreements on the same structure, the vertebra (Fig. 8), as seen in Table 2. In a reliable age determination, the structure with the lowest ageing error is to be preferred. In this study, it is the vertebra which was determined to have the most minimal ageing error.

Due to the lowest agreements and maximal ageing error, the scale is not a reliable bony structure for the ageing of this species (Fig. 9).

We recommend that in studies involving the rate of survival, growth and mortality, age composition and reproduction rate of stock of this species, the vertebra may be used as the most reliable structure for the age determination of *Pleuronectes flesus luscus*.

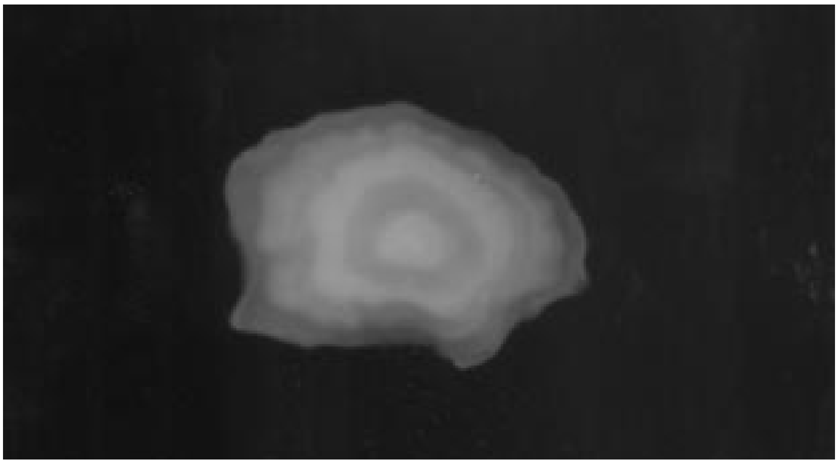

Figure 6. Blind side otolith with central focus. Age 3(4). (x15).

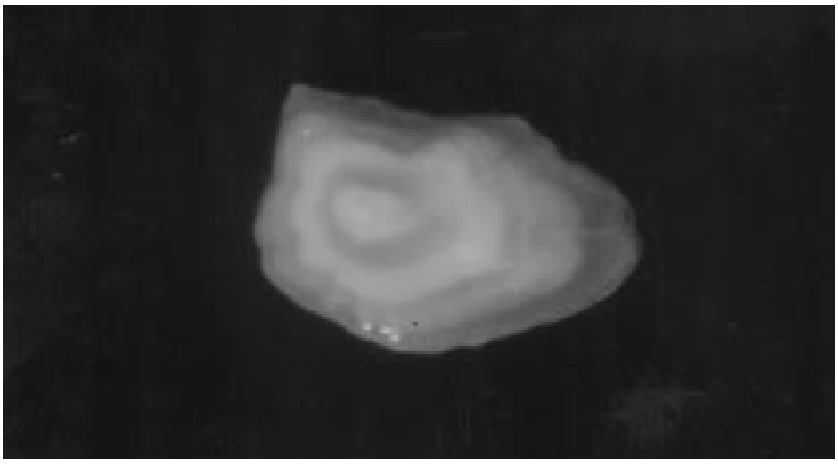

Figure 7. Ocular side otolith with focus closer to posterior. Age 3(4) on the anterior and 2(3) on the posterior. (x15).

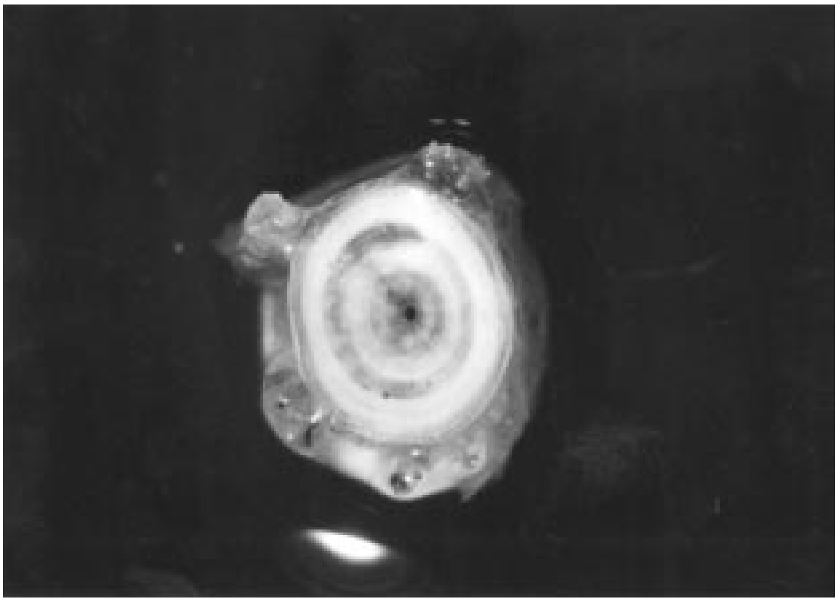

Figure 8. Vertebra from *Pleuronectes flesus luscus* age 2(3). (x15).

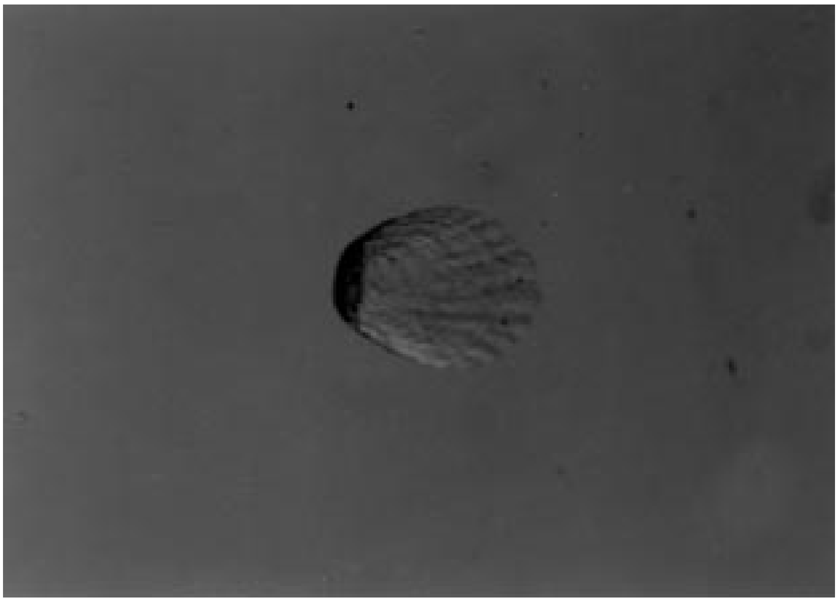

Figure 9. Scale from *Pleuronectes flesus luscus* age 2(3). (x20).

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