

NEW RECORDS OF THE GOATFISH, *UPENEUS VITTATUS* (FORSSKAL 1775) AND *UPENEUS SUPRAVITTATUS* (UIBLEIN AND HEEMSTRA 2010) (PERCIFORMES, MULLIDAE), FROM SAINT MARTIN'S ISLAND IN THE BAY OF BENGAL, BANGLADESH

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Abstract

Genus *Upeneus* distributed in Indo-Pacific, South Africa, north to southern Japan and south to New Caledonia. Although there is difficulty in identifying accurately goatfish solely on the basis of morphology, *U. vittatus* and *U. supravittatus* were confirmed by both using morphological characters and DNA barcoding cytochrome oxidase I subunit (COI) approach for the first time in St. Martin's Island, Bay of Bengal, Bangladesh. Increased number of species of the genus *Upeneus* was found in Bangladesh from 3 to 5 and extended the distribution range.

Key words: Coral ecosystem, Cytochrome oxidase I subunit (COI), Morphometrics, New record, *Upeneus vittatus*, *U. supravittatus*

Introduction

The goatfishes (Perciformes: Mullidae) are characterized by a pair of long barbels at the front of the chin, two well-separated dorsal fins with 7 or 8 spines and a deeply forked caudal fin. The family Mullidae consisted of 6 genera that are differentiated mainly by dentition as shown in the key of Lachner in Schultz and collaborators (1960). Species of *Upeneus* are only ones with teeth in both jaws and on the vomer and palatines. There are 29 species of the genus *Upeneus* (Uiblein and Heemstra 2011, Yamashita *et al.* 2011) found in shallow coastal waters above 100 m. The yellow striped goatfish, *Upeneus vittatus* (Kuitert and Tonzuka 2001) is a marine, brackish, coral reef associated fish species. Sandy lagoons, sheltered coastal waters and also muddy bottoms (Fischer *et al.* 1990) are preferred habitats of *U. vittatus*. This species is euryhaline (Lewis and Pring 1986), benthopelagic (Mundy 2005) often found in groups (Lieske and Myers 1994) and feeds primarily on small crustaceans (Sousa and Dias 1981). The long fin goatfish, *U. supravittatus* (Uiblein and Heemstra 2010) is a marine, coral reef associated fish species. They use their two sensitive chin barbels to dig into the sediment to locate food items. Invertebrates such as small crustaceans and worms are hunted by small goatfish that forage on the bottom in loose schools. In contrast, large individuals (20 - 40 cm in length) also feed on small fishes (Sea-Ex). At present, only three species of this genus, *U. sulphureus* (Tomascik 1997), *U. sundaicus* and *U. tragula* (Rahman *et al.* 2009) have been recorded in Bangladesh and studied

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only on the basis of morphological characters. However, specimens of *Upeneus* are often difficult to identify to species, as a promising alternative to the traditional species identification based on morphological characters, partial cytochrome c oxidase subunit I (COI) sequences (DNA barcodes) have been suggested for standardized and routine species identification (Hebert *et al.* 2003). Newly generated DNA barcodes can be analyzed together with available data (shown in bold face overleaf) to resolve taxonomic conflicts. Although barcodes for almost one-third (10,267) of all described fish species are already available (Ward 2012), the permanent addition of new barcode data is indispensable to increase the taxonomic resolution. The present study to identify *U. vittatus* and *U. supravittatus* was based on morphomeristics as well as DNA barcoding for the first time in Bangladesh, and distinguished from each other and also from other *Upeneus* spp. recorded before in Bangladesh.

Materials and Methods

Sample collection: Fish specimens were collected on 14 December, 2015 from local fishermen of St. Martin's Island (Fig. 1) (20° 34'N - 20° 38.8'N and 92°18'E - 92°20.8'E) who collected these fishes using gear named ber jal which is one kind of seine net and not destructive. Specimens were collected in full accordance with local government regulation, and in compliance with appropriate animal care standards. After that specimens were carried with cooling box to -20°C in Fisheries laboratory, Department of Zoology, Jagannath University, Dhaka until further study. After study, all specimens were deposited in the museum of Zoology Department, Jagannath University as voucher specimen with registration number *Upeneus vittatus*-F1215SM02 and *Upeneus supravittatus*-F1215SM03.

