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First Report of the Scandium Element in *Pagrus pagrus* Otoliths: A Potential Indicator of Thermal Activity From Gökçeada Island, Turkey

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ABSTRACT

In this study, surface morphology and chemical structure of *Pagrus pagrus* (Linnaeus 1758) sagittal otolith from the shores of Gizli liman to Kefalos Cape in the north of Gökçeada Island, Turkey were investigated using scanning electron microscopy (SEM) and energy dispersion X-ray spectroscopy (EDS) analyses. According to the results of SEM analysis, the species was found to be over two years old. The age rings of *P. pagrus* were clearly identified on the otolith by SEM. According to the EDS results, it was examined that the element contents of age rings and regions between the sagittal otoliths obtained by EDS analysis. It was observed that calcium, carbon, and oxygen elements that form the calcium carbonate were the highest in EDS analysis. In addition, it has been found that the percentage of protein in ring structures, especially, in rings related to the age factor showed an increase. It can be said that this structural difference plays a major role in the clarification of age ring. Furthermore, the scandium (Sc) element was firstly discovered in the structure of the sagittal otoliths of *P. pagrus* from Gökçeada, Turkey. Scandium is one of the indicator elements in thermal regions. This study describes the presence of scandium trace element for the first time in the structure of otolith.

INTRODUCTION

The red porgy, *Pagrus pagrus*, is a demersal marine fish associated with a variety of temperate to subtropical habitats (Vassilopoulou & Papaconstantinou, 1992; Labropoulou et al., 1999). This species distributed throughout the Atlantic Ocean and Mediterranean Sea at depths of 18 to 280 m (Manooch & Hassler, 1978). Adults of this species inhabits rocky or gravel habitats (Manooch & Hassler, 1978; Alekseev, 1982). This species is a protogynous hermaphrodite that revealed an unbalanced sex ratio in favor of females (Manooch & Hassler, 1978; Vassilopoulou & Papaconstantinou, 1992). Red porgy is a carnivorous fish species that can reach up to 15-20 kg in weight. It has great economic importance for coastal fisheries in the Turkish waters. According to TurkStat data, commercial landings of red porgy were experienced a serious decline since 2009. Based on these data, it will be possible to mention that red porgy stocks are being overexploited.

The chemical composition of otoliths has significant contribution to the fisheries science. Fish otoliths record the chemical composition of the water that fish live in and travel

through. Otoliths are metabolically inert aragonite structures that vary depending on the physical and chemical properties of environment (Campana, 1999). Therefore, they become a potential tool that keeps a chronological record of the environments in which the fish live or temporarily present (Campana & Neilson, 1985; Campana, 1999). In addition, chemical structures found in otoliths are also used to determine differences between fish stocks (Campana et al., 2000; Gillanders & Kingsford, 2000; Rooker et al., 2003). Thus, the otolith chemistry is also used in order to detect the migration paths of fish (Ashford et al., 2008; Steer et al., 2010).

The aim of the current study is to determine the chemical structure of the sagittal otoliths of red porgy from Gökçeada Island, Turkey. This study is the initial step in our attempt to find out the specific environment that fish live or pass through.

MATERIAL AND METHODS

This study has been carried out from the shores of Gizli liman to Kefalos Cape in the north of Gökçeada Island, Turkey (Figure 1). The samples were collected using long lines, from 40–120 m depths between March and June in 2018.

The red porgy was measured for the purpose of obtaining the total length (TL) and total weight (W). Sagittal otoliths of red porgy were extracted and stored in Eppendorf tubes.

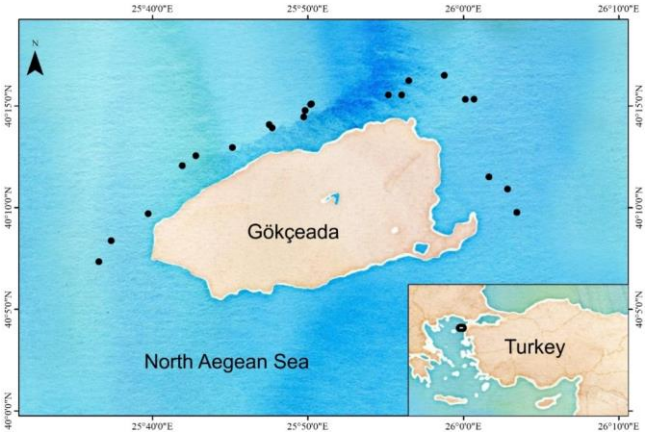

Figure 1. Sampling stations where red porgy, *Pagrus pagrus* were collected with long lines from the island of Gökçeada, Turkey, March–June in 2018

