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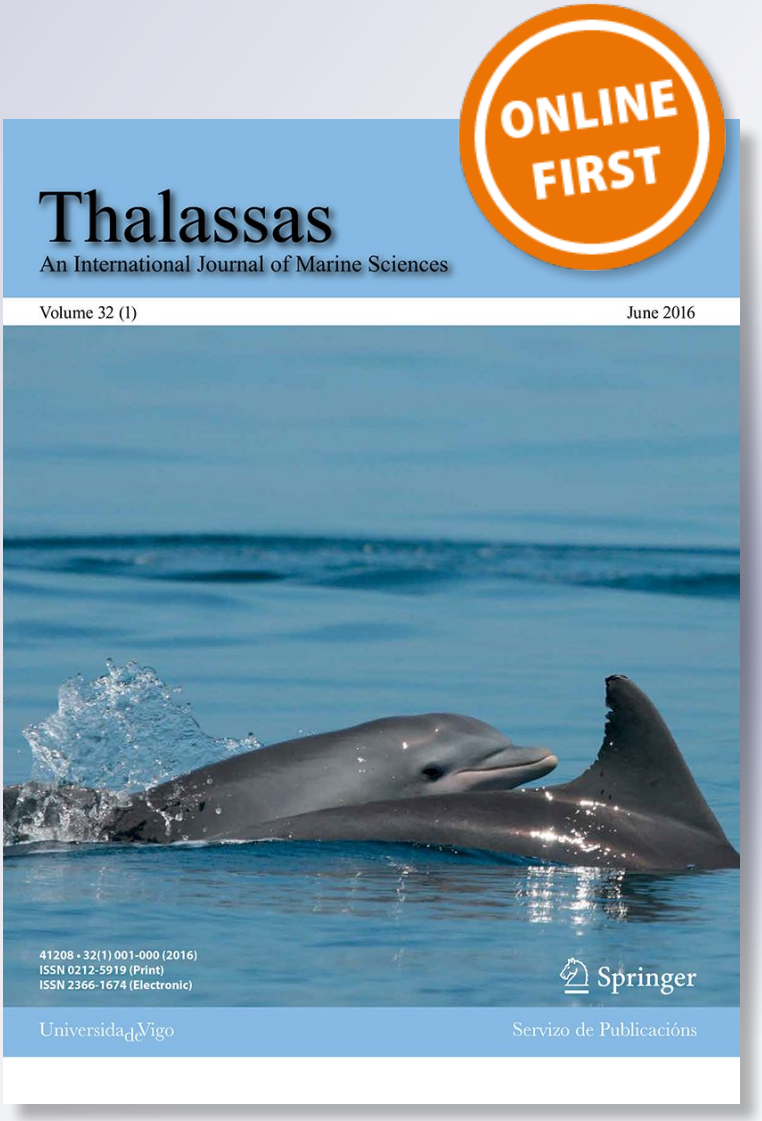

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New Distributional Record of Deep Sea Snake Fish *Acanthocepola indica* (Day, 1888) from the Southwest Coast of India

V. Mahesh[1] · P. K. Asokan[1] · N. S. Jeena[2] · K. Vinod[1] · K. P. Said Koya[1] · P. U. Zacharia[2]

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Abstract
Deep sea snake fishes or band fishes belonging to the Family Cepolidae are widespread in eastern Atlantic, but rare in the central Indo-Pacific Ocean. A specimen of *Acanthocepola* sp. measuring 556 mm in total length (TL) weighing 139 g was collected from a multiday trawler, operated off the southwest coast of India and landed at Beypore Fisheries harbour, Calicut, Kerala on 9th November 2017. The species was identified as *A. indicus* based on morphometric and meristic characteristics which was later confirmed by DNA barcoding.