Morphological studies: Morphometric and meristic characters were compared with previous records by Uiblein and Heemstra (2010) from Western Indian Ocean and other 3 species of *Upeneus* already recorded from Bangladesh water. Lengths were measured in cm scale and weight in gm. The meristic abbreviations used in this study are as follows: D₁, first dorsal fin; D₂, second dorsal fin; P, pectoral fin; V, pelvic fin; A, anal fin; C, caudal fin; Li, lateral-line scale; aLi, transverse scale above lateral-line; bLi, transverse scale below lateral-line; Br, branchiostegal ray. Detailed morphomeristics study was carried out.

Molecular phylogenetic studies: Intra- and inter-specific genetic distances were determined using a molecular marker cytochrome oxidase subunit I (COI) gene of mitochondrial DNA. Muscle tissue was isolated from just below the dorsal fin. DNA was extracted by TIANamp Marine Animal DNA Kit. Approximately 700 bp were amplified from the 5' region of the COI gene using C FishF1t1/C FishR1t1 primer cocktails (Ivanova *et al.* 2007) due to its high effectiveness in generating amplicons that sequence cleanly for the DNA barcode region of diverse fish taxa and other groups of vertebrate. There are two primers (FishF2 t1/VF2 t1) for cocktail C FishF1t1 and

also two primers (FishR2 t1/FR1d t1) for C FishR1t1. To facilitate sequencing of products, all PCR primers were tailed with M13 sequences. The primer's nucleotide sequences were:

VF2_t1-5'-***TGTA**AAACGACGGCCAGTCAACCAACCACAAAGACATTGGCC -3'

FishF2_t1-5'-***TGTA**AAACGACGGCCAGTCGACTAATCATAAAGATATCGGCAC-3'

FishR2_t1-5'-***CAGG**AAACAGCTATGACACTTCAGGGTGACCGAAGAATCAGA-3'

FR1d_t1-5'-***CAGG**AAACAGCTATGACACCTCAGGGTGTCCGAARAAYCARAA-3'

[*The M13 primer sequence in bold face]

The PCR reactions were carried out in 20 µl reaction mixture containing 6.0 µl of distilled water, 10 µl of master mix, 1 µl of each primer (5 µmol/l) and 2 µl of DNA template. The thermal regime consisted of an initial step of 2 min at 94°C followed by 35 cycles of 30 sec at 94°C, 40 sec at 52°C, and 1 min at 72°C, followed in turn by 10 min at 72°C. Then soak at 4°C. PCR products were purified by using QIAquick PCR purification kit. After purification, the products were sequenced in both directions by using commercial sequencer, Macrogen, Korea. Sequences were manually edited using the software Chromas Lite. For phylogenetic analysis, COI sequences of same species and species in the same genus were downloaded from GenBank. The species and their GenBank accession numbers are following: KX024776 for *U. vittatus*; KR057896 for *U. supravittatus*; KP293705, KP293708 for *U. suahelicus*; EF607613, EF607614 for *U. tragula*; JN313348 for *U. sulphureus* and FJ237883 for *Parupeneus indicus*. The software MEGA 6 (Tamura *et al.* 2013) was used for alignment and construction of neighbor-joining tree (Saitou and Nei 1987) on the basis of evolutionary distances calculated using the Kimura two parameter (K2P) model (Kimura 1980), where bootstrapping replications were 1000. Molecular phylogenetic analysis was conducted in the Aquatic Biodiversity Research Laboratory, Department of Fisheries, Sher-e-Bangla Agricultural University, Dhaka-1207, Bangladesh.

Results and Discussion

Upeneus vittatus (Forsskal 1775)

Identification: The morphometric measurements of this species are provided in Table 1 and meristics in Table 2. Body elongated, laterally compressed, dorsal profile convex slightly and moderate in size. Head medium and obtuse. Large, rounded, highly protractile, superior mouth placed sub-terminally with large mouth gap. Unequal jaws with cardiform teeth, also present on palatine and vomer. Two barbels present on chin.

Large operculum with flap and large opercular opening. Lateral line is single, complete and curved. Scales are ctenoid, rhomboidal, large and overlapping. Dorsal fin double, separate, medium and situated at the middle of the body, which starts from the middle of pectoral fin and reaches up to the just opposite to end of anal fin, having spines in front. Pectoral fin large, wide spread and placed ventro-laterally. Pelvic fin moderate in

size and present at the forward. Anal fin medium, contains spines in front and reaches up to the opposite of 2nd dorsal fin. Caudal fin forked and homocercal type.

