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First Morphological and Genetic Record and Confirmation of Korean Rockfish *Sebastes schlegelii* Hilgendorf, 1880 in the Black Sea Coast of Türkiye

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Abstract

One specimen of Korean rockfish *Sebastes schlegelii* Hilgendorf, 1880 was caught by using a fish net at a depth of 7 m on 14 May 2022 from Akçakoca, Düzce in the Black Sea. In this study, the Korean rockfish *Sebastes schlegelii* from the Black Sea coast of Türkiye is reported for the first time with both morphologic and genetic evidence. *S. schlegelii* is characterised by 5 spines on the preoperculum and 2 spines on the operculum. Genetic analyses using mtDNA COI gene region also confirmed the species as *Sebastes schlegelii*. This species is thought to have been transported to the Black Sea from other seas of the world, probably via ballast waters or aquarium escape. The most important reason for this is that the species has not been detected in the Mediterranean.

Keywords:

Sebastes schlegelii, rockfish, first record, Black Sea, genetic evidence

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Introduction

The introduction of new species to the Black Sea can significantly disrupt ecosystem stability and functioning, posing a major threat to biodiversity. Mediterranean species have been increasingly

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recorded in the Black Sea since the 1920s, leading to the expansion of Black Sea ichthyofauna diversity, a phenomenon known as "Mediterranization" (Pusanov, 1967; Turan et al., 2009; Yağlıoğlu & Turan, 2021).

The Korean rockfish *Sebastes schlegelii* belongs to the Sebastinae subfamily and represents the Scorpaenidae family. The Sebastinae subfamily comprises about 124 valid species in 4 valid genera (Fricke et al., 2023). They are generally known as rockfishes. Its natural distribution area is the Pacific (North) Ocean (Hyde & Vetter, 2007; Kai & Soes, 2009; Kai et al., 2013;). Only six species are found in the Atlantic Ocean: *S. capensis* and *S. oculatus* on the southern Atlantic coast, and *S. fasciatus, S. mentella, S. norvegicus* and *S. viviparus* on the north coast of the Atlantic Ocean. Recently, *Sebastes schlegelii* has been reported on the Crimean coast of the Black Sea (Karpova et al., 2021). On the other hand, this genus has not been recorded in the Mediterranean, but in the Black Sea.

In this study, we first reported the Korean rockfish *Sebastes schlegelii* from the Black Sea coast of Türkiye is given for the first time with both morphologic and genetic evidence.

Materials and Methods

One specimen of the Korean rockfish *Sebastes schlegelii* from Akçakoca, Düzce, Türkiye, South Black Sea coast 41.086195, 31.090505, (41° 5' 10.302" N - 31° 5' 25.818" E) (Figure 1) on 14 May 2022 at a depth of about 7 m was captured by fisherman (Hüseyin Demircioğlu) using fish net. Captured species were immediately frozen and transported to the laboratory for detailed measurements in the laboratory. Each body length (±0.1 mm) and total body weight (W) (±0.01 g) were measured. The sample of *S. schlegelii* (Figure 2) was carefully examined and identified using previous records from the Black Sea by Karpova et al., (2021) and from Dutch coastal waters by Kai & Soes (2009). After that, the specimen was preserved in 98% Ethanol solution and deposited at the Duzce University, Faculty of Arts and Sciences, Department of Biology (catalogue number: DUFC/2022-001).

