Iran. J. Ichthyol. (2024) 11(2): 79-92
P-ISSN: 2383-1561; E-ISSN: 2383-0964

ORIGINAL ARTICLE

Morpho-molecular approaches reveal presence of an additional grouper fish, the oblique-banded grouper, *Epinephelus radiatus* (Teleostei: Epinephelidae), in the northeast coast of the Oman Sea with a note on its scale, otolith and caudal skeleton morphology

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Article history:
Accepted 28 May 2024

Abstract

Consistent and ongoing biodiversity exploration is crucial for understanding diversity patterns over time and space and evaluating conservation efforts. Despite increased research in the western Indo-Pacific, recent ichthyological studies continue to reveal high rates of new species discoveries and records. In this study, the presence, general morphology, scale and otolith shape variations, osteology, distribution, and molecular phylogenetic affinity of the banded grouper *Epinephelus radiatus* have been documented/presented from the northwest of the Oman Sea. The morphological and meristic characteristics of specimens were well fited with *E. radiatus*. The scales were cycloid type, being oval/ ovoid, oval/ true oval, and oval/ reversed ovoid in different body regions of fish. The lateral line scale has a wide and long canal extending from the posterior end of the scale to just before the anterior margin, becoming wider in the anterior region. The otolith shape is elliptic-oblong, with a crenate/ irregular dorsal margin, sinuates ventral margin, and dentate and irregular posterior margin. Sulcus groove medium, heterosulcoid; opening ostial. Ostium funnel-like. Cauda tubular, strongly curved, ending close to the ventral margin. Colliculum heteromorph. Crista superior and inferior are well-developed and ridgelike. The vertebral column includes 10 abdominal and 14 caudal vertebrae, with a total vertebral count of 24. DNA barcoding based on mitochondrial COI sequences revealed that some species of the genus *Epinephelus* including *E. radiatus* are nested within the *Mycteroperca* clade, and thus both the genera are not monophyletic.

Keywords: Oman Sea, Biodiversity, DNA barcoding, DNA taxonomy, Otolith and scale morphology, Osteology.

INTRODUCTION

Long-term, consistent, and continuous biodiversity surveys across different regions are crucial for understanding temporal and spatial diversity patterns and for assessing the effectiveness of current conservation measures. Such efforts are vital for revealing how biodiversity changes over time and space, which in turn informs conservation strategies and policies. Despite the significant recent increase in research within the western Indo-Pacific, this region continues to be a hotspot for ichthyological discoveries. New investigations frequently result in the identification of new species and the documentation of previously unrecorded occurrences (Esmaeili et al. 2022). This ongoing trend highlights the rich biodiversity of the area and underscores the necessity for sustained scientific. Hence, documentation of new records and new species, and preparation of a list of fish species at different regional or worldwide scales, and different taxonomic levels (e.g., family, order) are significant for studying marine biodiversity and conservation.

Groupers of the family Epinephelidae comprise a species-rich assemblage of marine reef fishes whose classification has undergone many changes and remains inconsistent across various ranks of the taxonomic hierarchy from species to the family (Ma & Craig 2018). Over the past two decades, a number of molecular phylogenetic studies, using a variety of markers and covering many taxonomic levels, have contributed significantly to clarifying the relationships among groupers and providing the context for a classification scheme that reflects these relationships (see Ma et al. 2016; Ma & Craig 2018). Epinephelid fishes have long been considered as a subfamily of Serranidae which are generally known as groupers and sea basses (Parenti & Randall 2020). The subfamily Epinephelinae was previously divided into five tribes:

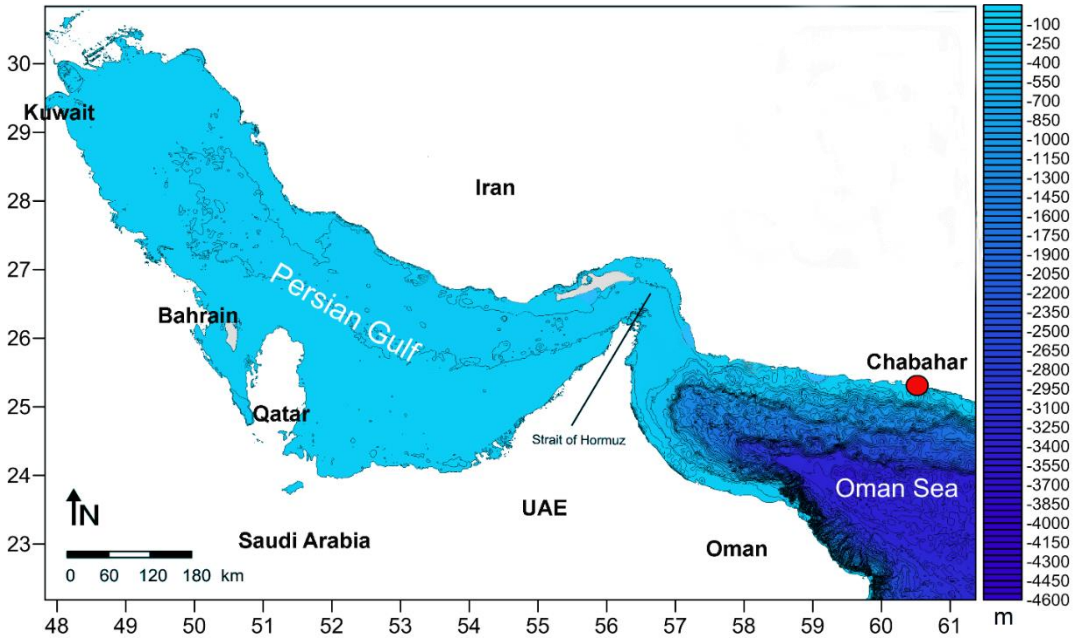

Fig.1. Sampled area of *Epinephelus radiatus,* Chabahar, Iran, Oman Sea.

Epinephelini, Diploprionini, Grammistini, Liopropomini, and Niphonini (Baldwin & Johnson 1993; Parenti & Randall 2020). Later, Heemstra (2022) considered it as a distinct family with 5 subfamilies: Diploprioninae (soapfishes), Epinephelinae (groupers), Grammistinae (soapfishes), Liopropomatinae (basslets) and Pseudogramminae (podges), with about 30 genera and ~237 species; 19 genera and at least 88 species in western Indian Ocean (Heemstra 2022). Fricke et al. (2024) considered the following five families: Serranidae, Anthiadidae, Epinephelidae, Liopropomatidae (Liopropomatinae, and Diploprioninae), and Grammistidae. Here, we follow the arrangement adopted from the online version of Eschmeyer's Catalog of Fishes (Fricke et al. 2024).