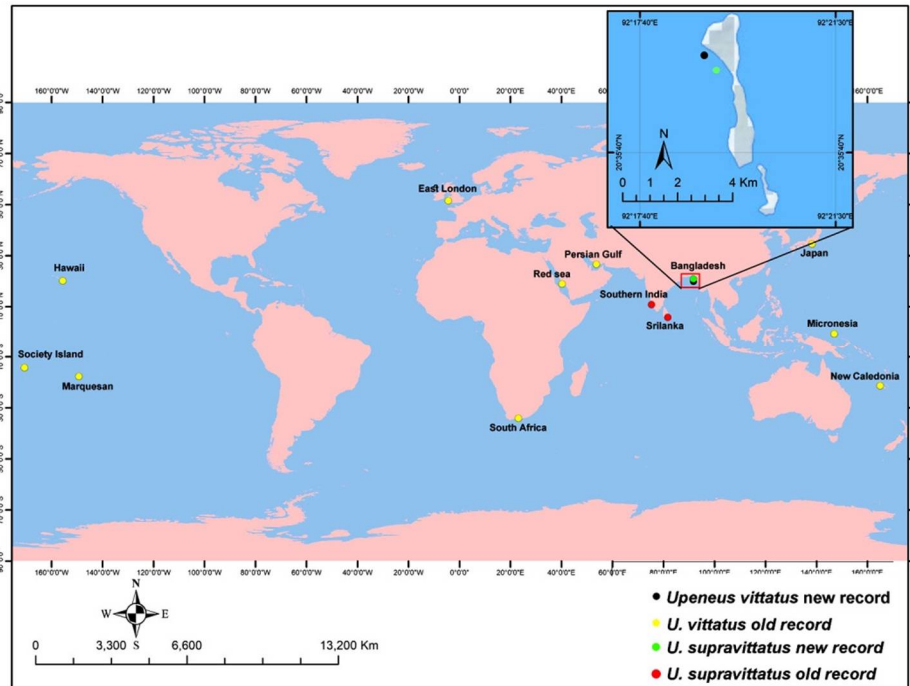


Fig. 1. Location of new and old record of *Upeneus vittatus* and *U. supravittatus* species in the St. Martin's Island, Bay of Bengal, Bangladesh.

Coloration: Body reddish dorsally, silver ventrally in fresh fish. Four golden stripes present, among them 2 mid lateral, one starts from eye and extends up to base of caudal fin, attaches the adjacent bar of upper caudal fin lobe and the other stripe starts from below of pectoral fin base and extends up to caudal peduncle and joined by adjacent bar of lower caudal fin lobe; 2 dorsolateral stripes, where lower one well defined, starts from operculum and extends up to behind of second dorsal fin, the upper one ambiguous and much shorter, starts from below of first dorsal fin origin and surrounded dorsally by a horizontal series of pale spots (Fig. 2a). Two yellow stripes present on white membrane in both dorsal fin and a broad black spot on the tip of 1st dorsal fin anteriorly, the straight length of the black spot similar in size to broadness of broadest lower caudal fin lobe bar (Fig. 2c); reddish pectoral, brown pelvic, reddish anal fin and yellowish patches along pelvic and anal fin bases; five yellow black oblique bars on upper lobe of caudal fin and 3 on lower (Fig. 2e) those increasing

distally in width with the widest distal-most bar black or dark brown, while the other bars are yellow blackish; broadness of broadest lower caudal fin lobe bar and/or light colored gap between bars that are most distally located equal to or larger than eye diameter; light colored tip of lower fin lobe. Barbels are white.