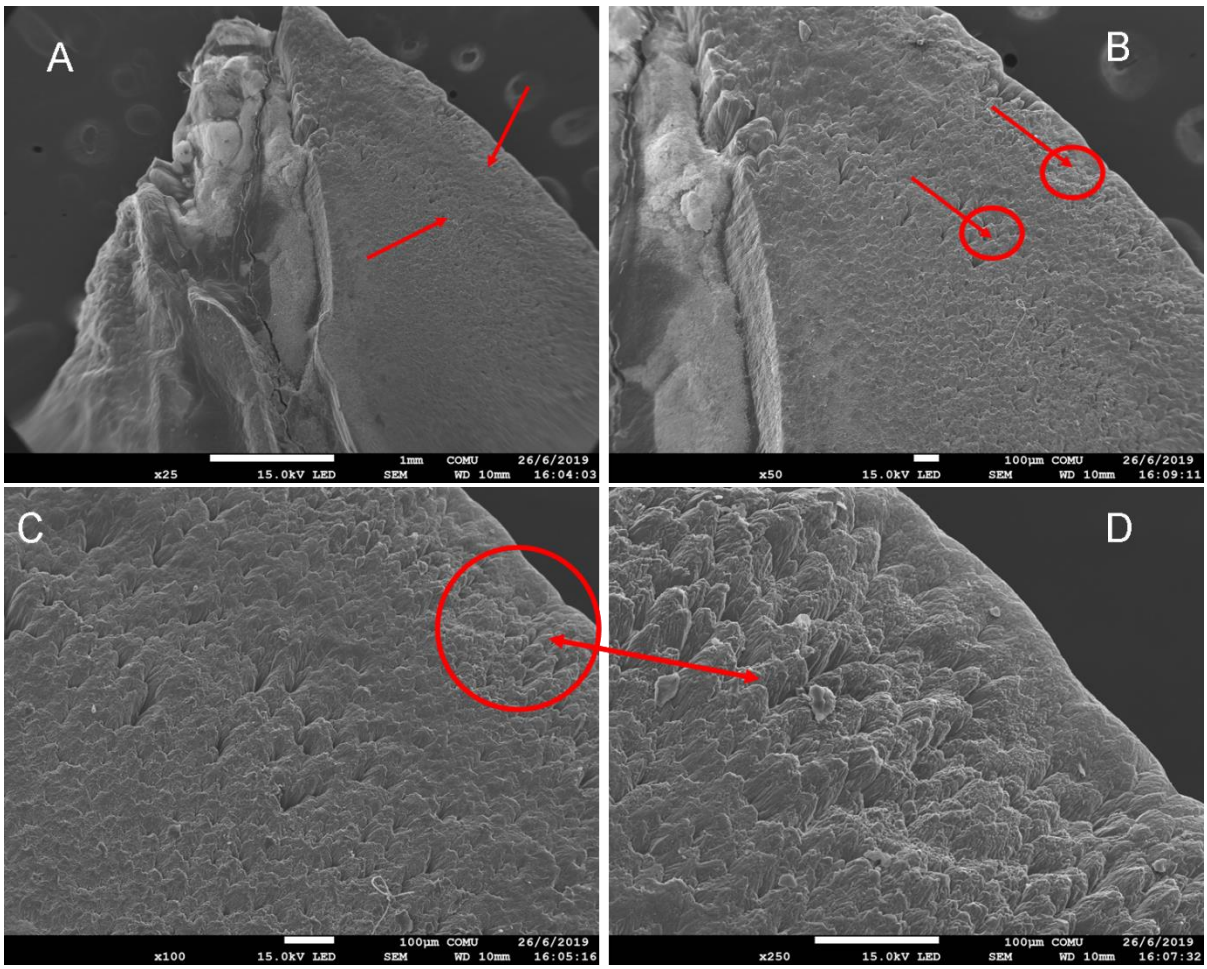

Figure 2. Scanning electron microscopy (SEM) images of *Pagrus pagrus*

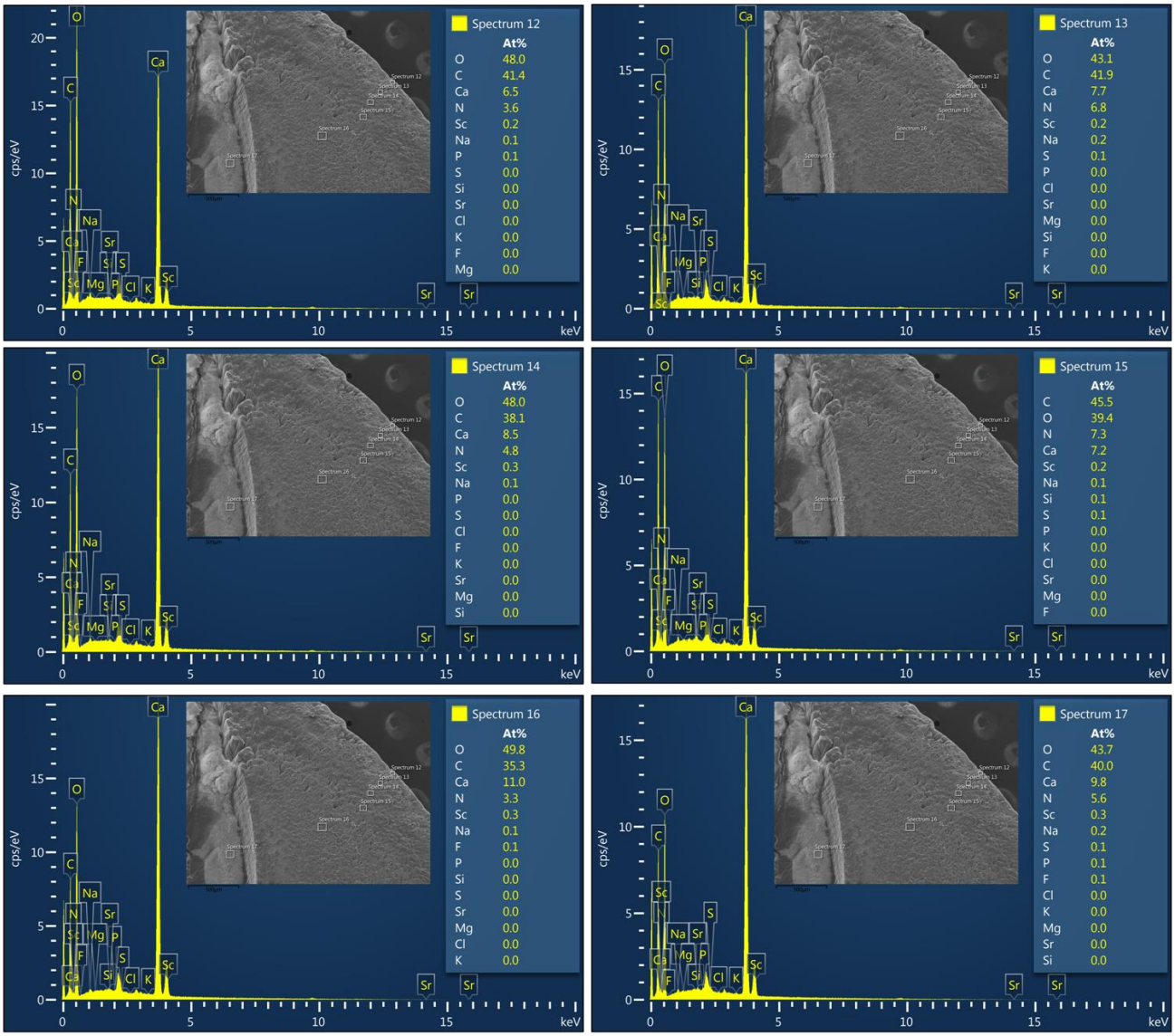

Figure 3. Energy dispersion X-ray spectroscopy (EDS) analyses of *Pagrus pagrus*

Scanning Electron Microscopy (SEM) Image and Energy Dispersion X-Ray Spectroscopy (EDS) Analysis

SEM images and EDS analyses were performed in the Science and Technology Application and Research Center of Çanakkale Onsekiz Mart University using JEOL JSM-7100F scanning electron microscope. The magnification capacity of the instrument and the accelerator voltage is between × 40 and 300,000 and 0.2 and 30 kV, respectively. The sample was coated with gold-palladium (80-20%) to increase the conductivity properties in 10 mA voltage and 8×10^{-1} mbar/Pa vacuum using Quorum coating device. It was used as secondary electrons for SEM images. Results related to EDX analysis are given in the form of percentages.

RESULTS

SEM images of the sagittal otolith of *P. pagrus* are given in Figure 2. There are two sharp and distinct layers on otolith showing in SEM images (A) and (B) and these layers were separated from each other. It seems that this fish is over two years old.