Epinephelidae (groupers) includes 16 genera and 177 species (Fricke et al. 2024). They are demersal fishes of tropical, subtropical, and temperate (few species) seas. They inhabit coral reefs, rocky bottoms, or sandy substrates. Found on seagrass beds and mud or sandy bottoms. They are at the top of the alimentary chain, feeding mainly on fish, large crustaceans, and cephalopods. Size: 20-300cm with a maximum weight of 400Kg (Parenti & Randall 2020).

In Iranian waters of the Persian Gulf and the Sea of Oman, 15 species belonging to the genus *Epinephelus* have been reported so far (Eagderi et al. 2019). In this study, we found another species the oblique-banded grouper *E. radiatus* in the Iranian waters of the Oman Sea in the Chabahar region.

This study aims i) to document the further expansion of *E. radiatus* to the western part of the Indian Ocean, ii) to provide a detailed description of the collected specimens, iii) to illustrate the morphology of scale, otolith, and caudal skeleton of *E. radiatus*, iv) to reconstruct its phylogenetic tree, and v) to compare the new findings with the available information.

MATERIAL AND METHODS

In November 2023, three specimens of a species of grouper fish were sampled from the northwest Oman Sea (Chabahar) (N: 25°21´14.1˝ and E: 60°36´04.5˝) of Iran (Fig. 1) at a depth of approximately 30m from coral-associated. The specimens were entangled in fish traps set at a depth of 20-25m over a soft bottom, scattered with rocks. The collected specimens were stored in an ice box and transported to the laboratory for identification and further analysis. In the laboratory, 13 morphometric measurements and 7 meristic counts were taken from the collected species

Fig.2. Lateral view of *Epinephelus radiatus* collected from Chabahar, northwestern Oman Sea.

(Appendix 1- T1) using a measuring caliper nearest to 0.1mm. A digital electric balance was used to measure the weight of sampled specimens up to 0.1g. The specimens were identified as *E. radiatus* according to traditional morphology-based taxonomic keys (Randall & Heemstra 1991; Heemstra & Randall 1993; Baldwin et al. 1994) and color patterns. The examined specimens (SE10-01 to SE10-03) were deposited in the Zoological Museum and Collection of Biology Department of Shiraz University (ZM-CBSU), Shiraz, Iran (Fig. 2).

Scale study: For the preparation of scales, Gholami et al. (2013) and Esmaeili et al. (2019) followed. The scales were removed from seven regions (Fig. 3a). The left side of the fish body along the longitudinal axis was divided into seven regions as follows: region A (the operculular region), region B (the head region), region C (key scale: the third to fourth rows between the dorsal-fin origin and the lateral line), region D (the lateral line), region E (a region in the front of the pectoral fin), region F (a region in the front of the anal fin), and region G (the caudal peduncle region) (Fig. 3b). For each fish specimen, five scales were removed from the selected regions (i.e., 35 scales per specimen: 7 regions × 5 scales per region). The scales were gently removed with fine forceps from the left side in such a way that, while removing the scales, no damage was done. Scales were immediately rinsed in distilled water, cleaned mechanically to remove irrelevant matter using a fine brush, and transferred into 1% KOH solution for approximaly 40 min to remove soft tissues from the surface. The cleaned scales were dehydrated in 30, 50, 70, and 90% ethanol for 20 min respectively, and dried on filter paper (Lippitsch 1990). To avoid curling the margins of the scales, they were kept between the two micro sides for 2-3 days. Thereafter, scales were subjected to digital imaging using a Nikon Eclipse 80i Digital Imaging Head connected to a computer. Digital images were then used for morphological descriptions and measurements (Echreshavi et al. 2021), and for scanning electron microscopy, to avoid curling the margins of the scales, images were immediately mounted on aluminum stubs with double adhesive tape with the dorsal surface upward. Stubs were coated with gold to a thickness of 100 Å in a gold coating unit and various images of the scales were photographed and captured with a TESCAN vega3 instrument (Shiraz University, Iran) at 20 kV.

The terminology of scale features and characters: Specific terms are set to describe the different parts of the scale (Fig. 3c), which we follow Lagler (1947) and Lippitsch (1990) define.

• Circulus/Circuli: usually looking like continuous concentric lines, interrupted by radii or other grooves (Fig. 3c).

• Focus: focus refers to the first part, often central, of the scale to appear in growth (Fig. 3c).

• Radius/Radii: grooves, which usually radiating from focus to margins. Depending on their point of origin