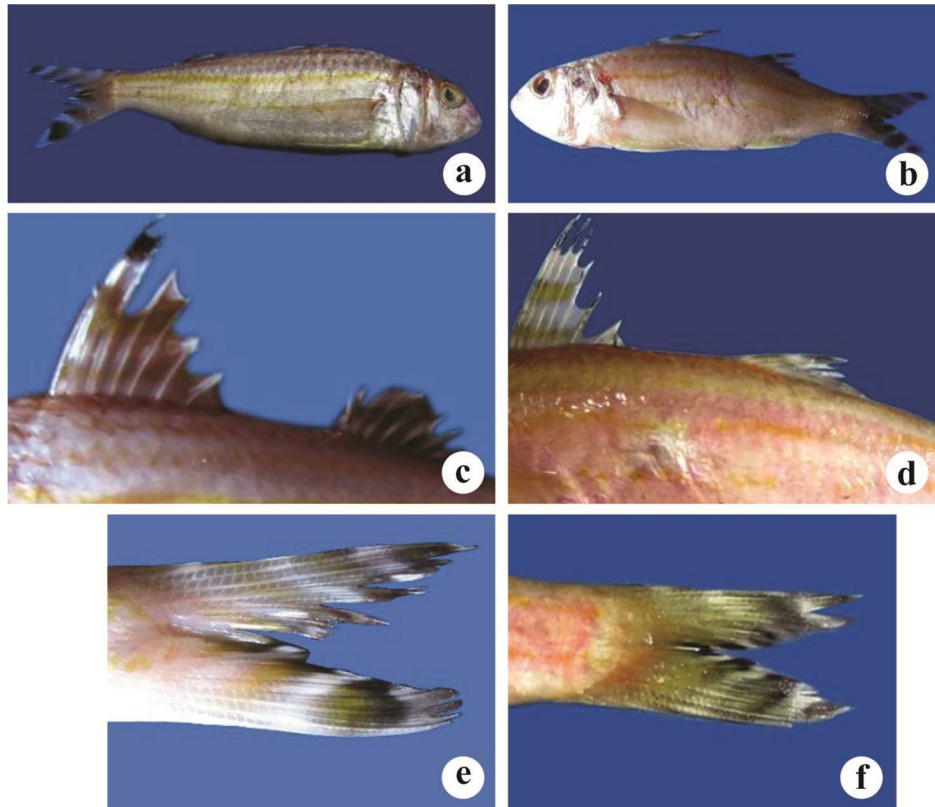


Fig. 2. (a) *Upeneus vittatus* (10.6 cm SL), (b) *Upeneus supravittatus* (11.1 cm SL), (c) dorsal fin of *Upeneus vittatus*, (d) dorsal fin *Upeneus supravittatus*, (e) caudal fin of *Upeneus vittatus* and (f) caudal fin of *Upeneus supravittatus*.

***Upeneus supravittatus* (Uiblein and Heemstra 2010)**

Identification: The morphometric measurements of this species are provided in Table 3 and meristics in Table 4. Body elongated, laterally compressed, dorsal profile convex slightly and moderate in size. Head medium and obtuse. Large, rounded, highly protractile, superior mouth placed sub-terminally with large mouth gap. Unequal jaws with cardiform teeth, also present on palatine and vomer. Two barbels present on chin. Large operculum with flap and large opercular opening. Lateral line single, complete

and curved. Scales are ctenoid, rhomboidal, large and overlapping. Dorsal fin double, separate, medium and situated at the middle of the body, which starts from the middle of pectoral fin and reaches upto the just opposite to end of anal fin, having spines in front. Pectoral fin large, wide spread and placed ventro-laterally. Pelvic fin moderate in size and present at the forward. Anal fin medium, contains spines in front and reaches upto the opposite of 2nd dorsal fin. Caudal fin forked and heterocercal, lower lobe slightly shorter than upper.

Table 1. Morphometric measurements of *Upeneus vittatus* from Bangladesh and comparative materials reported from other places.

Characteristics	Measurements (Present study)	Uiblein and Heemstra (2010)	Characteristics	Measurements (Present study)	Uiblein and Heemstra (2010)
Percentage of standard length					
Head length	35.85	-	Longest anal spine	10.38 (1 st)	-
Body depth	29.25	-	Longest anal fin ray length	16.98	15-16
Body width	17.92	-	Pectoral fin base length	5.66	-
Caudal peduncle depth	9.43	-	Longest pectoral fin ray length	25.47	22-24
Caudal peduncle length	18.87	-	Pelvic fin base length	4.71	-
Caudal concavity	20.75	-	Longest pelvic fin ray length	19.81	18-21
Caudal fin base length	12.26	-	Barbel length	19.81	17-21
Predorsal length	44.34	-	Inter-orbital length	9.43	-
Preanal length	73.58	-	Preorbital length	12.26	-
Prepectoral length	44.34	-	Post-orbital length	14.15	-
Prepelvic length	37.74	-	Eye diameter	9.43	7.0-8.7
First dorsal fin height	23.5	22-25	Snout length	7.56	-
Dorsal fin base length (1 st)	21.70	-	Upper jaw length	14.15	11-13
Dorsal fin base length (2 nd)	14.15	-	Lower jaw length	9.43	-
Longest dorsal spine	23.58(1 st)	-	Jaw gap	15.09	-
Anal fin base length	12.26	-			

Table 2. Meristic features of *Upeneus vittatus* and comparative materials reported from other places.

