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Report on the occurrence of Margaretha's Goatfish, *Upeneus margarethae* Uiblein & Heemstra, 2010 (Perciformes: Mullidae) from Gulf of Mannar

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A single specimen of Goatfish with the standard length of 88.43 mm was collected from the Gulf of Mannar, Southeast coast of India on 28.12.2018. The specimen was confirmed as *Upeneus margarethae* Uiblein & Heemstra, 2010, based on the morphometric and partial mitochondrial cytochrome oxidase sub-unit I gene analysis. The presence of the species suggests the extended distributional range of *U. margarethae* in Gulf of Mannar (GoM), Southeast coast of India. Morphometristic characters of the present specimen are being compared with the earlier records of the species.

[Keywords: Goatfish, Gulf of Mannar, Mitochondrial COI gene, Upeneus margarethae]

Introduction

Goatfishes are mainly found in the tropical and subtropical waters and belong to the family Mullidae, which consists of 6 genera and 97 valid species^{1,2}. Goatfishes are ecologically and commercially important group, inhabiting shallow waters and have a strong association with the coral reefs. In general, they are recognized based on the bright coloured body with stripes and presence of two chin barbels. The taxonomic status of goatfish species has dramatically increased in the last decade due to the extensive taxonomic research efforts on the coastal fish diversity¹⁻⁸.

The diagnostic features of family Mullidae are the presence of 7 or 8 spines in dorsal fin, villiform teeth, chin barbels, caudal fin with oblique bars and body with lateral stripes^{2,7-9}. In Mullidae, *Upeneus* is one of the dominant genera comprising 42 species, which is characterised by 7-8 dorsal spines, 13-17 pectoral fin rays, 18-33 total gill rakers on first gill arch, 28-39 scales along lateral line, presence of teeth on vomer, palatines and both jaws and presence/absence of bars on caudal fin^{2,5,8}. Out of the 42 valid species, 37 species are assembled into six phenotypically similar groups taxonomic viz. japonicus, tragula, margarethae, moluccensis, vittatus and stenopsis. Five species are ungrouped^{5,8} and Upeneus margarethae is placed in the "margarethae group" encompassing six species, which are distinguished

from other groups based on the characters including 7-8 dorsal fin spines, absence of dark pigmentation at the tip of first dorsal fin, pectoral fin rays 12-15, gill rakers on first gill arch 21-25, 28-30 lateral line scales and the presence of mid lateral body stripe with or without dark spots⁸.

Upeneus margarethae was first described from the Mozambic⁵ and is further reported from Red Sea, Persian Gulf, Arafura Sea, Madagascar, African coast, Southwest, Southeast and Northwest coasts of India, off Sri Lanka, Andaman Sea, off Myanmar, Thailand and from Australian coast^{5,8}. Morphological variations were also observed between the specimens of *U. margarethae* obtained from Mozambique, Somalia and Madagascar due to the ecological impacts⁵. The current study reports the presence of *U. margarethae* from the Gulf of Mannar, Southeast coast of India by analysing the morphomeristic and genetic characters.

Materials and Methods

Sample collection

On 28^{th} December 2018, a single male specimen of *U. margarethae* was obtained from a commercial trawler at the Gulf of Mannar vicinity at depth of about 40-55 m (8°10'26.32" N 78°28'37.56" E; Fig. 1). The collected specimen was photographed and the muscle tissue was excised and preserved in absolute alcohol for molecular analysis. Digital vernier caliper was used for taking measurements



Fig. 1 — Bathymetry of Gulf of Mannar and capture location of *Upeneus margarethae* (*) Bay of Bengal

with an accuracy of 0.1 mm. Measurements are expressed as proportions of standard length (% of SL). The collected specimens were identified according to Uiblein & Heemstra⁵. Further, the specimen was fixed in 10 % buffered formalin and was subsequently deposited in the National Repository of ICAR-National Bureau of Fish Genetic Resources, Lucknow, India (Accession number: MULUMAR/NBFGR).