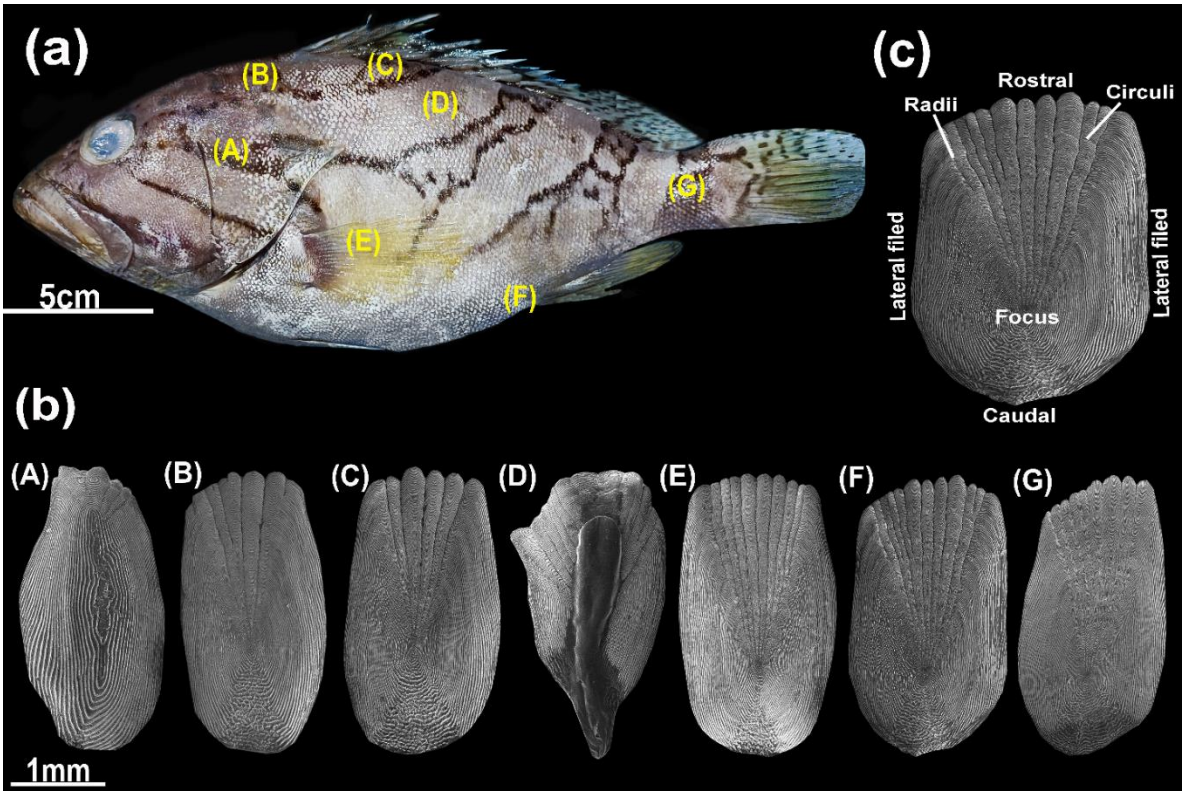

Fig.3. (a) Image of *Epinephelus radiatus* showing seven studied body regions, where scales were removed from the left side of fish, (b) scales of seven body regions (A= the Operculum region, B= the Head region, C= the third to fourth rows between the dorsal-fin origin and the lateral line, D= the lateral line, E= a region in the front of the pectoral fin, F= a region in the front of the anal fin, and G= the caudal peduncle region), (c) terminology of the scale.

in the scale, they are in three types, including primary radii (extend from the focus to margin), secondary radii (begin outward from, not at, focus), and tertiary radii (are positioned between the scale margin and the focus) (Fig. 3c).

• Fields: The area of the scale surface in the anterior, posterior, and two lateral regions (Fig. 3c).

Otolith study: Fishes were dissected under a stereomicroscope (Zeiss Stemi SV6) and the left sagittal otolith of each specimen was extracted and the remaining tissue in a 4% KOH solution for a few minutes. Otoliths were then washed in distilled water and allowed to dry at room temperature before being them in plastic trays. The otoliths were then mounted on an aluminum stub using double-sided sticker tape and coated with a 30-nm layer of gold and Scanning Electron Microscopy (SEM) was used to image the inner face of the left saccular otolith (Fig. 4).

The terminology of otolith characters: Terms are illustrated in Figure 4, and in this study, we followed Smale et al. (1995).

• Margin: shape and sculpturing of otolith edges.

• Sulcus acusticus: a longitudinal depression on the proximal surface of the otolith. It may be divided into ostium (that portion of the sulcus anterior to the neck) and cauda (that portion of the sulcus posterior to the neck).

• Crista: the rim of the sulcus dorsally (crista superior) and ventrally (crista inferior), ranging in development from absent or broken, through to a continuous, extended feature, which in turn may be poorly developed, or well developed, through to a raised, ridge-like margin to the sulcus.

• Rostrum: where present, the portion of the otolith extending ventrally and anteriorly from the excisura notch.

• Antirostrum: where present, the portion of the otolith extending dorsally and anteriorly from the excisura notch.

• Excisura: where present, the opening of the sulcus

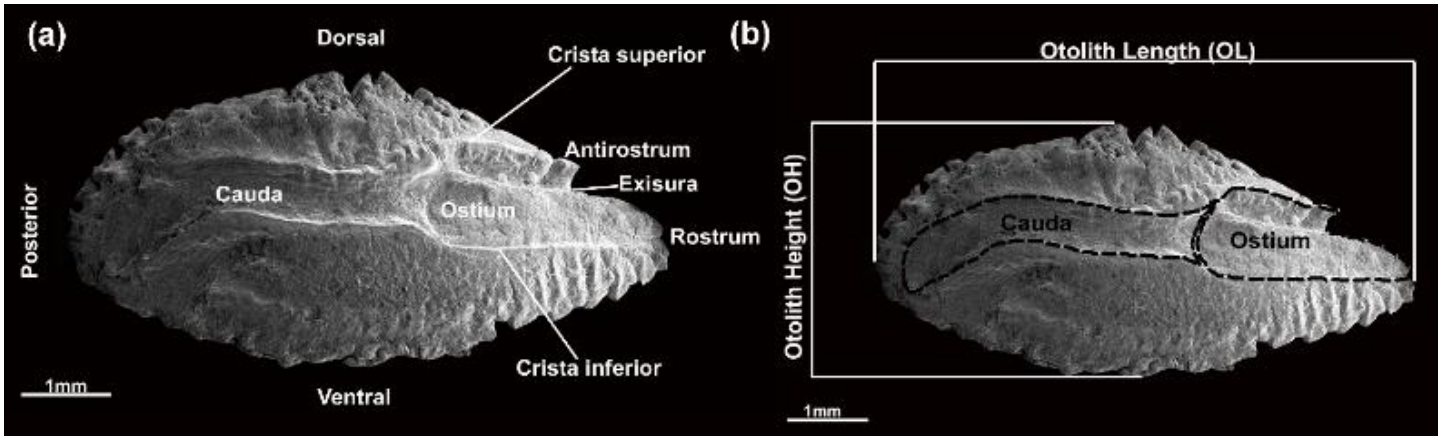

Fig.4. (a) Left sagitta otolith showing the terminology of otolith characters according to Tuset et al. (2008), (b) measurements according to Reichenbacher et al. (2007).