Characteristics	Present study	Uiblein and Heemstra (2010)	Kuiter and Tonozuka (2001)	FAO (1983a)	FAO (1983b)	FAO (1983c)
D ₁	VII	-	VIII	VII-VIII	-	-
D ₂	I/8 br	-	-	I/9 br	-	-
P	14	-	-	13 to 17	-	-
V	I/5 br	-	-	I/5	-	-
A	I/6 br	-	-	-	I/6-7	-
C	13 br	-	-	-	-	13 br
L _i	37	36-38	-	-	-	-
aLi	2	-	-	-	-	-
bLi	6	-	-	-	-	-
Br	2 pair	-	-	-	-	-

Table 3. Morphometric measurements of *Upeneus supravittatus* from Bangladesh and comparative materials reported from other places.

Characteristics	Measurements (Present study)	Uiblein and Heemstra (2010)	Characteristics	Measurements (Present study)	Uiblein and Heemstra (2010)
Percentage of standard length					
Head length	33.33	30-33	Anal fin base length	11.71	-
Body depth	27.93	26-29	Longest anal fin ray length	15.32	14-17
Body width	16.22	-	Pectoral fin base length	6.31	-
Caudal peduncle depth	10.81	9.9-11	Longest pectoral fin ray length	26.13	25-28
Caudal peduncle length	19.82	-	Pelvic fin base length	5.41	-
Caudal concavity	13.51	-	Longest pelvic fin ray length	19.82	18-21
Caudal fin base length	11.71	-	Barbel length	19.82	19-23
Predorsal length	42.34	-	Interorbital length	9.0	-
Preanal length	72.07	-	Preorbital length	11.71	-
Prepectoral length	34.23	-	Postorbital length	13.51	-
Prepelvic length	36.04	-	Eye diameter	8.12	6.8-8.5
First dorsal fin height	26.1	23-26	Snout length	6.31	-
Dorsal fin base length (1 st)	17.12	-	Upper jaw length	12.61	12-14
Dorsal fin base length (2 nd)	14.41	-	Lower jaw length	12.61	-
Longest dorsal spine	26.13 (2 nd)	-	Jaw gap	9.0	-

Table 4. Meristic features of *Upeneus supravittatus* and comparative materials reported from other places.

Characteristics	Present study	Uiblein and Heemstra (2010)	Kuiter and Tonzuka (2001)	FAO (1983a)	FAO (1983b)	FAO (1983c)
D ₁	VII	-	VIII	VII-VIII	-	-
D ₂	I/8 br	-	-	I/9 br	-	-
P	14	-	-	16 to 17	-	-
V	I/5 br	-	-	I/5	-	-
A	I/6 br	-	-	-	I/6-7	-
C	17 br	-	-	-	-	13 br
L _i	35	34-36	-	-	-	-
aLi	2	-	-	-	-	-
bLi	6	-	-	-	-	-
Br	2 pair	-	-	-	-	-

Coloration: Body reddish dorsally, silver ventrally in fresh fish which become pale after preservation. Two yellow stripes present on body (Fig. 2b), one starts from behind head and extends up to base of caudal fin, attaches the adjacent bar of upper caudal fin lobe and the other stripe starts from below of pectoral fin base and extends up to caudal peduncle and joined by adjacent bar of lower caudal fin lobe. In both dorsal fin 2 yellow stripes present on white membrane and the tip of first dorsal fin black (Fig. 2d). Reddish pectoral, brown pelvic, reddish anal fin and yellowish patches along pelvic and anal-fin bases; total 9 bars on caudal fin, 5 yellow blackish bars on upper caudal-fin lobe, 4 bars on lower lobe, increasing distally in width (Fig. 2f). Barbels are white.

Although *U. vittatus* and *U. supravittatus* at a glance are very similar but have distinct morphological difference and also clearly different from other three *Upeneus* spp. such as *U. sulphureus*, *U. sundaicus* and *U. tragula* which were recorded before in Bangladesh. So, comparative morphological difference among these five *Upeneus* spp. are summarized in Table 5.

Sequence analysis of the COI gene: COI gene fragments of single *U. vittatus* and *U. supravittatus* individual were amplified. Both the COI sequences were submitted to GenBank with the following accession numbers: MG099705 for *U. vittatus* and MG196640 for *U. supravittatus*. Including the eight downloaded COI sequences of family Mullidae, totally ten sequences were used in the analysis and their accession number with sampling location were shown in the associated figure. COI sequences of *U. vittatus* and *U. supravittatus* showed 100% similarity with COI sequences of the respected species in NCBI (<https://blast.ncbi.nlm.nih.gov/Blast.cgi>). The mean interspecies evolutionary divergence was 0.2 - 16.3% between *U. vittatus* and other five species of family Mullidae while 0.2 - 16.1% between *U. supravittatus* and other five species of family Mullidae (Table 6).