Keywords *Acanthocepola indica* · Band fish · Cepolidae · Calicut · India

Introduction

Perciformes is the most diversified order, comprising about 40% of bony fishes. The order is difficult to define because members do not share a unique character or a combination of characters. Many species have a generalized perch-like body form. Scales are usually ctenoid, although sometimes they are cycloid or otherwise modified. Most members of Perciformes are marine shore fishes, and the perciforms dominate the vertebrate ocean life (Nelson 2006; Bray and Martin 2018). The family Cepolidae comprises two sub families, Cepolinae and Owstoniinae, five genera and about 23 species. Most species live in self-made burrows in muddy or fine-sand areas, feeding on zooplankton and pelagic eggs (Froese and Pauly 2017). In the genus *Acanthocepola*, three species namely *Acanthocepola indica* (Day 1888), *A. limbata* (Valenciennes 1835) and *A. abbreviata* (Valenciennes 1835) are known from the Indian waters (Day 1888; Fischer and Bianchi 1984). These fishes are commonly called as band fishes. After Day's record of *A. indica* from Madras waters (Day 1888), there has been no record of the species from Indian waters. On 9th November, 2017 one specimen of *A. indica* measuring 556 mm in total length (TL) and weighing 139 g was collected from Beypore fisheries horbour, Calicut (Fig. 1). It formed part of the catch landed by a multiday trawler operated off South-west coast of India, at depth range between 100 and 180 m. The morphometric and meristic characters of the specimen were described and nucl17:39:00 eotide sequencing of the mitochondrial cytochrome c oxidase subunit I (COI) gene was carried out to confirm the species (Hubert and Hanner 2015).

Materials and Methods

Species identification was initially done based on morphometric and meristic characters as per the description of Day (1888) and Smith-Vaniz (1986). Subsequently, Genomic DNA was isolated from the tissue stored in 90% ethanol using phenol-chloroform method (Sambrook and Russell 2001). Amplification of partial sequences of COI gene was carried out using the primer set LCO1490/ HCO2198 (Folmer et al. 1994). PCR reactions were carried out in BIORAD T100 TM thermal cycler (Biorad, USA). The reactions were performed in 25 μl containing 2.5 μl 10x assay buffer, 1.5 μl $MgCl_2$ (1.5 μM), 0.5 μl of 10 μM of each primer, 0.5 μl of 10 μM dNTPs, 1 U Taq DNA polymerase (Sigma Aldrich, USA) and 1 μl of 50–100 ng template DNA. The PCR cycling profiles were as follows: An initial denaturation of 4 min at 94 °C, 30 cycles of denaturation for 30 s at 94 °C, 30 s of annealing at 42 °C, 45 s of extension at 72 °C, and a final extension of 7 min at 72 °C. The PCR products were checked on

✉ V. Mahesh
mahesh.fishco@gmail.com

[1] Calicut Research Centre of ICAR-Central Marine Fisheries Research Institute, P.B. No. 917, Westhill P.O., Kozhikode 673005, India

[2] ICAR-Central Marine Fisheries Research Institute, P.B. No. 1603, Ernakulam North P.O, Kochi 682018, India

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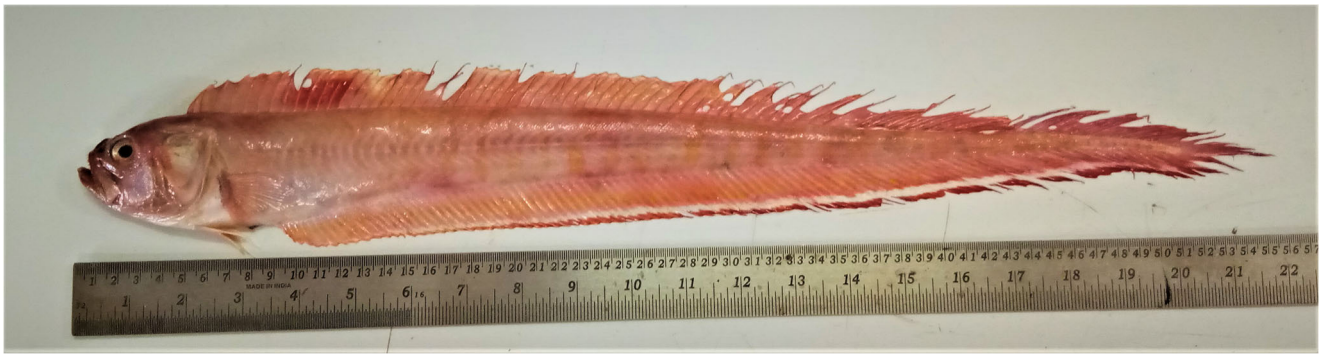