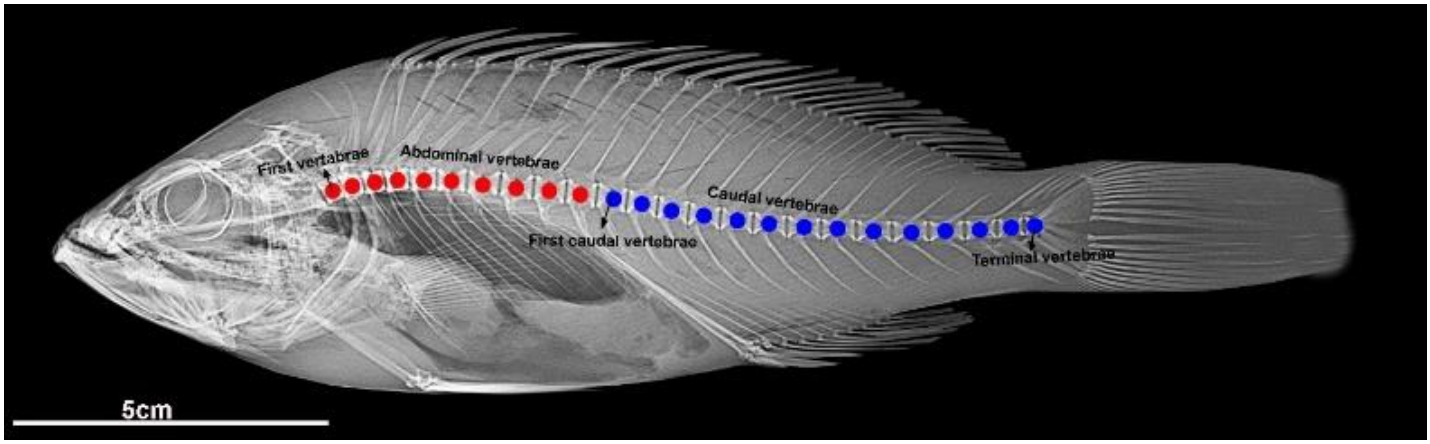

Fig.5. Digital radiographic image of *Epinephelus radiatus* showing the skeleton of the specimen.

onto the otolith anterior margin; often with an associated notch.

Skeleton study: Digital radiographic images were taken from the left side of three specimens using a radiology machine available in the veterinary faculty of Shiraz University, Shiraz, Iran (Fig. 5). The Digital radiograph images were used the osteological study of the caudal skeleton (Fig. 6). The shape of the individual bones of the caudal skeleton is described (see Figs. 5 and 6). The caudal skeleton is formed by the last caudal vertebra (terminal centrum), the uroneural, the hypural plates, the parhypural, and the epurals.

The terminology of the caudal skeleton in this study followed by Hubbs & Lagler (1947), Gosline (1961), Fujita (1990), Kim (2002), Arratia (2008), Schultze & Arratia (2013), and Jawad & Jig (2016) which is explained in the following:

• Terminal centrum: The most posterior centrum of the vertebral column.
• Uroneural: Modified neural arch.
• Hypurals (HYP): The hypural bones are triangular and articulated with the terminal centrum.
• Epural: Modified neural spine.
• Parhypural: It is related to the arch and haemal spine of the terminal centrum and can bear a hypurapophysis.
• Preural vertebra (PU): The neural and haemal spine of the preural vertebra each support a caudal ray at their distal tip. (See Fig. 6).

Molecular study: The right pectoral fin or tissue from below the dorsal fin on the right side of each specimen was removed and preserved in 96% ethanol.

DNA extraction and amplification: Total genomic DNA was extracted using PsPure Genomic DNA Extraction kit and the standard vertebrate DNA

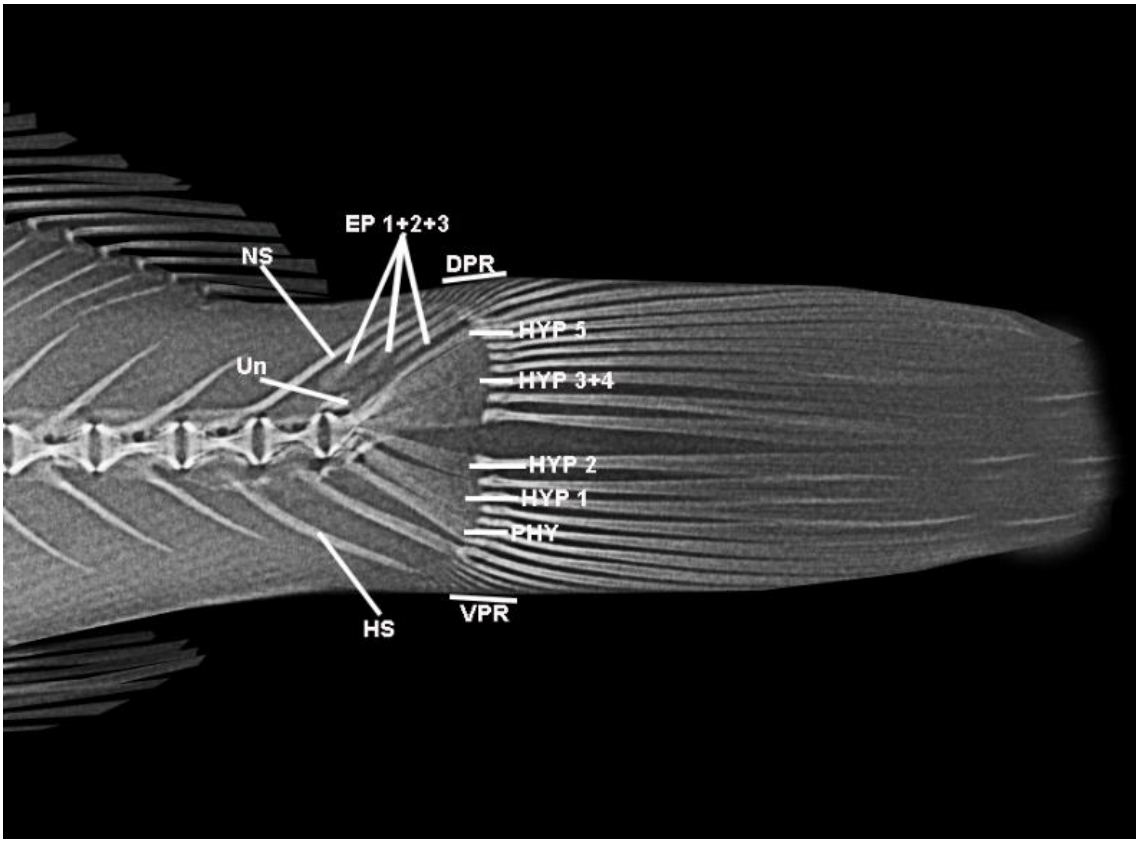

Fig.6. Digital radiographic image of *Epinephelus radiatus* showing the caudal skeleton. EP: Epural, HP: Hypurapophysis, HS: Haemal spine, HYP: Hypural plate, NS: Neural spine, PHY: Parhypural, PU: Preural centrum, TC: Terminal centrum, UN: Uroneural, VPR: Ventral procurrent rays; DPR: Dorsal procurrent rays.