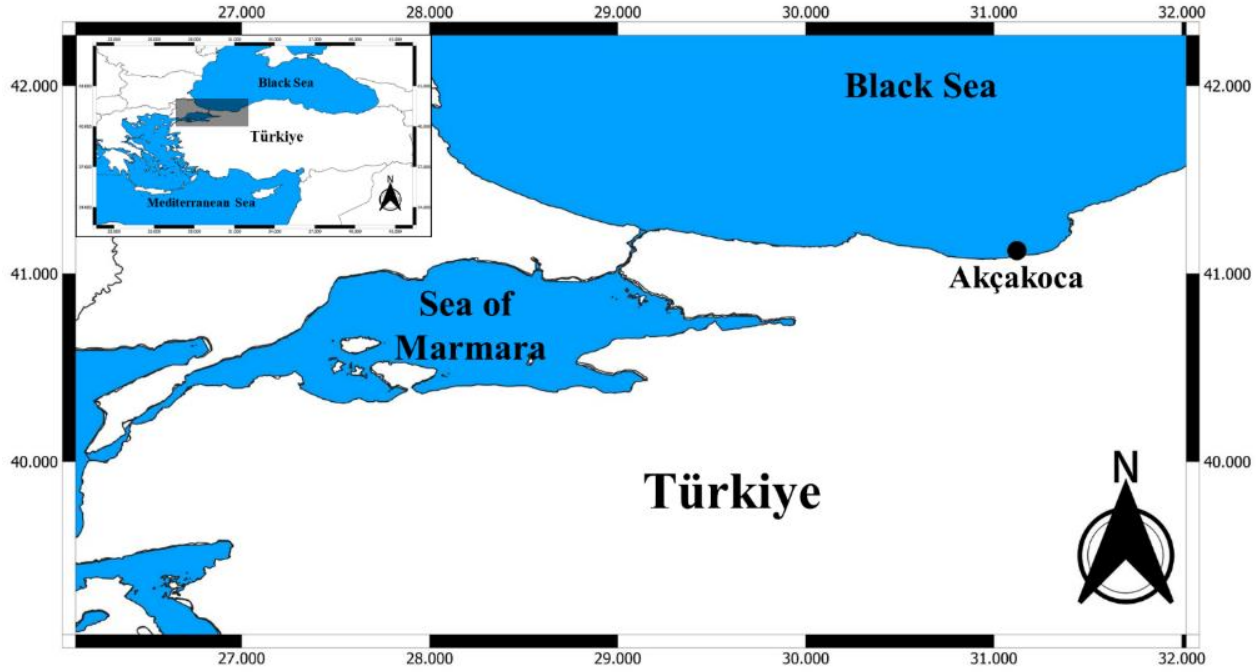

Figure 1. Sampling location of S. schlegelii from the Black Sea Coast of Türkiye.

Figure 2. *S. schlegelii* was captured in the southwestern Black Sea, Turkey.

Molecular Analysis

The specimen was delivered to the laboratory and stored in a deep freeze at -30 °C until DNA extraction. Total genomic DNA was extracted from the muscle sample using the DNeasy Blood and Tissue Kit (Qiagen, USA). The manufacturer's protocols were used during all steps. The mtDNA COI gene region was amplified through PCR with selective primers especially designed for groupers (Doğdu & Turan 2016). Fish_F: 5'-TCA ACC AAC CAC AAA GAC ATT GGC AC-'3' -Fish_R: 5'-ACT TCA GGG TGA CCG AAG AAT CAG AA-'3'.

The PCRs were conducted in a 50 µl total volume with 0.4 uM of each primer, 0.2 mM of dNTP and 1.25U of Taq DNA polymerase in a PCR buffer that included 20 mM of Tris–HCl (pH 8.0), 1.5 mM of MgCl2, 15 mM of KCl and 1-2 µl template DNA. The denaturation step was at 94 °C for 30 s, 50 °C for 30 s, and 72 °C for 45 s for 30 cycles followed by a final extension for 7 min at 72 °C. The PCR products were visualized using electrophoresis on 1.5 % agarose gel. DNA sequencing was attempted to determine the order of the nucleotides of the mtDNA COI gene region. The chain termination method by Sanger et al. (1977) was applied with Bigdye Cycle Sequencing Kit V3.1 and ABI 3130 XL genetic analyzer. The initial alignments of partial COI sequences were performed with the BioEdit (Hall et al., 2011). The intraspecific and interspecific genetic analyses were performed with maximum likelihood (ML), and Neighbor-Joining analyses. The best-fit substitution model (TN93) was provided by the MEGAX software (Kumar et al., 2018). After sequence alignment, MEGA X was used to determine the genetic diversity and sequence divergences and to construct the phylogenetic tree (Kumar et al., 2018). Sequences of other Sebastinae species were obtained from Genbank.