Fig. 1 *Acanthocepola indica,* 556 mm, landed at Beypore, Calicut, India

1.5% agarose gels, bi-directionally sequenced and aligned in MEGA 7 (Kumar et al. 2016). The COI sequence (678 bp) generated in this study was submitted to NCBI GenBank (Accession no.: MG886841) and was BLAST against GenBank database to confirm the identification. Similar sequences from related species of the same genus were aligned and assembled using MEGA7 and further phylogenetic analysis was carried out in the software with *Cepola macrophthalma* as out-group.

Results

Morphometric (Table 1) and meristic (Table 2) traits for the specimen are shown in the tables. Measurements are exhibited as a percentage against SL: Body depth 11.02; body width 5.31; head length 12.40; snout length 1.77; postorbital length 4.33; eye diameter 2.75; upper jaw length 4.92; interorbital width 1.96; predorsal fin length 10.23; prepectoral fin length 12.99; prepelvic fin length 10.43; preanal fin length 17.91; preanus length 15.35; pectoral fin length 6.69; pelvic fin length 7.08; length of longest dorsal fin ray 7.28; length of longest anal fin ray 8.26.

Description Body elongated, highly compressed laterally and gradually tapering to caudal fin. Snout short and blunt, its length shorter than eye diameter; eyes large, protractile, located laterally in anterior half of the head and middle part of eye bright dark. Interorbital space flatten. Mouth large and deeply oblique with triangular tongue, both jaws equally protruding, upper jaw broad at end and reach to the middle of eye. Preopercular margin with five blunt and one posterior serrated spine. A single row of fine, slightly curved canine teeth on both jaws in addition a row of ten teeth at anterior part of lower jaw, anterior teeth slightly bigger than lateral ones. Two nostrils, posterior nostril a simple pore, located just anterior edge of eye, slightly bigger than anterior one. Gill openings wide, semi-circular, gill rakers long and slender. A single large dorsal and pelvic fin inserted at the same distance on head and

Table 1 Morphometric measurements of *Acanthocepola indica* collected from Beypore F.H

Parameter	Measurements (mm)
Total length (TL)	556
Standard length (SL)	508
Head length	63
Snout	9
Eye diameter (Same along both axes)	14
Eye (middle dark portion)	7
Maxillary length	24
Mandibular length	16
Width of gill opening	46
Snout to insertion of dorsal fin	42
Length of dorsal fin	455
Snout to insertion of pectoral fin	48
Length of pectoral fin	34
Snout to insertion of pelvic fin	41
Length of pelvic fin	36
Snout to insertion of anal fin	76
Length of anal fin	420
Length of caudal fin	48
Snout to vent	71
Snout to origin of lateral line	46
Gape	23
Depth of body in line with eye	38
Depth of body at dorsal fin insertion	54
Depth of body at pectoral fin insertion	56
Depth of body at pelvic fin insertion	54
Depth of body at anal fin insertion	53
Depth of body at mid-length	30
Depth of body at caudal fin insertion	4
Inter-orbital distance	10
Distance between eyes	18
Breadth of body at dorsal fin insertion	23
Breadth of body at mid-length	9

Table 2 Comparison of meristic characters of *Acanthocepola indica*

Particulars	Present study	Day (1888)	Smith-Vaniz (1986)	Nakabo (2002)	Park et al. (2008)
Place of report	Calicut, India	Madrass, India	South Africa	Japan	Korea
Number of specimens	1	1	–	–	1
Total length (mm)	556	200.3	–	–	245.0
Standard length (mm)	508	–	–	–	220.4
Dorsal fin rays	83	ca.90	82–89	ca. 85	88
Anal fin rays	93	ca. 90	91–102	ca. 100	101
Pectoral fin rays	19	17	–	–	17
Pelvic fin rays	1,5	–	–	–	1,5
Caudal fin rays	10	–	–	–	–
Gill rakers	13 + 32	–	–	–	16 + 33
Gill filaments	100	–	–	–	–
Branchiostegal rays	6	6	–	–	6
Vertebrae	–	–	12 + 60–66	–	12 + 66
Pre opercula spine	1 + 5	1 + 5	–	–	–