Table 5. Comparison of *U. vittatus* and *U. supravittatus* and 3 other species of *Upeneus* previously recorded from Bangladesh.

	<i>U. vittatus</i> ^{a,b}	<i>U. supravittatus</i> ^{a,b}	<i>U. sulphureus</i> ^{a,b}	<i>U. sundaicus</i> ^{a,b}	<i>U. tragula</i> ^{a,b}
Body stripe	4 golden	2 yellow	2 yellow	1 broad yellow	1 broad black
Body color	Reddish dorsally, silver ventrally	Reddish dorsally, silver ventrally	Pale brown dorsally, white ventrally	Light reddish dorsally, white ventrally with yellow-green ventral margin	Light greenish dorsally, whitish ventrally
Spot, stripe or patch on dorsal fin	Broad black spot on the tip of 1 st dorsal fin	Absent	1 st dorsal fin tip black	3 red stripes	2 black and 2 white patches present alternately
Bar on caudal fin	5 yellow black oblique bars on upper lobe and 3 on lower	5 black oblique bars on upper lobe and 4 on lower	No bar	5 red bars on upper lobe and no bar on lower lobe	4 blackish oblique bars on upper lobe and 5 on lower
Lateral line scales	37 (36-38)	35 (34-36)	34(34-37)	34 (31-34)	31 (28-31)
Scales above/below lateral line	2/6	2/6	2/4	3/5	3/5

^aPresent study; ^bUiblein and Heemstra (2010) and reference values are shown in parentheses.

The construction of Neighbor-joining tree was done on the basis of K2P model, where replications of bootstrapping test was 1000. In the phylogenetic tree (Fig. 3) *U. vittatus* and *U. supravittatus* clustering together and they are apparently also very similar but with distinct difference in morphology.

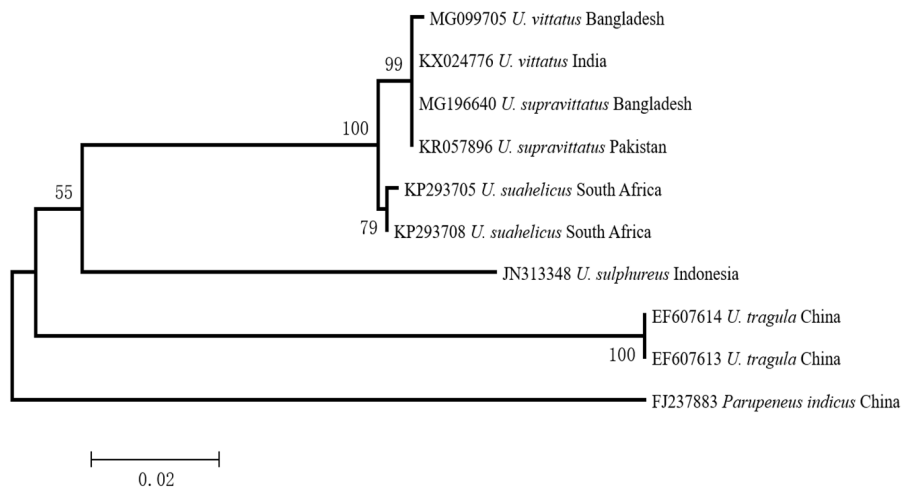


Fig. 3. Neighbor-joining tree constructed using the K2P model for COI gene sequences of six species of *Upeneus* with *Parupeneus indicus* as the outgroup.

Table 6. Mean interspecies genetic distances (K2P) between six species of family Mullidae.

	<i>U. vittatus</i>	<i>U. supravittatus</i>	<i>U. suahelicus</i>	<i>U. sulphureus</i>	<i>U. tragula</i>
<i>U. vittatus</i>					
<i>U. supravittatus</i>	0.002				
<i>U. suahelicus</i>	0.009	0.008			
<i>U. sulphureus</i>	0.118	0.116	0.115		
<i>U. tragula</i>	0.158	0.156	0.152	0.159	
<i>Parupeneus indicus</i>	0.163	0.161	0.155	0.183	0.198