Results

Diagnosis and Description

Taxonomic identification of the captured specimen (Figure 3) was classified as *Sebastes schlegelii* which can be distinguished from other Sebastinae species by the following characteristics: dorsal-fin rays XIII, 13; anal-fin rays III, 7; pectoral-fin rays 18; lateral-line scales 47; caudal fin rays 16 and convex; anal fin rounded. While it may be difficult to make a precise and definitive description regarding color and body patterns, it can be said that they are seen with a light coloration on the ventral region. Sebastinae species can exhibit variations in colour and patterns due to different age groups, gender differences, and their living environment. Therefore, it is not possible to state that this species is entirely of a specific colour or pattern. The presence of five distinct, sharp, bony, tooth-like structures on the preoperculum and two on the operculum. Moreover, the first spine of the opercular margin long arch being hook-shaped enables a definite distinction of the species.

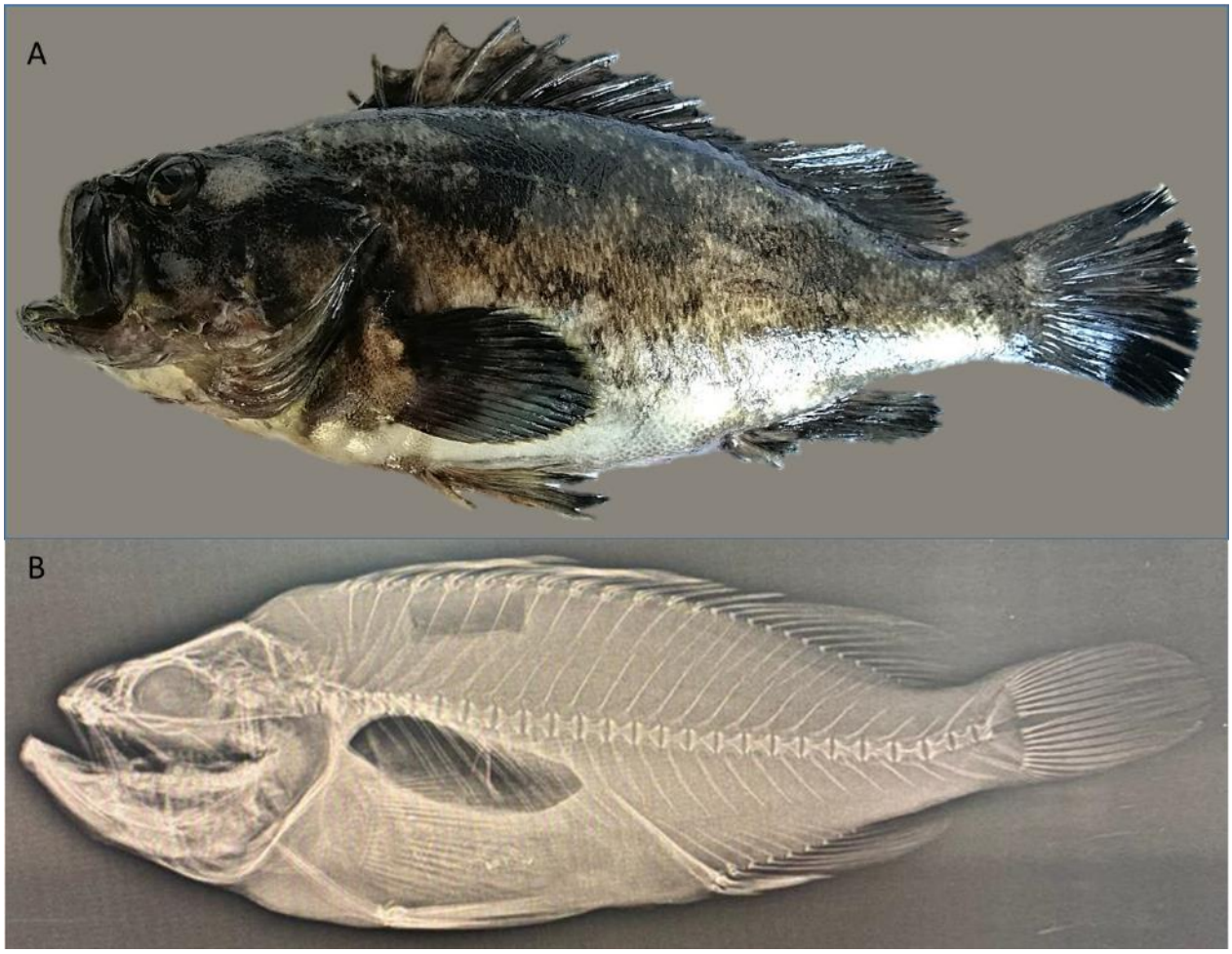

Figure 1. A) General view of S. schlegelii B) Radiograph of S. schlegelii DUFC/2022-001, 34.96 mm TL.

S. schlegelii is a fish with a wide mouth, protruding lower jaw, and a robust body relative to its size, featuring a large head covered in tough, tight scales. Its color can vary from light tones to black, with noticeable differences. It stands out with its lighter color on the ventral area. The tail fin, or caudal fin, is characterized by its upward structure and is not forked. Generally, its fins have a rounded shape. The dorsal fin consists of 13 spines and 13 soft rays. All the morphometric measurements and meristic characters of *S. schlegelii* are given in Table 1 with previously recorded.