barcode region of the COI (cytochrome oxidase subunit 1) was amplified using primer pairs for COI named FishF1 (5'TCA ACC AAC CAC AAA GAC ATT GGC AC3') and FishR1 (5'TAG ACT TCT GGG TGG CCA AAG AAT CA3') (Ward et al., 2005). The following amplification protocol for COI primer (Fish F1 and Fish R1): 94°C for 1 min for initial denaturing, 35 cycles of 94°C for 30 seconds, 52°C for 45 seconds, and 72°C for 45 seconds, followed by 72°C for 5 minutes as the final extension on a Bioer thermal cycler. The amplification process was performed using Master Mix in a total volume of 25µl containing 12.5µl of a Ready 2X PCR Master Mix (Genet bio, Cat. no. G-2000), 0.5µl of each primer (10pmol/µl), 5µl of the DNA template and 6.5µl dd water. The amplification was performed using a Bioer XP Thermal Cycler (Bioer Technology Co. Ltd., Hangzhou, China). Amplification products were determined by electrophoresis in 1% agarose gel in 0.5X TBE. Purification and sequencing of the PCR products were conducted at Niagene Noor (Tehran, Iran) laboratories with the aforementioned primer pairs. An additional 90 sequences from Epinephelidae were obtained from the NCBI GenBank to achieve an integrative dataset for assessing the phylogenetic position of the *E. radiatus*. The *Liopropoma rubre* sequence was used as an appropriate outgroup (Fig. 7).

Data analyses: The mitochondrial COI sequences of *E. radiatus* were checked via BLAST (Basic Local Alignment Search Tool) searches at the NCBI website (http://www.ncbi.nih.gov). Data processing and edition were performed in BioEdit 7.2.5 (Hall 1999). MEGA11 (Tamura et al. 2013) was used to create a DNA sequence alignment using the ClustalW algorithm. No indications of unexpected stop-codons or nuclear copies of mitochondrial fragments occurred in any sequence. We generated maximum likelihood phylogenetic trees with 10,000 bootstrap replicates using RAxML software 8.2.5 (Stamatakis 2006) under

the GTR+G+I model of nucleotide substitution, with fast bootstrap, and also Bayesian phylogenetic analysis (BI) via the Markov Chain Monte Carlo method (MCMC), with 6,000,000 generations under the most generalizing model (GTR+G+I) using MrBayes 5.3.2 (Huelsenbeck & Ronquist 2001). Sequence divergence values between species were calculated using the Kimura two-parameter (K2P) distance model implemented in MEGA11 (Tamura et al. 2013).

RESULTS

Systematics

Class: Actinopterygii Klein 1885

Order: Perciformes Bleeker 1863

Family: Epinephelidae

Subfamily: Epinephelinae

Genus: *Epinephelus* Bloch 1793

Species: *Epinephelus radiatus* (Day 1868)

The genus *Epinephelus* Bloch 1793

Body oblong to oval, subcylindrical or deep and slightly compressed; body depth 2.3-3.7 in SL; HL 2.1-2.8 in SL. Dorsal fin with 11 spines and 12-19 rays; anal fin with 3 spines and 8-9 rays; caudal fin round, truncate, or slightly emarginated, with 15 branched rays. Preopercle margin rounded or angular, distinctly serrate, but no large antrorse spines on the lower edge. Jaws with bands of small depressible teeth and several canines at front; vomer and palatines with small slender teeth; tongue edentate. Caught with traps, hooks, lines, spears, trawls, and gill nets. About 108 species, 46 in the western Indian Ocean (Heemstra 2022).

E. radiatus (Day 1867)

Serranus radiatus Day 1867: 699 (near Chennai, India).

Epinephelus radiatus: Randall & Heemstra 1991; Craig et al. 2011; Psomadakis et al. 2015.

General morphology: Morphometric and meristic characters of *E. radiatus* are given in Appendix 1- T1, and general morphology in Figure 2. It is distinguished from other congeneric species of the western Indian Ocean by body with some dark spots and 5 irregular curved oblique dark brown bands (with age only the edges remain dark): 1st band from nape to the eye, 2nd band from middle dorsal-fin spines to upper end of gill opening, 3rd and 4th bands from dorsal-fin rays bifurcating ventrally, and 5th band on peduncle.

Dorsal head profile slightly convex, nearly flat interorbital, maxilla reaches to hind orbit margin; lower jaw with two rows of palatine teeth; posterior margin of preopercle serrated and five enlarged serrate at the corner; Body depth 2.6-3 in standard length (fish 11-42cm SL); head length 2.1-2.3 in SL; dorsal-fin spines 13, third dorsal spine longest, dorsal-fin rays 13-15; anal-fin spines 3, second and third anal spines almost equal, anal-fin rays 8; pectoral-fin rays 17-18, pelvic fin not extend to anus; caudal-fin margin convex to moderately rounded; gill rakes 8 or 9/16–18; lateral line scales 52-66; LSS 102-120 (Craig et al. 2011; Heemstra 2022).

The coloration is well described by Heemstra (2022): Juveniles (10-20cm SL) dark brown, with black-edged paler (or white) bands enclosing many black spots. Small juveniles (4-7cm SL) mostly dark green-brown, with dark-edged immaculate white markings (pale interspaces on larger specimens) and no spots; fins mostly hyaline white, except spinous dorsal fin coloured as body. Small adults (20-40cm SL) buff, with 5 oblique dark-edged pale bands with dark spots mainly in series in middle of band (1st band from upper half of eye; 2nd band branching just behind eye, bifurcating on opercle and extending onto spinous dorsal fin; 3rd band or lower branch of 2nd band from end of opercle to last 2 dorsal-fin spines and first few rays; 4th band from rear of dorsal fin, bifurcating towards anal fin; 5th band on peduncle); dark bands also containing scattered black spots and pale blotches; dark line from eye to edge of sub opercle; faint dark band along maxillary groove to edge of interopercle. Large adults (40-50cm SL) with dark-edged bands replaced by series of dark spots (except for dark line running back from lower edge of eye, and no spots on lower third of body); dorsal fin and most of caudal fin also covered with dark spots.

Attains 57cm SL, 79cm TL (7.3kg).