There are a few reports on the occurrence of *Upeneus* species in Bangladesh. For example, only three species of this genus, *U. sulphureus* (Tomascik 1997), *U. sundaicus* and *U. tragula* (Rahman *et al.* 2009) are reported in Bangladesh waters based on their identification only on external morphological characters. However, misidentifications are known to result from damaged or poor specimens and ambiguous morphological features (Arai and Wong 2016). Morphological characters are ineffective to distinguish species at some stages of development (Ward *et al.* 2009). Several authors have suggested that molecular genetic analyses are necessary for precise species confirmation (Arai *et al.* 2015, Arai and Wong 2016, Kadir *et al.* 2017). However, on the basis of morphological characters and DNA barcoding, *U. vittatus* and *U. supravittatus* were newly recorded in Bangladesh. Photographs, counts and measurements were taken for morphological characters, then comparison of the present study was done with the reports of Uiblein and Heemstra (2010), Kuitert and Tonzuka (2001) and FAO (1983a,b,c). Even though some do not precisely fall into the range reported by others, due to differences in growth rate and sampling size for some of the characters that either overlap or span the range. The K2P distance between *U. vittatus* and *U. supravittatus* is 0.2%. This is lower than threshold of species delimitation 3.5% (Ward *et al.* 2005, 2009). This may be due to lower genetic diversity between closely related and apparently morphologically similar species of the genus *Upeneus* such as *U. supravittatus* and *U. suahelicus* have only 0.8% genetic divergence (Uiblein and Gouws 2015) that is also lower than threshold of species delimitation 3.5% (Ward *et al.* 2005, 2009). For more confirmation need further study such as Kruck *et al.* 2013 suggested that when CO1 revealed low interspecific variation which is insufficient for genetic distance-based species identification and mismatch among morphological identifications then multi-gene spot test approach and efficient markers (recombination activating gene 2 in combination with NADH dehydrogenase 2 and ATP synthase) may help identifying problematic fish species.

Conclusion

For the first time in Bangladesh *Upeneus vittatus* and *U. supravittatus* were recorded based on both morphometrics and DNA barcoding. The morphometrics descriptions are compatible with descriptions of these species given by Zoologists. Moreover, these two species and other three previous Bangladeshi *Upeneus* species can be distinguished from each other by combination of characters like body stripe, body color, spot, stripe or patch on dorsal fin, bar on caudal fin, lateral line scales and scales above and below the lateral line. Although *U. vittatus* and *U. supravittatus* have low genetic variation but significant difference from other species of the same genus that were also confirmed from comparative COI sequence analysis of the present study with reference sequences. Therefore, *U. vittatus* and *U. supravittatus* are valid species at the genetic level.

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References

- Arai, T., T.C. Chin, K.O. Kwong, and M.N. Siti Azizah. 2015. Occurrence of the tropical eels, *Anguillabengalensis bengalensis* and *A. bicolor bicolor* in Peninsular Malaysia, Malaysia and implications for the eel taxonomy. *Mar. Biodiver. Reco.* **8**: e28. <https://doi.org/10.1017/S1755267215000950>
- Arai, T, and L.L. Wong. 2016. Validation of the occurrence of the tropical eels, *Anguilla bengalensis bengalensis* and *A. bicolor bicolor* at Langkawi Island in Peninsular Malaysia, Malaysia. *Tropi. Ecol.* **57**: 23-31.
- FAO, 1983a. FAO species identification sheets, Mullidae. Fishing area 51 (West Indian Ocean).1654 pp. <ftp://ftp.fao.org/docrep/fao/009/y4162e/y4162e32.pdf>
- FAO,1983b. FAO species identification sheets, Mullidae. Fishing area 51 (West Indian Ocean).3175 pp. <ftp://ftp.fao.org/docrep/fao/009/y0770e/y0770e39.pdf>
- FAO, 1983c. FAO species identification sheets, Mullidae. Fishing area 51 (West Indian Ocean). <ftp://ftp.fao.org/docrep/fao/009/ad468e/AD468eIF.pdf>
- Fischer, W., I. Sousa, and C. Silva.1990. Sheets FAO species identification of fishing activities. Field guide of commercial marine species. Roma, FAO. 424 pp.
- Hebert, P.D.N, A. Cywinska, and S.L. Ball.2003. Biological identifications through DNA barcodes. *Proc. R. Soc. Lond. B.* **270**: 313-321.
- Ivanova, N.V., T.S. Zemlak, and R.H. Hanner, 2007. Universal primer cocktails for fish DNA barcoding. *Mol Eco N.* **4**(7): 544-548.