ventral part. Pectoral fin inserted posterior to pelvic fin, originates at fourth dorsal fin ray. Posterior margin of pectoral fin rounded, all rays branched. Outermost ray of pelvic fin longest. First dorsal and anal fin rays short. Anal fin also long, commences against eighth dorsal fin ray, dorsal and anal fin bases confluent with caudal fin. Anus located just before origin of anal fin. Lateral line originates at the upper margin of gill opening, then running very close to dorsal fin base terminating posteriorly near end of fin. Caudal fin lanceolate. Scales minute, cycloid, present on cheek and body.

Colour Fresh specimen had pinkish coloured head and pinkish-red coloured body with 19 golden yellow bands on sides. The bands originate at 16th dorsal fin ray. A brick red oblong blotch is present on the dorsal fin between 9th and 15th rays, covering more than half of the height of rays. Membranous portion of the dorsal fin with golden yellow and anal fin with dark red colour runs along the fins and merges to caudal fin. A white band of about 3 mm wide originates at 1st ray of the anal fin, then runs throughout above the dark red margin of the fin and spreads widely on caudal fin. Pectoral fin translucent. Pelvic fin pinkish white in colour. A distinctive dark stripe is present on the membrane (hidden) connecting the premaxillary and maxillary bones of the upper jaw.

Discussion

Acanthocepola indica is known to be distributed in Indo-west Pacific: India (South west coast in present study; Madras, Day 1888), Korea (Park et al. 2008), Japan (Masuda et al. 1984; Nakabo 2002), China (Randall and Lim 2000) Taiwan (Shen et al. 1993) and Natal in South Africa (Smith-Vaniz 1986; Heemstra 1995). *A. indica* could be distinguished from *A. limbata* by the number of dorsal fin rays (83–88 in the former vs. 102–104 in the latter) and body depth 11–14 vs. 7–9 in SL). *A. indica* also differs from *A. abbreviata* by the number of dorsal fin rays (83–88 vs. 67–74) (Masuda et al.

Table 3 Comparison of meristic characters of *A. indica* with the prior reports of *A. limbata* from Indian waters

Particulars	*Acanthocepola indica*	*Acanthocepola limbata*	
	Present study	Joshi et al. 2014	Kulkarni and Balasubramanian 1978
Number of specimens	1	2	1
Dorsal fin rays	83	81–83	83
Anal fin rays	93	91–93	93
Pectoral fin rays	19	19	19
Pelvic fin rays	1,5	6	6
Caudal fin rays	10	10	10
Gill rakers	13 + 32	52–55	17 + 34
Gill filaments	100	100	100

