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Rapid Communication

A first record of non-native Korean (black) rockfish *Sebastes schlegelii* Hilgendorf, 1880 from the Bulgarian Black Sea coast

Petya P. Ivanova, Nina St. Dzhembekova, Violin S. Raykov and Yordan Raev

Institute of Oceanology – Bulgarian Academy of Sciences, 40 Parvi May St, Varna, 9000, Bulgaria

Corresponding author: Violin S. Raykov (vio_raykov@abv.bg)

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Abstract

A first record of non-native *Sebastes schlegelii* (Hilgendorf, 1880) was made in Bulgarian Black Sea waters. One specimen was collected from the coastal waters (Varna breakwater) of the Bulgarian Black Sea. The identification was based on morphological characteristics and DNA barcoding. The morphological measurements covered 34 morphometric and 6 meristic features. A fragment of cytochrome oxidase subunit I (COI) gene of mitochondrial DNA was sequenced to supplement the species identification. Moreover, five specimens were caught during 2022 and 2023 by sport fishermen. These collections represent the first official report of *S. schlegelii* along the western Black Sea coast.

Key words: non-native species, mtDNA, morphology

Introduction

The biota of the Mediterranean and the Black Sea has started to change significantly over the last few decades due to the introduction of non-indigenous species as a result of Lessepsian migration, ship ballast water, range expansion of Atlantic species, intentional or unintentional introduction and climate change. Reliable scientific data on the dynamics of their distribution and abundance are essential to understand their ecological and economic effects (Öztürk 2021).

Rockfish of the genus *Sebastes*, family Sebastidae, consist of approximately 110 species (Fang et al. 2015). Most of them occur in the North Pacific (Kai et al. 2003), concentrated mainly around an Asian center near Japan and North Atlantic (Roques et al. 2002) and across the southern hemisphere (Rocha-Olivares et al. 1999). These species display great diversity in body color, ecology, and behavior (Kai et al. 2003).

The Korean (black) rockfish *Sebastes schlegelii* (Class Actinopterygii, Order Scorpaeniformes, Family Sebastidae) is a demersal fish widely distributed in the northwest Pacific along the coast of China, Japan and the Korean Peninsula (Wang et al. 2017). It is a commercially valuable and ecologically important rockfish species (Gao et al. 2016), characterized by strong adult site fidelity and a preference for rocky reef, silt and sand habitat (Zhang et al. 2015).

Outside their native range, the species was described in the south-western coastal waters of the Netherlands (Kai and Soes 2009) and coastal zone of Crimea and Düzce, Turkey, Black Sea (Karpova et al. 2021; Yağlıoglu et al. 2023). However, the presence of this species along the Bulgarian Black Sea coast was hitherto unknown.

In the current study, the first occurrence of non-native species S. *schlegelii* from the Bulgarian Black Sea coast based on morphological and molecular analyses is reported, confirming its expansion in the Black Sea.

Materials and methods

Sampling

A total of six fish were collected from Bulgarian Black Sea coastal waters – Varna (5) and Kiten (1), between May 2022 and August 2023 by sport fishermen (Figure 1, Supplementary material Table S1). The sampling locations were: in marine terminal station of Varna, Varna breakwater east side wall at 4–5 m depth, 5 to 6 m distant from the wall and north-east side of the Kiten Pier, at 5–6 m depth, 2–3 m distance from the pier. The bottom of this area is sandy, covered with rocks and with seaweeds. All the specimens were photographed (Figure 2). One of the individuals caught at Varna breakwater on 23.08.2023, was used for morphological and molecular identification (Figure 3).

Morphological study

Counts and measurements generally followed Pravdin (1966) and Kai and Nakabo (2002). Standardization of morphometric data is expressed as a percentage of standard length (SL). Measurements were taken by common caliper, based on the length of the measured items, recorded to the nearest 0.1 mm. Meristic measurements (counts) included: dorsal fin spines and rays, pectoral fin rays, anal fin spines and rays, pelvic fin spines and rays, and pored lateral line scales. Terminology of head spines was according to Phillips (1957) and Wibowo and Motomura (2022).