Table 1. Morphometric measurements and meristic characters of the *S. schlegelii* captured in the Black Sea.

Morphometric Measurements	**This Study**	**Karpova et al. (2021)**	**Kai & Soes (2009)**
Total Length	34.96	35.07	20
Standard Length	29.93	29.75	-
Head Length	9.12	-	-
Snout Length	2.79	-	-
Body Depth	10.19	-	-
Body Width	5.95	-	-
Orbit Diameter	1.72	-	-
Interorbital Width	2.82	-	-
Length of Pelvic-Fin	10.19	-	-
Lengths of The Dorsal	4.72	-	-
Lengths of The Anal	5.32	-	-
Lengths of The Pectoral	6.03	-	-
Lengths of The Caudal Fins	5.08	-	-
Caudal-Peduncle Depth	3.04	-	-
Predorsal Length	9.3	-	-
Prepelvic Length	10.17	-	-
Longest Hard Dorsal Spine	4.26	-	-
Longest Soft Dorsal Rays	4.72	-	-
Meristic characters			
Gill Rakers	24	-	-
Lateral-Line Scales	47	47-49	-
Pectoral-Fin Rays	18	18	18
Anal-Fin Rays	III-7	III-7	III-7
Dorsal-Fin Rays	XIII-13	XIII-13	XIII-12/13
Pelvic-Fin Rays	I-5	I-5	I-5
Caudal-Fin Rays	16	-	-

Genetic analyses

The mtDNA COI gene region sequence of S. schlegelii was obtained at 612 bp and deposited to the Genbank with accession number: OR577041. The Genbank database was used to compare with sequences of other Sebastinae species distributed all over the world. Neighbor-Joining (NJ) and

Maximum Parsimony (MP) trees of the cuptured and other species from Genbank are given in Figure 4 and Figure 5.