- Kadir, A., S.R., M.H.F. Abdul Rasid, K.O. Kwong, L.L. Wong, and T. Arai. 2017. Occurrence and the ecological implication of a tropical anguillid eel *Anguilla marmorata* from peninsular Malaysia. *ZooKeys*.**695**: 103-110. <https://doi.org/10.3897/zookeys.695.13298>
- Kimura, M.1980.A simple method for estimating evolutionary rates of base substitutions through comparative studies of nucleotide sequences. *J. Mol. Evo.* **16**:111-120.
- Kruck, N.C., I.R. Tibbetts, R. D. Ward, J.W. Johnson, W.K.W. Loh, and J.R. Ovenden. 2013. Multi-gene barcoding to discriminate sibling species within a morphologically difficult fish genus (*Sillago*). *Fishe. Res.* **143**: 39-46.
- Kuiter, R.H, and T. Tonozuka. 2001. Pictorial guide to Indonesian reef fishes. Part 1. Eels-Snappers, Muraenidae - Lutjanidae. *Zoone. Aus.* 1-302 pp.
- Lewis, A.D, and C.K. Pring.1986. *Freshwater and brackish water fish and fisheries of Fiji*. 38-59 p. *In*: Reports and papers presented at the Indo-Pacific Fishery Commission Expert Consultation on inland fisheries of the larger Indo-Pacific islands. Bangkok, Thailand, 4-6 August 1986. FAO Fish. Rep. (371) Suppl.:258 p. (eds. Petr T)
- Lieske, E, and R. Myers. 1994. Collins Pocket Guide. Coral reef fishes. Indo-Pacific & Caribbean including the Red Sea. Haper Collins Publishers. 400 pp.
- Mundy, B.C.2005.Checklist of the fishes of the Hawaiian Archipelago. Bishop Museum Bulletins in Zoology. *Bishop Mus. Bull. Zool.* **6**: 1-704.
- Rahman, A.K.A., S.M.H. Kabir, M. Ahmed, A.T.A. Ahmed, Z.U. Ahmed, Z.N.T. Begum, M.A. Hasan, and M. Khondker. (eds.) 2009. Encyclopedia of Flora and Fauna of Bangladesh, vol. 24 Marine Fishes. Asiatic Society of Bangladesh, Dhaka. **24**: 276-279.
- Saitou, N, and M. Nei. 1987.The neighbor-joining method: A new method for reconstructing phylogenetic trees. *Mol Biol Evol.* **4**: 406-425.
- Schultz L.P, and collaborators. 1960. Fishes of the Marshall and Marianas Islands. *Bull. US Natl. Mus.* vol. **2**. 202: ix + 438 pp.
- Sea-Ex (<http://www.sea-ex.com/fishphotos/goatfish.htm>)
- Sousa, M. I, and M. Dias. 1981. Catalog Fish Mozambique - South Zone Fishing Development Institute, Maputo. 121 pp.
- Tamura, K., G. Stecher, and D. Peterson. 2013. MEGA6: Molecular Evolutionary Genetics Analysis Version 6.0. *Mol. Biol. Evol.* **30**: 2725-2729.
- Tomascik, T.1997. Management plan for coral resources of Narikel Jinjira (St. Martin's Island), national conservation strategy implementation project-1, Ministry of Environment and Forest, Government of Bangladesh. 126 pp.
- Uiblein, F, and P.C. Heemstra.2010.A taxonomic review of the Western Indian Ocean goatfishes of the genus *Upeneus* (Family Mullidae), with descriptions of four new species. *Smithiana*.**11**: 35-71.
- Uiblein, F, and P.C. Heemstra. 2011. Description of a new goat fish species, *Upeneus seychellensis* sp. nov. (Mullidae) from the seychelles Bank with remarks on *Upeneus guttatus* geographic variation and a key for all Western Indian Ocean *Upeneus* species. *Mar. Biol. Res.***7**: 637-650.

- Uiblein, F, and G. Gouws. 2015. Distinction and relatedness –Taxonomic and genetic studies reveal a new species group of goatfishes (*Upeneus*; Mullidae), *Mar. Biol. Res.* **11**:10, 1021-1042, DOI: 10.1080/17451000.2015.1064963
- Ward, R.D., T.S. Zemplak, and B.H. Innes. 2005. Barcoding Australia's fish species. *Philos. Trans. R. Soc. of Lond B.* **360**: 1847-1857.
- Ward, R.D., R. Hanner, and P.D.N. Hebert. 2009. The campaign to DNA barcode all fishes, FISH-BOL. *J. Fish Biol.* **74**: 329-356.
- Ward, R.D. 2012. FISH-BOL, a case study for DNA barcodes. *Methods Mol. Biol.* **858**: 423-439.
- Yamashita, Y., D. Golani, and H. Motomura. 2011. A new species of *Upeneus* (Perciformes: Mullidae) from southern Japan. *Zootaxa.* **3107**: 47-58.

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