Three samples obtained in this study had an average standard length of 181.60±0.04mm, an average total length of 219.35±0.01mm, and an average total weight of 174.43±0.02g it was (Appendix 2- T2).

Distribution and habitat: *Epinephelus radiatus* is distributed in the Indo-Pacific Ocean from East Africa to Tonga, including Zanzibar, Chagos, Laccadive, St. Brandon Shoals, Maldives, Sri Lanka, India, Nazareth Bank, Sumatra, Fiji (Randall et al. 2003). They live in coral reefs and relatively deep tropical waters of rocky areas. However young *E. radiatus* are mostly found in shallow rocky areas.

Distribution in the western Indian Ocean: Gulf of Oman to India and Sri Lanka, Red Sea and Gulf of Aden, South Africa (KwaZulu-Natal), Comoros, Reunion, Mauritius, and Chagos; elsewhere to southeast India, Sri Lanka, Japan, New Guinea and Australia (Fricke et al. 2024).

IUCN status: Least Concern (LC) (Barreiros 2018).

Scale morphology: The general morphology of the seven region scales of the studied *E. radiatus* is shown in Figure 3b. Scales are usually divided into four parts: rostral, caudal, ventral, and dorsal fields. The scales are cycloid type in (Fig. 3b), being oval/ ovoid (A region), oval/ true oval (B, C, E-G regions), and oval/ reversed ovoid (D region) as given in Figure 3b.

The rostral margin of the scales is scalloped, and the posterior margin is true cycloid (Fig. 3b). Focus is oval and long in A region, and round and small in B, C, E-G regions (Fig. 3b). Focus is positioned as a long oval depression in the middle of the scale (A region), and postero-central (B, C, and E-G regions). Radii are found in the rostral and lateral parts of the scale in the forms of primary, secondary, and tertiary depending on their position on the scale (Fig. 3b). The direction of the rays was parallel. The circuli are distinct and discontinuous in the anterior field, but distinct and continuous in the lateral fields (Fig. 3b).

The lateral line scale with a wide and long canal extending from the posterior end of scale to the area before anterior margin. The anterior part of the canal is wider than the central and posterior parts. The canal bears 3 pores (Fig. 3b).

Otolith morphology: Otolith shape is elliptic-oblong. Dorsal margin crenate/ irregular; ventral margin sinuates; posterior margin dentate and irregular. Sulcus groove medium, heterosulcoid; opening ostial. Ostium funnel-like. Cauda tubular, strongly curved, ending close to the ventral margin. Colliculum heteromorph. Crista superior and inferior well-developed and ridgelike. Anterior region peaked; rostrum broad, medium, pointed; antirostrum poorly defined; excisura wide with a shallow notch (Fig. 4a). Then some otolith parameters were also measured, and their average is OL= 6.66±0.01 mm, OH= 3.39±0.01mm, OWt= 0.041±0.02g (Fig. 4b, Appendix 2- T2).

Axial skeleton morphology: The radiological images taken from the axial skeleton and caudal skeleton of *E. radiatus* species are shown in Figures 5 and 6.

In the axial skeleton, each vertebra consists of a centrum, a neural arch, and a neural spine, except the last vertebra (= terminal centrum, TC) (Fig. 5). The neural spines are well-developed. Abdominal vertebrae, except the first one or two, were associated with ribs; caudal vertebrae (except TC) were characterized by the presence of a closed haemal spine (Fig. 5). There are 10 abdominal and 14 caudal vertebrae in the vertebral column (Fig. 5). In general, each vertebra centrum has a cylinder shape and is amphicoelous.

Caudal skeleton: In this species studied, the caudal skeleton is composed of the preural centra 2 and 3 and their neural and haemal spines, the uroneural, three epurals, the parhypural, the terminal centrum, and the hypural plates. The hypurals are plate-like bones consisting of five elements. Hypurals 1 and 2 separate and hypurals 3 and 4 are usually fused and hypural 5 as well separate and the shape is rectangular; it is narrow at the end (Fig. 6). The terminal centrum is fused to the hypural plates 3 and 4 (Fig. 6). Three separate epurals bones are present; they have unequal lengths and epural 3 is generally shorter (Fig. 6). Epural 1 is usually wider than epurals 2 and 3 and is