Molecular analyses

Extraction of the genomic DNA from muscle tissue was carried out by the Qiagen DNeasy blood and tissue kit following the protocol of the manufacture. The primers Fish F1 (5'—TCA ACC AAC CAC AAA GAC ATT GGC AC—3') and Fish R1 (5'— TAG ACT TCT GGG TGG CCA AAG AAT CA— 3')^(ref. 10) were used for amplifying the COI gene. PCR amplification, sequencing and analysis followed Jayakumar *et al.*¹¹. Mean genetic distances and neighbour joining (NJ) tree were constructed according to Kimura 2-parameter (K2P) model using MEGA version 6^(ref. 12). Mitochondrial COI gene sequence was deposited in the NCBI GenBank.

Results

Morphometric measurements and meristic counts of the specimen are presented in Tables 1 & 2.

Systematics

Order:	Perciformes
Family:	Mullidae Rafinesque, 1815
Genus:	Upeneus Cuvier, 1829
Upeneus mai	rgarethae Uiblein & Heemstra, 2010

Common name: Margaretha's goat fish, (Fig. 2; Table 1 & 2)

Diagnosis

Body elongated with two separate dorsal fins and a pair of unbranched barbels on the chin. Dorsal fin spines 8, rays 9; first spine minute; pectoral fin rays 13; total gill rakers on the first gill arch 6+15 = 21 and 29 lateral line scales. Measurements are in percentage standard length, body depth at dorsal fin origin 24; depth at anus 21; head depth 20.3; head length 30.9; orbit length 6.9; barbel length 18; maxilla length 10; first dorsal fin height 20.5; second dorsal fin height 18.09; length of first dorsal fin base 14; length of second dorsal fin base 14.7; pectoral fin length 21; pectoral fin width 4.9; pelvic fin length 21-82; anal fin height 15.7; anal fin base 10.7; caudal peduncle width 5.5; caudal peduncle depth 10.7 and caudal fin length 28.6 (Tables 1 & 2).

Colour

Head and body portion dorsally red constituting of irregular markings; first dorsal fin pale with two horizontal red stripe; dorsal fin tip pale; 3 red stripes on second dorsal fin; barbels white; body covered with scattered red irregular blotches above and below the lateral line, pale white colouration on ventral part of head and belly; hyaline pectoral fins and pale pelvic fins; 3 bars on upper caudal fin lobe and one bar at base; lower caudal fin lobe with a broad red band (separate bars are not clearly visible as it might have fused to form a red band; bars may be more conspicuous in fresh specimen), single mid-lateral body stripe running through eye to base of caudal fin, which is red coloured from snout tip to eve and yellow from behind eye to base of caudal fin (Fig. 2). Preserved fish become entirely pale brown in colour without any stripes or bars.

Comparisons with congeners

Upeneus margarethae differs from U. luzonius and U. sundaicus, in having higher gill raker counts, lesser lateral line scales and shallower caudal peduncle. Moreover, the mid-lateral stripe starts from the snout of Upeneus margarethae (vs. starts behind the eye of U. sundaicus). Furthermore, U. margarethae can be clearly distinguished from U. taeniopterus, in having lesser lateral line scales, greater eye diameter, lengthier second dorsal, pelvic and pectoral fins. Upeneus margarethae can be distinguished from U. tragula by longer pectoral fin, shorter jaws, paler