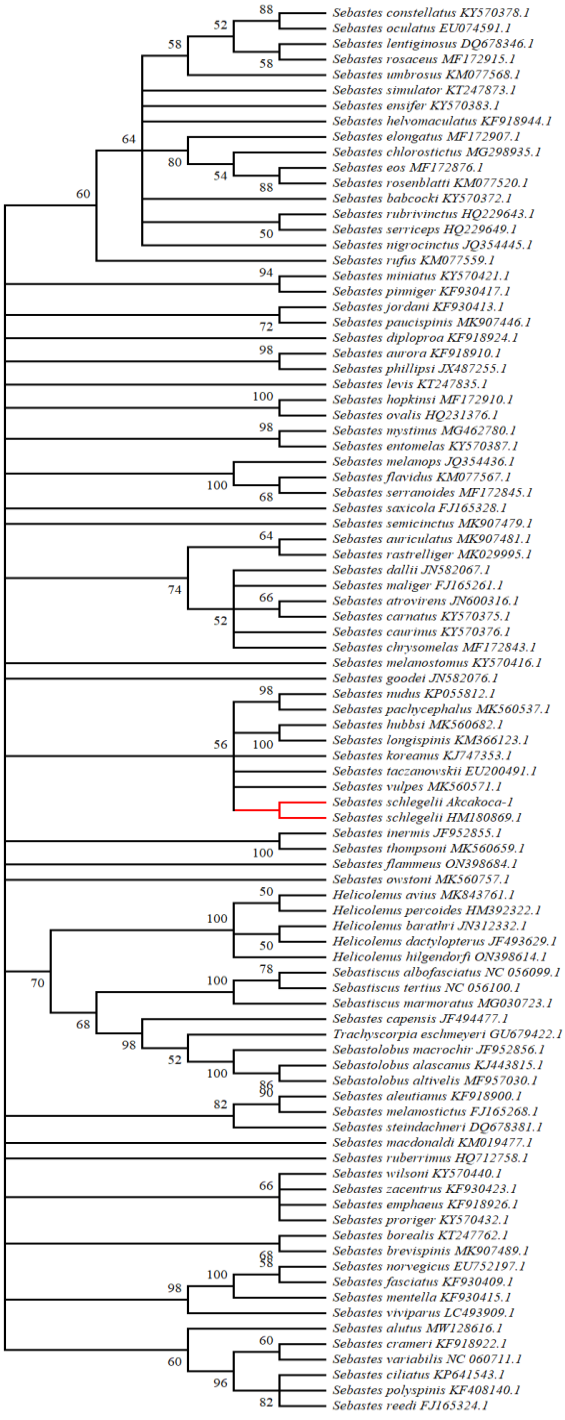

Figure 4. Neighbour-Joining tree analyses of S. schlegelii and other Sebastinae species distributed in the World. The red branch of the tree shows that the species we captured matches the record in the GenBank database.