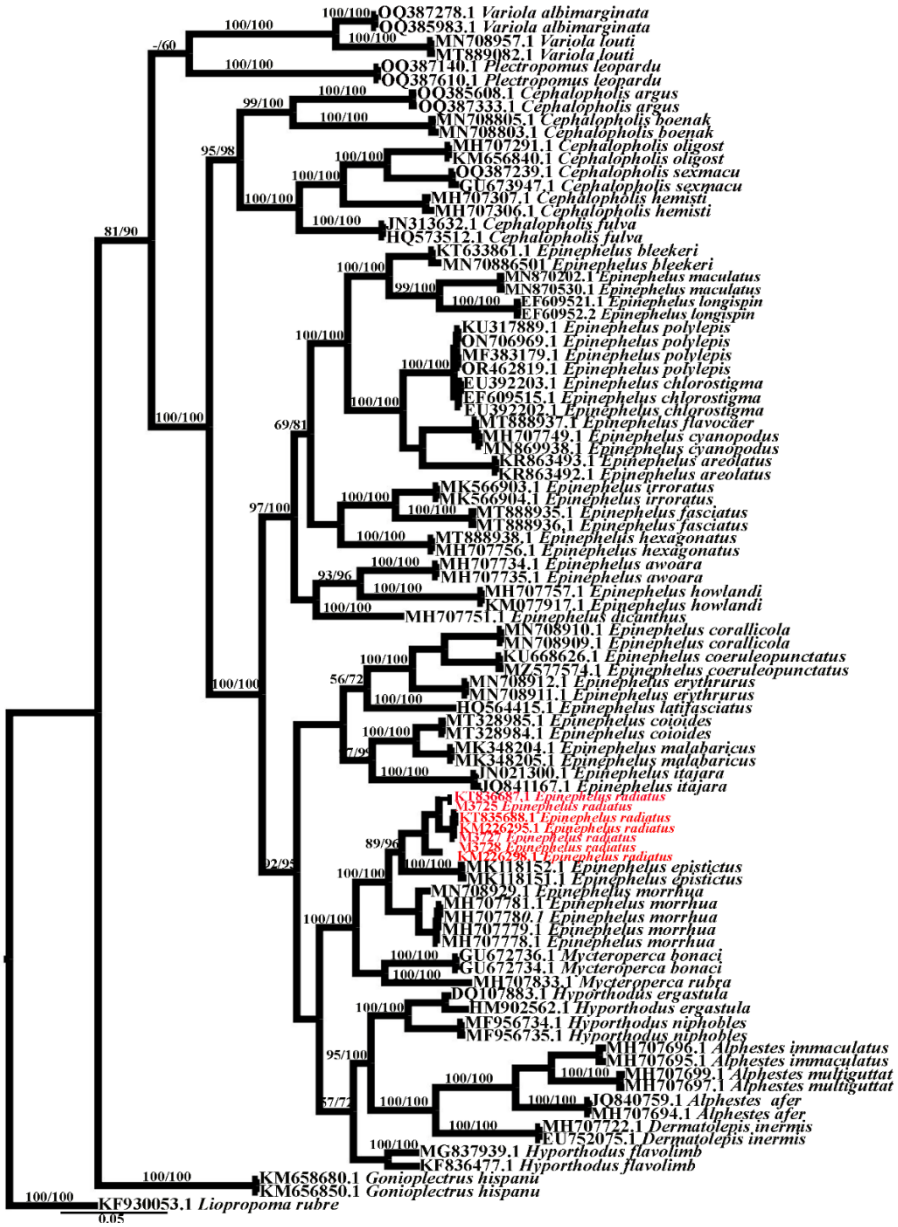

Fig.7. Maximum Likelihood and Bayesian phylogeny reconstructed based on COI gene sequences. The values beside the branches before and after a slash are BI posterior and ML bootstrap probability values, respectively.

wide and paddle-like at the end (Fig. 6). Epural 2 is narrow and straight at the end (Fig. 6), and Epural 3 is narrow and sharp at the end. The parhypural is long and broad, with a straight ventral edge. It has the same length as the hypural plate 1, and it reaches the ventral edge of the fish body (Fig. 6). The uroneural is lying on the dorsal region of the terminal centrum and it is triangular (Fig. 6). The neural spine of the penultimate vertebra 3 (PU3) reaches the dorsal edge of the body (Fig. 6). The haemal spines of PU2 and PU3 are broad and long. The haemal spine of PU2 is rectangular and wide at the end, whereas the haemal spine of PU3 is slightly narrow at the end (Fig. 6).

Phylogenetic relationship: A total of 94 sequences of the several genera had 655 bp after aligning the sequences. Both Maximum likelihood (ML) and Bayesian Inference (BI) were similar in topology and thus only the Maximum likelihood (ML) tree is given here. All species are monophyletic and bootstrap values range from 56% to 100% (Fig. 3). All three *E. radiatus* samples were clustered with high bootstrap values (96%) along with the sequences obtained from

the NCBI gene bank of *E. radiatus* (KT835688.1), (KT835687.1), (KM226298.1), and (KM226295.1) species (Fig. 7).

Genetic distances between grouper species based on consensus sequences of 655 base pairs of COI and the reference sequences related to the K2P model are given in Appendix 3- T3. The maximum genetic distance was between *E. radiatus* and *Variola louti* (22.5%), and the minimum distance was between *E. radiatus* and *Mycteroperca bonaci* (9.1%). Interspecies differences among other species of the Epinephelidae species e.g., between *E. maculatus* and *V. louti* was 24.7%, and *E. chlorostigma* and *E. polylepis* (2%).

DISCUSSION

So far, extensive studies on the diversity of rock and coral fish, in the Iranian waters of the Persian Gulf and the Sea of Oman have not been conducted, and the possibility of new species records and new species arrivals has been suggested for the area (Esmaeili et al. 2022), although some of the documented species may have been overlooked. Thus, continuous long-term monitoring and molecular analysis of fish populations in the region are required to provide further evidence to support such a hypothesis. The surveys in the northwestern Indian Ocean (northern Oman Sea) revealed occurrence of a grouper, *Epinephelus radiatus*. The geographical distribution of species belonging to the Epinephelidae family likely indicates their preference for a specific type of habitat (Hanif et al. 2019). The factors such as the mobility and agility of fish, their ability to navigate physical barriers, and their lack of a fixed home mean they have wide dispersal patterns, necessitating a study of their dispersal (Pinsky et al. 2013). Climate change can also directly affect the population size, survival, and distribution of organisms (Walther et al. 2002; Preuss et al. 2014; Lu et al. 2015; Su et al. 2015; Hanif et al. 2017; Siddik & Hanif 2020).

Ranjan et al. (2017) reported 54 species of Epinephelidae in the waters of India. In the updated list provided by Eagderi et al. (2019), 25 species of groupers belonging to the *Aethaloperca* (1 species), *Anyperodon* (1 species), *Cephalopholis* (4 species), *Epinephelus* (15 species), *Hyporthodus* (1 species), and *Pseudanthias* (3 species) were reported in the Iranian waters of the Persian Gulf and the Sea of Oman. Esmaeili et al. (2022) reported a new record for *Liopropoma randalli* in the Sea of Oman (northwestern Indian Ocean), indicating the potential first occurrence of fish belonging to this family in the Persian Gulf and the Sea of Oman. Presence of *E. radiatus* in neighboring waters (India: Odisha coast) was reported by Sahoo et al. (2020). As Oman Sea sits within the natural distribution range of the newly recorded *E. radiatus* species, thus, the possibility of artificial release can be ignored.

Both morphological and molecular characteristics confirm presence of *E. radiatus* in northwestern Oman Sea. Most identification of species belonging to the Epinephelidae family is based on body coloration, patterns, and markings, as well as morphological and meristic characteristics such as shape, head size, and fin size (Elamin et al. 2011). However, often the body color, lines, and spots present on the body, and some meristic counts obtained for juveniles, differ from those of adult specimens of the same species. In this study, it was found that the body color pattern and morphological characteristics, especially meristic traits, correspond to previous studies by Heemstra and Randall (1993), Han et al. (2014), and Hanif et al. (2021) as given in Table 1.