DNA extraction and PCR amplification

A tissue sample from the pectoral fin was cut and preserved in 96% ethanol at 4 °C. The genomic DNA was isolated using the DNeasy Blood & Tissue Kit (QIAGEN), and the target DNA was amplified with universal set of mitochondrial primers – cytochrome *c* oxidase subunit I (COI) FishF2: 5'TCGACTAATCATAAAGATATCGGCAC3' and FishR2: 5'ACTTCAG GGTGACCGAAGAATCAGAA3' (Ward et al. 2005). The polymerase chain reaction (PCR) was carried out in a reaction volume of 50 µl containing 1 µl of each primer, 25 µl of the mastermix (MyTaqTM HS Mix) and 2 µl of the target DNA. The PCR conditions included the following parameters: 95 °C for 1 min, and 95 °C for 30 sec, 54 °C for 30 sec, 72 °C for 1 min

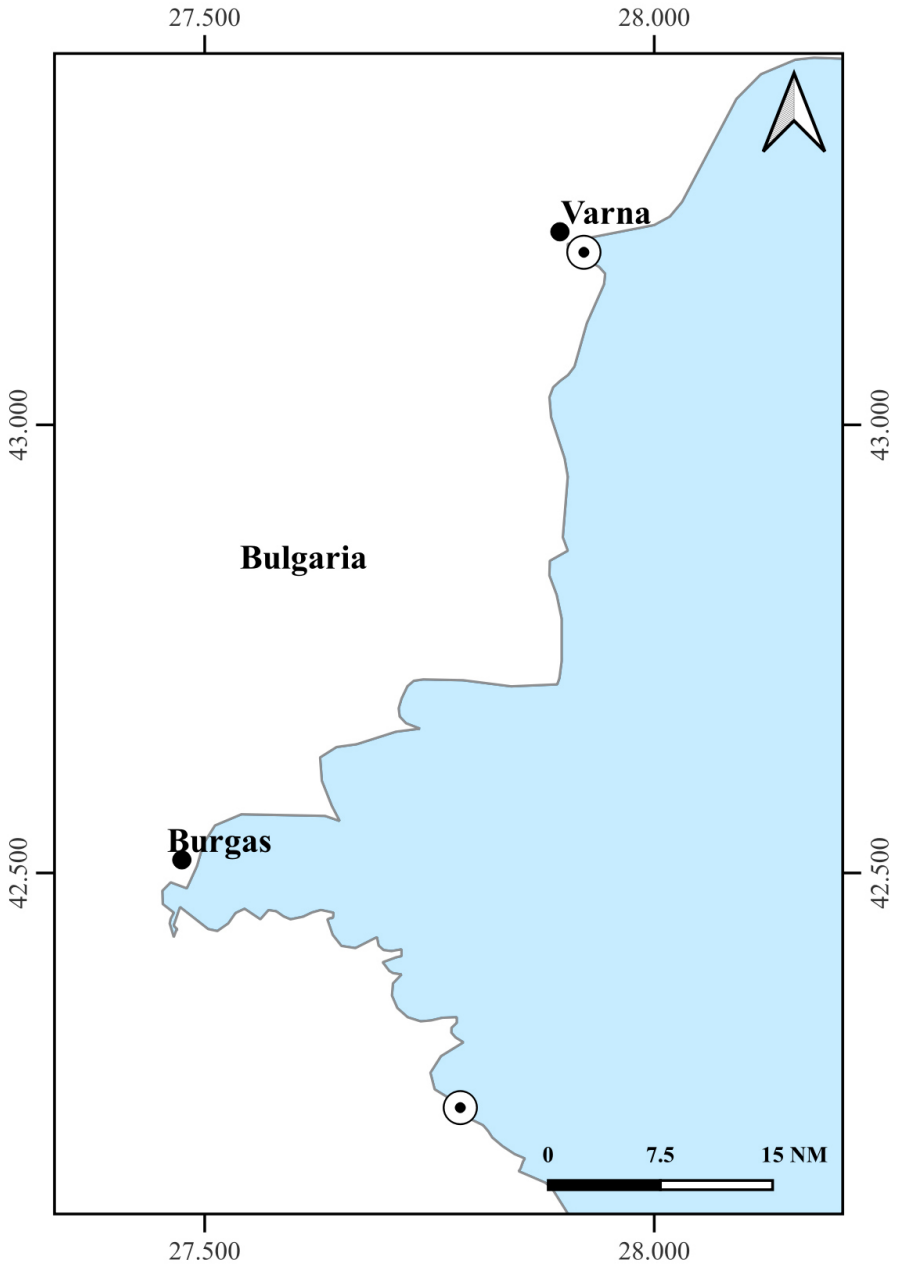

Figure 1. Locations of sampling sites (Varna and Kiten) for *S. schlegelii* along the Bulgarian Black Sea coast in the period 2022–2023.

(35 cycles), 72 °C for 10 min. A quality control of the PCR product was performed by electrophoresis on 1% agarose gel. The DNA sequencing was performed by Macrogen Europe B.V. The obtained sequence was submitted to GenBank under the accession number OR 528864.

Results and discussion

A first record of non-native Korean rockfish (*S. schlegelii*) from Bulgarian Black Sea coast was documented based on morphological characters and DNA barcoding.