Table 1 — Morphometric characters (in %SL) of <i>Upeneus margarethae</i> , off Gulf of Mannar in comparison with holotype and previous records								
Morphometric characters in % SL	Present study	Uiblein & Heemstra ⁵ Holotype	Uiblein <i>et al.</i> ⁸ Eastern Indian Ocean and N Australia (n = 22)		Uiblein <i>et al.</i> ⁸ Western Indian Ocean Proper (n = 44)		Uiblein <i>et al.</i> ⁸ Red Sea (n = 21)	
			Min	Max	Min	Max	Min	Max
Standard Length (SL) mm	88.43	82	81	117	78	80	67	116
Body depth at first dorsal	24.6	26	24	27	25	26	23	26
Body depth at anal	21.2	24	21	23	20	20	20	24
Caudal peduncle depth	10.7	11	10	12	10	11	10	11
Caudal peduncle width	5.5	4.8	3.8	5.3	3.6	4.1	3.5	4.5
Interorbital length	8.1	8.2	7.6	8.8	7.6	8.4	7.5	8.9
Head length	31	30	27	30	28	31	27	29
Snout length	11.5	12	10	12	11	12	10	12
Postorbital length	11.6	12	11	13	11	12	11	12
Orbit length	7.8	8.7	6.7	8	7.3	7.4	6.2	8.1
Upper jaw length	10.2	11	10	12	11	11	9.8	12
Lower jaw length	8.6	10	8.9	12	10	10	9.5	11
Barbel length	18.1	17	16	20	18	19	15	18
First predorsal length	34.5	40	34	40	37	38	34	38
Second predorsal length	62.2	67	61	67	62	65	60	67
Inter dorsal distance	16.3	15	12	16	13	15	12	16
Caudal-peduncle length	22.7	23	22	25	24	24	22	25
Preanal length	63.8	66	61	68	64	66	61	69
Prepelvic length	31.7	29	30	35	33	34	29	33
Prepectoral length	28.6	31	28	32	30	33	28	31
Length of first dorsal fin base	14	15	14	17	14	15	14	18
Length of second dorsal fin base	14.7	15	12	15	13	13	13	16
Length of anal fin base	10.7	12	9.8	13	11	11	11	12
Pelvic fin length	21.8	23	20	23	21	22	20	22
Pectoral fin length	21.	23	20	23	21	22	19	22
Pectoral fin width	4.9	4.8	4.2	5.2	4.9	5.2	4.1	5.1
First dorsal fin height	20.6	21	18	22	20	22	20	23
Second dorsal fin height	18.1	18	15	18	16	16	16	19

Table 1 — Morphometric characters (i	n %SL) of	f Upeneus	marga	<i>irethae</i> , o	off Gulf of Mannai	in comparison with
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Table 2 — Meristic characters of Upeneus margarethae in comparison with holotype, holotype and previous records

Meristic Characters	Present study	Uiblein & Heemstra ⁵	Uiblein <i>et al.</i> ⁸ Western Indian	Uiblein <i>et al.</i> ⁸ Red Sea $(n - 21)$	Uiblein <i>et al.</i> ⁸ Eastern Indian Ocean and N	
		Holotype	Ocean Proper	(II = 21)	Australia ($n = 22$)	
Dorsal fin spines	8	8	8	8	8	
Pectoral fin rays	13	14	13-14	13-14	13-15	
Rudimentary gill rakers on upper limb	3	3	2-5	2	2-4	
Developed gill rakers on upper limb	3	3	2-4	2-3	2-3	
Total gill rakers on upper limb	6	6	5-7	4-5	5-6	
Rudimentary gill rakers on lower limb	4	5	3-6	4	4-6	
Developed gill rakers on lower limb	11	12	11-14	13	11-12	
Total gill rakers on lower limb	16	17	16-18	17	15-18	
Total gill rakers	21	23	21-24	21-22	21-24	
Lateral line scales	29	28	28-30	29-30	29-30	



Fig. 3 — Neighbour-Joining tree constructed for Upeneus margarethae from Gulf of Mannar with available data of COI gene

dorsal fin tips and from presence of yellow, beige or orange body stripe on lateral side (vs. brown to black in U. tragula). Upeneus guttatus is distinguished from U. margarethae by having 7 spines in dorsal fin (vs. 8 in U. margarethae), subequal length of pectoral and pelvic fins and absence of lateral body stripe (vs. present in U. margarethae). Upeneus vittatus differs from U. margarethae, in having higher gill raker counts and presence of 3 to 4 stripes on lateral side of body for fresh fish (vs. one lateral stripe on body in U. *margarethae*). Upeneus moluccensis and U. sulphureus differ from U. margarethae with the absence of bars on the lower lobe of caudal fin (vs. present in U. margarethae). Furthermore, Upeneus sulphureus differs in having two conspicuous yellow lateral body stripes and black tipped first dorsal fin (vs. dorsal fin tip pale in U. margarethae) and U. moluccensis has 6-8 red bars on upper caudal lobe (vs. 4-5 bars on upper caudal lobe in U. margarethae).