According to SEM images (C) and (D), it has been clearly seen that the calcium carbonate ($CaCO_3$) layers forming the main building block of otolith. Ca (CO_3) layers progressed layer by layer. It can be said that Ca (CO_3) layers progress depending on the growth and age of the fish on daily and/or annually.

The main chemical structure of otoliths is calcium carbonate ($CaCO_3$) and it is consisted of a small amount of protein as binding. Some specific elements of environment in which a fish lives are likely to include in an otolith. In addition to SEM images of *P. pagrus* (PP_{121}), important information has been obtained about the element contents and distributions by EDS analysis determined on different regions of the sagittal otolith. Analysis and the spectrum EDS results from 6 different regions of the sagittal otoliths of *P. pagrus* (PP_{121}) are shown in Figure 3. Calcium (Ca), carbon (C)

and oxygen (O) are the most common and basic elements in otoliths which are the main structural elements in the formation of Ca (CO_3) as shown in Figure 3. The minor elements of red porgy sagittal otoliths are represented by nitrogen (N), scandium (Sc), sodium (Na), silicon (Si), chlorine (Cl), phosphorus (P), sulfur (S), strontium (Sr) and magnesium (Mg) captured from Gökçeada Island. Particularly, the presence of Sc element is noteworthy. In addition, it has been seen that the amount of nitrogen (N) in proteins of otolith structure varies between 3.6% and 7.3%. EDS spectrum from different regions of otoliths provided important information about the basic structure of otolith. As Figure 3 indicates, especially for the spectrum 13 and 15, these regions belonging to the transition regions between the first and second age. In addition, according to the element results of EDS analysis, the nitrogen (N) amount is higher than the other regions.

It is believed that the ring lines of the fish age consist of protein-based structure instead of pure calcium carbonate. This situation might possibly cause to make clearer the age ring. The elemental distributions of 16, 14 and 12 spectrums in the intermediate regions of age rings showed that the nitrogen amount is low.

DISCUSSION

Scanning electron microscopy provides detailed and comprehensive information about surface morphology. Thus, the surfaces of the sagittal otoliths of red porgy can be examined in detail. In this study, surface morphology and chemical structure of *Pagrus pagrus* (Linnaeus 1758) sagittal otolith from the shores of Gizli liman to Kefalos Cape in the north of Gökçeada Island, Turkey were investigated using scanning electron microscopy (SEM) and energy dispersion X-ray spectroscopy (EDS) analyses. Otoliths are mainly composed of aragonite and elemental ratios which record the physico-chemical properties of water inhabited by a fish at any point during its life (Thorrold & Swearer, 2009). According to the results obtained from SEM analysis, it was observed that the surface morphology and properties of otoliths changed according to age factor. Age rings were observed in otolith structure and the regions inside the age rings differs in terms of surface properties.

According to the obtained element contents, it has been seen that Ca, C and O elements, the basic structure of $CaCO_3$, are formed in otolith structure. In addition, it is found that the protein structure in otolith is also determined with the presence of N element in the results of EDS analysis. Na, K and Li elements were not determined by EDS analysis. This is mainly due to the EDS analysis performs a percentage amounts to determine the detection limit of elements. This does not mean that there are no undetectable elements in the results of EDS analysis. Furthermore, that's why, Sr and similar trace elements cannot be determined by EDS analysis. In this study, surface properties, age rings, basic structure, and trace elements of *P. pagrus* otoliths were determined by SEM and EDS analyses.

CONCLUSION

The scandium (Sc) element was firstly discovered in the structure of sagittal otoliths of *P. pagrus* from Gökçeada, Turkey. Scandium is one of the indicator elements in thermal regions. This study describes the presence of scandium trace element for first time in the structure of otolith. There is information about Sc, hydrothermal events and erosion and volcanic rocks being mobilized during metamorphosis (Finlowbates & Stumpfl, 1981). Oudin & Cocherie (1988) conducted sedimentary analysis in Red Sea and associated hydrothermal activities and the accumulation of some trace elements, including Sc in sediment. Accordingly, a hypothesis can be established that the *P. pagrus* used in this study spend a certain part of their lives in specific environment or pass through with hydrothermal resources. Consequently, it is recommended to test this hypothesis in future studies.

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Compliance with Ethical Standards

Authors' Contributions

All authors made contributions in necessary fields during the preparation of samples, conduction of experiments, evaluation of results, and writing of the article.

Conflict of Interest

The authors declare that there is no conflict of interest.

Ethical Approval

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

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