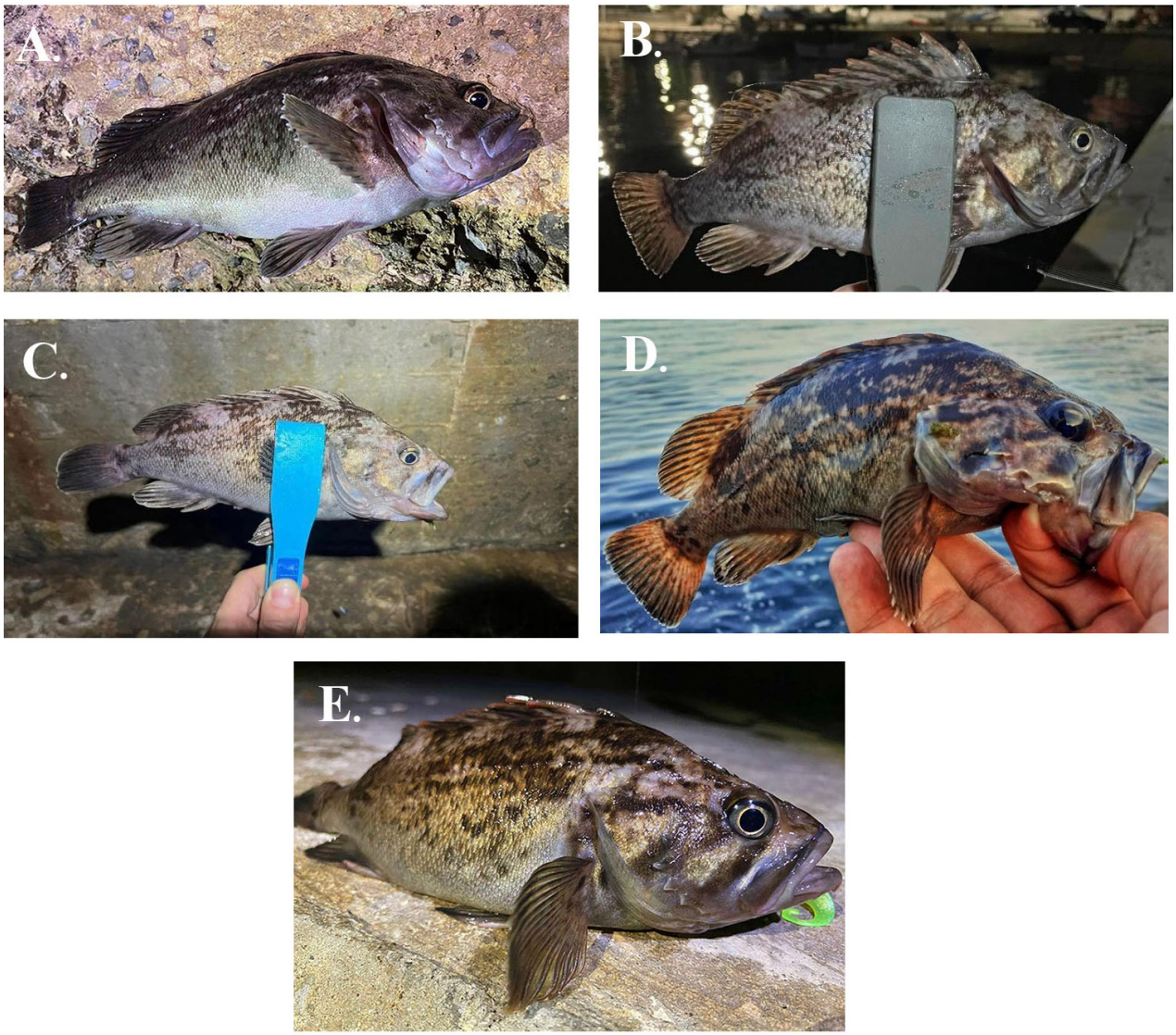

Figure 2. Photos of the samples sent by sport fishermen: A–D – Varna breakwater (N = 4) and E – Kiten Pier (N = 1). Photo by Hristian Hristov and Dian Stoev.

Morphological characters

The coloration of the specimen was dark gray with a marbled pattern of small indistinct darker spots and the following meristic characters were recorded: dorsal fin XIII–13, pectoral fin 17, the contour of the pectoral fin is rounded, anal fin III–8, pelvic fin I–5, caudal fin 15, pored lateral line scales 47 (Figure 2, Table 1). Head covered with scales. Three lachrymal spines observed. Nasal, preocular, postocular, tympanic, parietal, and five preopercular spines were present (Figure 3B, C). Mouth oblique directed upwards, the lower jaw protrudes, the anterior teeth of the lower jaw do not protrude. The upper and lower lips were dark gray, without transverse bands. The total length of the analyzed specimen was 230 mm, whereas the other five samples it varied between 230 and 280 mm.

The measured morphometric characters were compared with the previously reported *S. schlegelii* records for the Black Sea (Karpova et al. 2021 and Yağlıoglu et al. 2023). A significant data overlap was observed (Table 1).

Figure 3. The *Sebastes schlegelii* specimen caught along the Bulgarian Black Sea coast (Varna breakwater), A. specimen lateral view, B. head lateral view and C. frontal view of the head. Photographs by Petya Ivanova and Nina Dzhembekova.

The meristic data (the number of fin rays in the dorsal fin, fin spine in the anal fin and fin spine and rays in pelvic and caudal fins) were not fully described by Karpova et al. (2021) and are not presented in Table 1.

Molecular data

The obtained COI gene fragment (664 bp) showed 100% similarity with *S. schlegelii* sequences from China and South Korea (GenBank accession numbers OQ932947 China, HM180869 South Korea, KP330553 and KY275270 China) and 99.18% with a *S. schlegelii* isolate from the Turkish Black Sea (GenBank accession number OR577041) supporting the species identification.

Species spreading and potential impacts

Changes in the marine environment, particularly climate change, can have large effects on the distribution patterns of various marine species, and alter the biodiversity, structure and functions of the affected ecosystems (Chen et al. 2021). Due to its geographical position, the survival and establishment of marine alien species into the Black Sea might be amplified

Table 1. Morphological measurements of *Sebastes schlegelii*, Black Sea expressed as percentages of standard length.