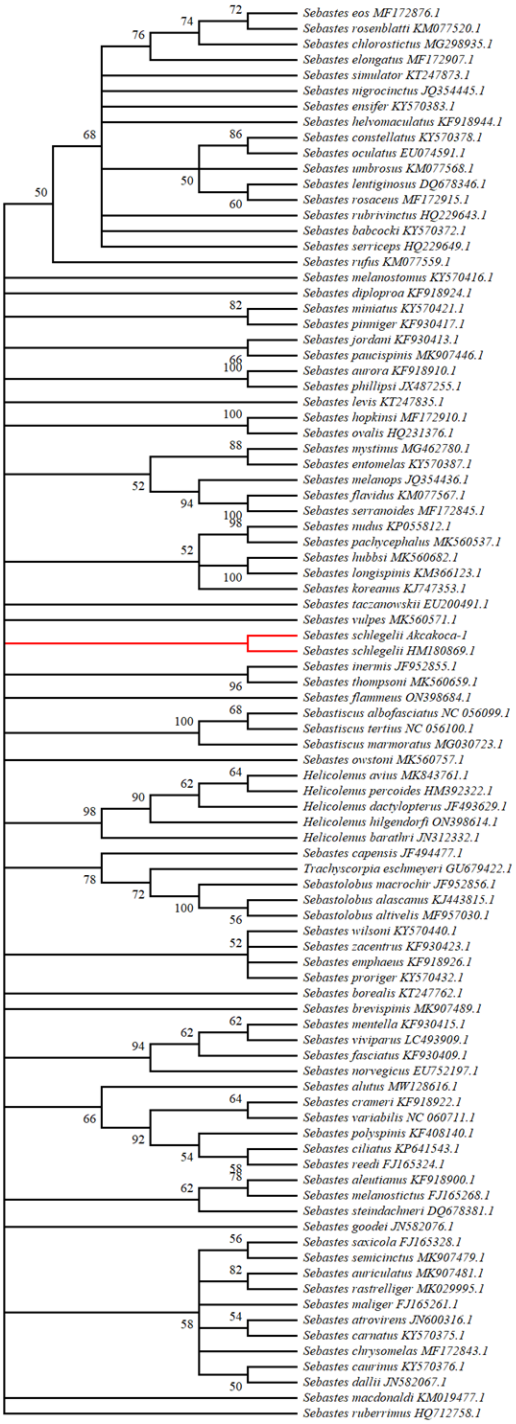

Figure 5. Maximum Parsimony tree analyses of S. schlegelii and other Sebastinae species distributed in the World. The red branch of the tree shows that the species we captured matches the record in the GenBank database.

Neighbour-joining and Maximum Parsimony analyses demonstrated that the captured species is *Sebastes schlegelii*, revealing the genetic confirmation of this record on the Black Sea coast of Türkiye.

Discussion

In this study, we first reported on the Korean rockfish *Sebastes schlegelii* from the Black Sea coast of Türkiye with both morphologic and genetic evidence. *S. schlegelii* is characterised by 5 spines on the preoperculum and 2 spines on the operculum. Genetic and morphological analyses confirmed that this species is *S. schlegelii*. This species is thought to have been transported to the Black Sea from other seas of the world, probably via ballast waters or aquarium escape. The most important reason for this is that the species has not been detected elsewhere in the Mediterranean. The species was only reported from the Black Sea Crimean Coast by Karpova et al. (2021).

In recent years, the occurrence of Atlanto-Mediterranean species in the Black Sea has been reported several times in various studies (Yağlıoğlu et al., 2014; Lipej et al., 2017; Yağlıoğlu & Turan, 2021). The migration of Lessepsian fish to the Turkish coast of the Black Sea was also reported for the first time by Turan et al. (2017). The changing ecological structure of the Black Sea, due to factors such as the increase in water temperature related to global climate change and variations in the influx of freshwater from closed seas like the Black Sea over the years, is considered the main reason for the entry of Mediterranean species into the Black Sea (Ben Rais Lasram et al., 2010; Turan et al., 2018). Turan et al., (2016) previously stated that there is a tendency of increasing temperatures in the Black Sea due to global climate change, and this may lead to an increase in the number of Mediterranean-Atlantic and Lessepsian fish species in the Black Sea.

The abundance and distribution of S. schlegelii should be monitored and tracked to understand its relationship with the native fauna of the Black Sea. Additionally, the potential impact of global climate change on the Black Sea environment and water quality parameters may accelerate or facilitate the Mediterraneanization process of the Black Sea. Therefore, further scientific research and support are crucial to comprehend the settlement process of species transported to and established in the Black Sea. Additionally, although we do not have samples on hand, based on photographs and videos received from fishermen engaged in recreational fishing along the Amasra and Samsun coasts, it is evident that the species is spreading along the Black Sea coast. It is believed that in the near future, this species may emerge as an economically important species in the Black Sea.

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assistance in providing us with photographs of the individuals caught in other provinces along the Black Sea coast of Türkiye.

Conflict of Interest

The authors declare that they have no competing interests.

Author Contributions

C.T. performed all the experiments and drafted the main manuscript text. D.Y., and S.A.D.. C.T. D.Y., and S.A.D.. performed genetic analysis. C.T., D.Y., and S.A.D. contributed morphological analysis and drafting of the manuscript. All authors reviewed and approved the final version of the manuscript.

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