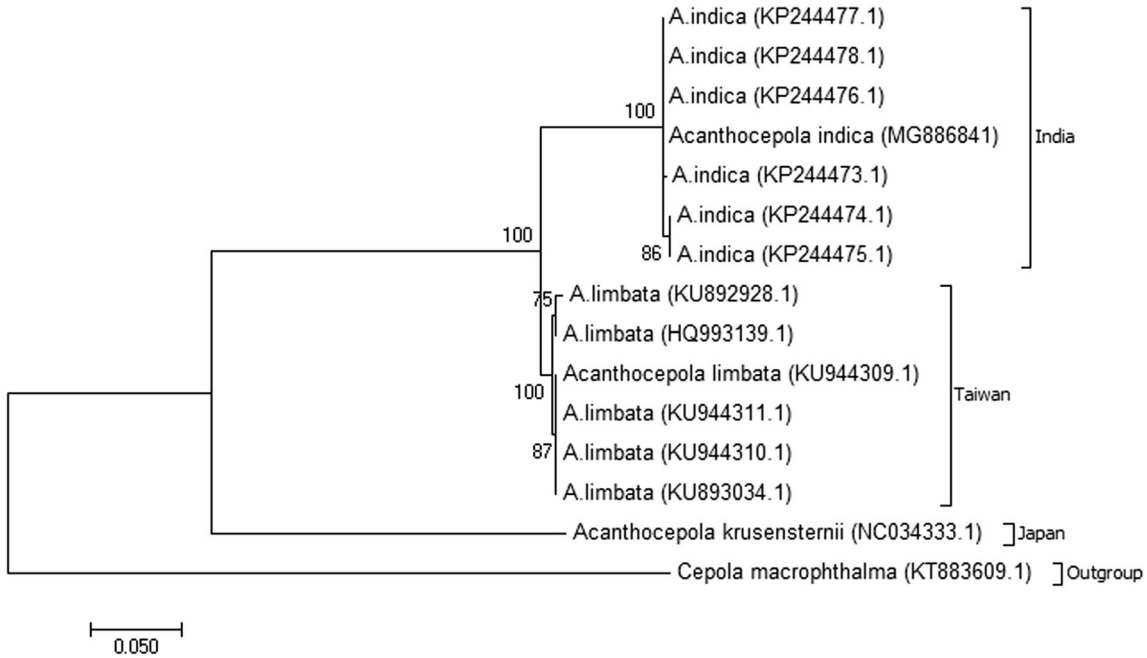

Fig. 2 Molecular phylogenetic analysis of *Acanthocepola* spp. by Maximum Likelihood method

1984; Smith-Vaniz 1986; Nakabo 2002; Park et al. 2008). Since the meristic counts and description of the present study coincided with the earlier reports of *A. limbata* from Indian waters (Kulkarni and Balasubramanian 1978; Manojkumar and Pavithran 2011 and Joshi et al. 2014) (Table 3), molecular identification method was also employed for confirmation of species. The COI sequence (Accession no.: MG886841) from the present specimen exhibited an identity of 95–100% to *A. indica* sequences available in GenBank. The evolutionary history for three species of the genus was inferred from 525 bp long COI region and phylogeny was reconstructed using the Maximum Likelihood method based on the Hasegawa-Kishino-Yano model (Hasegawa et al. 1985) (Fig. 2). The high bootstrap values in the phylogenetic tree showed independent status of the three taxa under comparison. *A. indica* from the Southwest coast of India (Accession no.: MG886841) clustered with the samples from the Southeast coast of India. The evolutionary divergence between sequences was calculated using the Kimura 2-parameter model (Kimura 1980). On group wise comparison of the data, it was found that the genetic distance between *A. indica* & *A. limbata* was 7.4%. The highest genetic distance of both species was from *A. krusensternii* (>21%).

The morphometric and meristic characters of the specimen agree well with the description of Day (1888) and Smith-Vaniz (1986). DNA barcoding which is a validated tool for species identification (Henriques et al. 2015) was employed as a supplemental identification method. The species of this study, showed high sequence divergence values well beyond the species delineation limit of 2% (Hebert et al. 2003) from related species of the genus. However morphologically the present specimen showed difference with the Korean specimen of *A. indica* in respect to percentage of SL viz. Body depth (11.0 in the former vs. 13.7 in the latter), body width (5.3 vs. 5.7), head length (12.4 vs. 14.4), snout length (1.7 vs. 3.3), postorbital length (4.3 vs. 6.8), eye diameter (2.8 vs. 4.4), upper jaw length (4.9 vs. 6.0) inter orbital width (2.0 vs. 3.2) pre dorsal fin length (10.2 vs. 11.3) pre pectoral fin length (13.0 vs. 14.4), pre pelvic fin length (10.4 vs. 13.8), pre anal fin length (17.9 vs. 17.2), pre anus length (15.4 vs. 16.6), pectoral fin length (6.7 vs. 8.8), pelvic fin length (7.1 vs. 7.8), length of longest dorsal fin ray (7.3 vs. 8.8) and length of longest anal fin ray (8.3 vs. 8.1). Meristic characteristics such as number of dorsal fin rays, anal fin rays, pectoral fin rays and gill rakers also differed from the Korean specimen (Table 2). This would be due to the geographic variations between the coasts.

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Compliance with Ethical Standards

Conflict of Interest The authors declare that they have no conflict of interest.

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