Measurements	*Sebastes schlegelii*, Varna Bulgaria	*Sebastes schlegelii*, Crimea and Caucasus (Karpova et al. 2021) mean values	*Sebastes schlegelii*,Turkish Black Sea coast, Yağlıoglu et al. (2023)
TL, mm	230	350.7	349.6
SL, mm	200	297.5	299.3
W, g	227	918.5	
Counts			
Dorsal fin	XIII-13	XIII	XIII-13
Anal fin	III-8	7	III-7
Pectoral fin	17	18	18
Pelvic fin	I-5		I-5
Caudal fin	15		16
Pored lateral line scales	47	47-49	47
(% SL)			
Head length	36.5	37.8	30.5
Head height	27	28	
Snout length	6.5	11.6	9.3
Orbit length	6.5	7.4	5.7
Interorbital width	7.5		9.4
Postorbital length	17.5	19.8	
Upper jaw length	15	18.3	
Lower jaw length	23.5	20.7	
Maximum body depth	32	35.1	34
Maximum body width	18	22.5	19.9
Caudal peduncle depth	9	10.3	10.2
Caudal peduncle width	3.5	4.1	
Upper peduncle length	6.5		
Lower peduncle length	12.5		
Pectoral-fin length	22.5	22.2	20.1
Pelvic-fin length	19.5	20.4	
Dorsal-fin base length	56	62.6	
Spinous dorsal-fin base length	37.5		
Soft dorsal-fin base length	18		
Preanal length	64	68.4	
Predorsal length	27.5	34.4	30.2
Postdorsal length	11.5	12.8	
Prepelvic length	38	38.1	34
Prepectoral length	34	34.6	
Anal-fin base length	16.5	15.9	
Pelvic-to-anal-fin length	27.5	24.4	
1st dorsal-fin spine length	4		
2nd dorsal-fin spine length	7.5		
3rd dorsal-fin spine length	11.5		
1st anal-fin spine length	3		
2nd anal-fin spine length	9		
3rd anal-fin spine length	9.5		
Pelvic-fin spine length	9.5		
Gill raker length	22.5		

by climatic change. Increases in the mean annual water temperature might make the Black Sea environment more suitable or even favorable to certain alien species (Băncilă et al. 2022). Chen et al. (2021) consider the potential of climate change-induced impacts on *S. schlegelii* distribution.

The first record of *S. schlegelii* in the Black Sea was in 2013 along the coastal zone of Crimea but sightings have become more frequent, suggesting successful introduction (Karpova et al. 2021). Our data support this hypothesis considering the fact that six samples were caught along the Bulgarian Black

Sea coast in the period of one year. In our study, individuals of different sizes were described, which suggests a naturalization of the species in the Black Sea, rather than penetration of single specimens (Karpova et al. 2021). Random introduction of *S. schlegelii* with ship ballast waters or during acclimatization of the giant oyster *Magallana gigas* as a potential object for mariculture is assumed to be the most probable reason for the appearance of the species in the Black Sea (Mitov et al. 2020; Karpova et al. 2021; Yağlıoglu et al. 2023).

Research on the impacts of alien species to the ecosystem and fisheries economy has been very limited in the Black Sea. In order to support the ecosystem recovery in spite of all types of negative impacts for the sustainable management of the natural resources, it is essential to understand, analyze and take urgent management measures regarding all aspects of the ecosystem (Kasapoglu et al. 2015).

According to its ecological features, *S. schlegelii* can compete with local Black Sea species (e.g. scorpionfish *Scorpaena porcus* Linnaeus, 1758), however, no influence of the invasive species on the coastal ecosystems was reported till now, possibly due to its small numbers (Karpova et al. 2021).

The source of introduction, current extent of spread, and ecosystem impacts of non-native *S. schlegelii* are still unknown and special investigation is needed to answer these questions and to understand the effects of this species on the native aquatic species. The monitoring of non-native species is essential in order to minimize and reduce its possible negative impacts in the Black Sea ecosystem and protect marine biodiversity.

Authors' contribution

All authors contributed to the study conception and design. Material preparation, data collection and analysis were performed by Petya Ivanova, Nina Dzhembekova, Violin Raykov and Yordan Raev. The first draft of the manuscript was written by Petya Ivanova with the contributions of Nina Dzhembekova and Violin Raykov. All authors commented on previous versions of the manuscript. All authors read and approved the final manuscript.

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Supplementary material

The following supplementary material is available for this article:

Table S1. Locations of the sampling sites of black rockfish along the Bulgarian Black Sea coast for the period May 2022–August 2023.

This material is available as part of online article from:
http://www.reabic.net/journals/bir/2024/Supplements/BIR_2024_Ivanova_etal_SupplementaryMaterial.pdf