Molecular taxonomic analysis

The COI gene sequence of the present specimen constituting 655 base pairs is being deposited in GenBank (Accession no: MK562407). The sequence generated in the present study was compared with sequences of the species from other regions retrieved from GenBank. The overall intra-species genetic divergence was 0.9 % which is very well within the acceptable range. The phylogenetic tree constructed using Neighbor-Joining model with 1000 bootstrap (Fig. 3) also confirmed the specimen as *U. margarethae*.

Discussion

Taxonomic status of many species of the family Mullidae is still ambiguous pertaining to prominent differentiating characters. The counts of dorsal fin spines, pectoral fin rays, gill rakers and lateral line scale are important in species differentiation of this family^{13,14}. The species also exhibit ontogenetic variation in body colour and morphometrics leading to difficulty in identification⁵.

Fourteen species of goatfishes were described by Day¹⁵ from the Indian Ocean, but he raised concerns regarding the specific identity of few species and urged for an in-depth study. Weber & Beaufort¹⁶ documented 28 goatfish species from the Indo-Australian Archipelago mentioning uncertainty in the identification of three species. Thomas¹⁷ reported 19 goatfish species from the Indian waters. Later, Talwar¹⁸ compiled details of about 20 goatfish species from India on the basis of the study carried out by Thomas¹⁷. Fourteen species were documented from Andaman and Nicobar Islands by Rajan *et al*¹⁹. Hitherto, three genera and eighteen species of goatfishes are known from the Indian waters²⁰. Ten species of the genus Upeneus have been reported so far from India⁸ and nine species are reported from Gulf of Mannar, which includes Upeneus luzonius, U. vittatus, U. tragula, U. moluccensis, U. sulphureus, U. bensasi, U. oligospilus, U. taeniopterus, and U. sundaicus²¹. This study reports the presence of U. margarethae from Gulf of Mannar, Southeastern coast of India, adding to the ichthyofaunal diversity of the region and suggests its extended distribution along the eastern Indian coast.

The morphomeristic characters of Upeneus margarethae are in agreement with the ranges of holotype and specimens collected from the Indian Ocean, Red Sea and Australia⁸. In order to support the limited information on taxonomic features of this species, molecular analysis was incorporated in this study. Mitochondrial DNA which has faster evolutionary rate than nuclear DNA is widely used for phylogenetic studies, and it is also used for differentiating the closely related species²². In the current study, the specimen is confirmed as U. margarethae with COI sequences and phylogenetic tree constructed by Neighbor-Joining model through 1000 bootstrap was used for comparing sequences of species recorded from different regions. It is revealed that the specimen from India forms a robust monophyletic clade with Malayian specimens and falls far with the Australian specimen. The molecular analysis also revealed the collected specimen to be U. margarethae with acceptable level of intra-specific level of genetic divergence values, when compared with available molecular data of the species. A comprehensive integrated approach is indispensable to understand the distinction and relatedness among different species and to unravel the diversity and

evolutionary differences in the genus. The paucity in taxonomy and systematics of goat fishes in the Indian waters warrants extensive study in the area. The current record brings new insight and implications to conserve the species in Indian waters.

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Conflict of Interest

The authors declare that they have no conflict of interest.

Author Contributions

AK conceived the concept, performed molecular analysis and prepared the manuscript. SJ helped in laboratory work and morphological analysis. TJTK performed taxonomic investigation, morphological analysis and manuscript correction. TTAK carried out specimen collection and manuscript edition. KKL provided the guidance and support for carrying out the research.

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