Genetically, the results presented by Ma et al. (2016) indicate that groupers of the family Epinephelidae originated in what is now the East Atlantic during the mid-Eocene, diverged successively to form six strongly supported main clades, and that the most species-rich clade, which includes the genus *Epinephelus* (including *Epinephelus radiatus*), primarily consists of Indo-Pacific shallow water species. Based on Ma and Craig (2018), the genera *Epinephelus*, and *Mycteroperca* are not monophyletic. According to them, the genus *Epinephelus* is monophyletic if the monotypic *Anyperodon* (*Anyperodon leucogrammicus*) is

included. *Anyperodon* shares 11 dorsal-fin spines and the absence of trisegmental pterygiophores with *Epinephelus*. However, it has been retained in a monotypic genus due to the absence of palatine teeth and its elongated and markedly compressed head and body (Heemstra & Randall 1993). It appears that *Anyperodon* is no more elongated or compressed than any other species of *Epinephelus*. The absence of palatine teeth is thus a curious autapomorphic specialization in the species that is worthy of future investigation; though, it is insufficient to exclude it from *Epinephelus* (Ma & Craig 2018). However, Fricke et al. (2024) did not adhere to the taxonomic proposed by Ma & Craig (2018).

There are controversial debates about the taxonomic status of *E. radiatus* within the genus *Epinephelus*. It has been shown that several species of *Epinephelus* are nested within the clade containing *Mycteroperca* (Ma & Craig 2018). They include *E. albomarginatus, E. andersoni, E. caninus*, *E. cosaretae, E. epistictus*, *E. goreensis*, *E. heniochus*, *E. marginatus*, *E. morrhua*, *E. poecilonotus*, *E. posteli*, and *E. radiatus*. Although the species of *Epinephelus* within the *Mycteroperca* clade have 8-9 anal-fin rays and a rounded caudal fin, several (*E. andersoni, E. goreensis* and *E. posteli*) share the characteristic evenness of body depth typical of *Mycteroperca.* Though morphological synapomorphies have yet to be found, all molecular data provide strong support for the monophyly of the *Mycteroperca* clade of groupers, and hence it seems most prudent to include the 11 species of *Epinephelus* including *E. radiatus* in *Mycteroperca* (Ma & Craig 2018). The type species for *Epinephelus* is *E. fasciatus*, and it is recovered within the *Epinephelus* clade (Ma & Craig 2018). However, Heemstra (2022) and Fricke et al. (2024) have not followed this new taxonomic status for *Mycteroperca radiatus* proposed by Ma & Craig (2018), and consider *E. radiatus* as a valid species. Further morphological and molecular data are needed to clarify the status of *E. radiatus*. Comparative study of scales, otoliths, and caudal skeletons in members of the family Epinephelidae might shed light on the taxonomic status of the genera.

CONCLUSION

In the present study, the existence of a grouper with the oblique band, *E. radiatus* in the Iranian waters of the Oman Sea is confirmed based on both morphological traits (color pattern, morphological and meristic traits) and molecular data (mitochondrial gene COI). Furthermore, morphology of scale, otolith, and skeleton was described in detail. The results of this study help to better understand the biology, taxonomy, morphology, genetic and phylogenetic diversity, as well as the distribution of this species, which is useful for the sustainable management of this grouper species in Iranian marine waters.

ACKNOWLEDGEMENT

This work is based upon research funded by Iran National Science Foundation (INSF) under project No.4014682.

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Iran. J. Ichthyol. (2024) 11(2): 79-92
P-ISSN: 2383-1561; E-ISSN: 2383-0964
http://www.ijichthyol.org

doi: 10.22034/iji.v11i2.1018

مقاله کامل

رویکردهای ریختی-مولکولی حضور گونه‌ای دیگر از هامورماهی، هامور نوار مورب، *Epinephelus radiatus* (ماهیان استخوانی عالی: هامورنما ماهیان) در سواحل شمال شرقی دریای عمان، همراه با یادداشتی بر ریخت‌شناسی فلس، اتولیت و اسکلت ساقه دمی آن

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آزمایشگاه تحقیقاتی ماهی‌شناسی و سیستماتیک مولکولی، بخش جانورشناسی، گروه زیست‌شناسی، دانشکده علوم، دانشگاه شیراز، شیراز، ایران.

چکیده: این مقاله بر اهمیت کاوش‌های مستمر و پایدار در زمینه تنوع زیستی تأکید دارد که برای درک الگوهای تنوع در طول زمان و مکان و ارزیابی تلاش‌های حفاظتی ضروری است. با وجود افزایش پژوهش های مختلف در منطقه غربی اقیانوس هند و آرام، مطالعات اخیر نشان می‌دهد که همچنان نرخ بالایی از کشف گونه‌های جدید و ثبت‌های تازه در این منطقه وجود دارد. در این پژوهش، حضور، ریخت‌شناسی عمومی، گوناگونی در شکل فلس‌ها، اتولیت‌ها و استخوان‌شناسی، پراکنش و تبارشناسی مولکولی ماهی هامور نوار مورب *Epinephelus radiatus* از شمال غربی دریای عمان مستند شده است. ویژگی‌های ریخت‌شناسی و شمارشی نمونه‌ها با *E. radiatus* به‌خوبی منطبق بود. فلس‌ها از نوع گرد (سیکلوئید) بوده و شکل آن‌ها در نواحی مختلف بدن ماهی بیضوی بودند. فلس‌های خط جانبی دارای کانال‌های بلند و پهنی بودند که از انتهای پشتی فلس تا نزدیک حاشیه جلویی امتداد داشته و در قسمت جلویی پهن‌تر شدند. شکل اتولیت بیضوی و کشیده با حاشیه پشتی نامنظم و دندانه‌دار، حاشیه شکمی موج‌دار و حاشیه خلفی نامنظم و دندانه‌دار. شیار سولکوس متوسط و هتروسولکوئید بود و استیوم شبیه به قیف بود. بخش دمی (کودا) لوله‌ای، بسیار خمیده و نزدیک به حاشیه شکمی پایان یافت. کریستای فوقانی و تحتانی به‌خوبی توسعه‌یافته و دارای لبه تیز بودند. ستون فقرات شامل ۱۰ مهره شکمی و ۱۴ مهره دمی بود که در مجموع تعداد مهره‌ها به ۲۴ رسید. بارکدگذاری DNA براساس توالی‌های میتوکندریایی COI نشان داد که برخی گونه‌های جنس *Epinephelus* از جمله *E. radiatus* درون کلاد *Mycteroperca* قرار می‌گیرند و هر دو جنس تک‌تبار نیستند.

کلمات‌کلیدی: دریای عمان، تنوع زیستی، بارکدگذاری DNA، آرایه‌شناسی DNA، ریخت‌شناسی اتولیت و فلس، استخوان‌شناسی.