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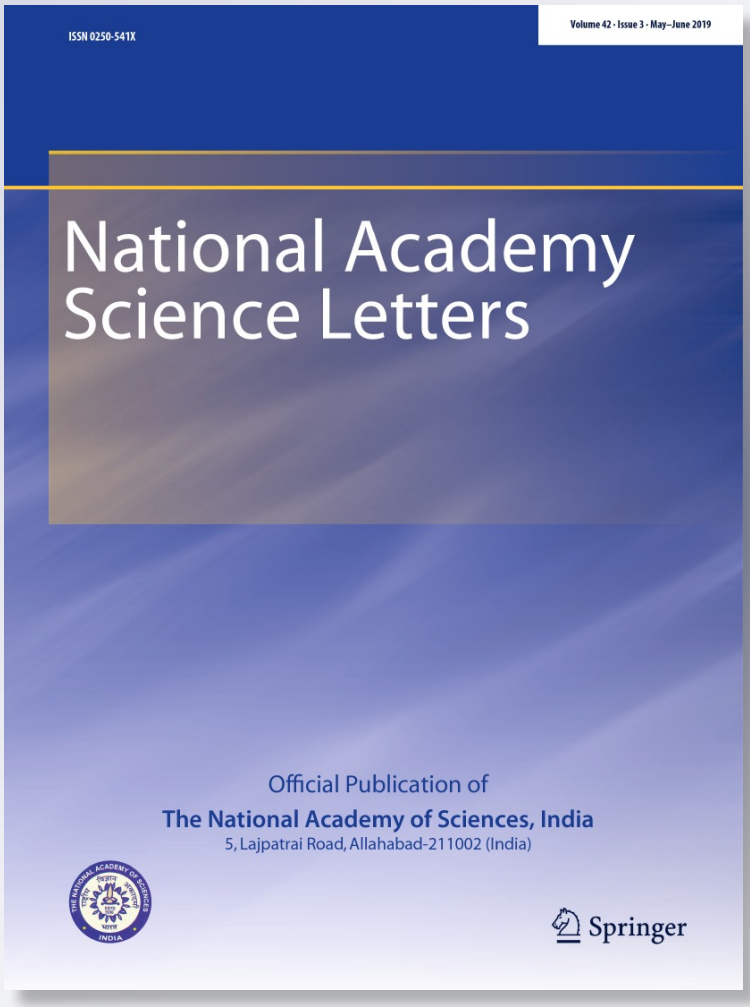

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SHORT COMMUNICATION

First Report on Infection of *Argulus quadristriatus* (Arthropoda: Crustacea: Branchiura) on Marine Fish Cobia in Brood Stock Pond Culture

R. Subburaj[1] · B. A. Venmathi Maran[2] · A. R. T. Arasu[1] · M. Kailasam[1] · S. Elangeshwaran[1] · Prem Kumar[1] · G. Thiagarajan[1] · K. Sukumaran[1]

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Abstract A total of 30 specimens of fish cobia *Rachycentron canadum* (Total Length = 45–120 cm, Weight = 3.0–25 kg) were stocked at the density of 1 kg/m^3 in the polythene lined earthen pond. After 3 months of stocking, fish cobia was found with infection of ectoparasites. Then fishes were sampled at fortnight interval to find the percentage distribution of ectoparasites in different parts of the body for a year and also any pathological symptoms. Identification of the parasite was made through light and electron microscopies. The parasite was identified as *Argulus quadristriatus* Devaraj and Ameer Hamsa, 1977 (Crustacea: Branchiura: Argulidae) commonly called as fish lice. The maximum distribution of pathogenic argulid was observed on the head and operculum of cobia and was found high in summer months from April to June. Pathological symptoms were observed on cobia as erratic swimming, rubbing against substrate in the pond and lesions of epithelial tissues on the infected regions. It must be due to continuous rupturing and feeding of argulids on the skin of cobia using its powerful antennae. Scanning electron micrographs revealed some important morphological features of *A. quadristriatus*. This is a first report of *A. quadristriatus* infection on cobia reared in a land-based pond ecosystem.

Keywords Cobia · Fish louse · Argulid · Brood stock · Lesions

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Argulids (Crustacea: Branchiura: Argulidae) are commonly called as fish lice, occurring as ectoparasites of freshwater, marine and estuarine fishes [1]. The subclass Branchiura contains a single family, the Argulidae including four valid genera and one among them is *Argulus*. The genus *Argulus* Müller, 1785 is cosmopolitan in distribution and is found in Africa, Europe, Asia, Australia and North and South America [1, 2]. In India, about 12 species are described in freshwater fishes and one each in marine and estuary [3, 4]. Species of *Argulus* are the only ones to possess an oral spine. It can easily attach and detach from their host causing damage to epithelial tissue of the host and leads to secondary infection from bacteria and virus. It also acts as a vector by transmitting diseases from one host to another host [1, 5, 6].

The marine fish cobia, *Rachycentron canadum* (Linnaeus, 1766) (Perciformes: Rachycentridae) (Fig. 1) is well distributed in the Indo-Pacific, temperate waters of west and east Atlantic and throughout the Caribbean, Australia and Japan [7]. It is one of the most commercially important fishes and considered as a prominent species for aquaculture. In recent years, several Asian countries have started raising cobia commercially [7, 8]. In India, a successful seed production of cobia was recently achieved using brooders maintained in sea cages and pond reared broodstock [8, 9]. Central Institute of Brackishwater Aquaculture (CIBA), Chennai, India has standardized the methodology of broodstock development of cobia in pond culture system [9]. During the process of broodstock development in pond culture system, infection of ectoparasites in different body parts was noticed. The present work was conducted to identify the infected ectoparasites through light and electron microscopies, and also to observe behavior of cobia, examine pathological symptoms and percentage distribution of ectoparasites on different body parts of cobia.

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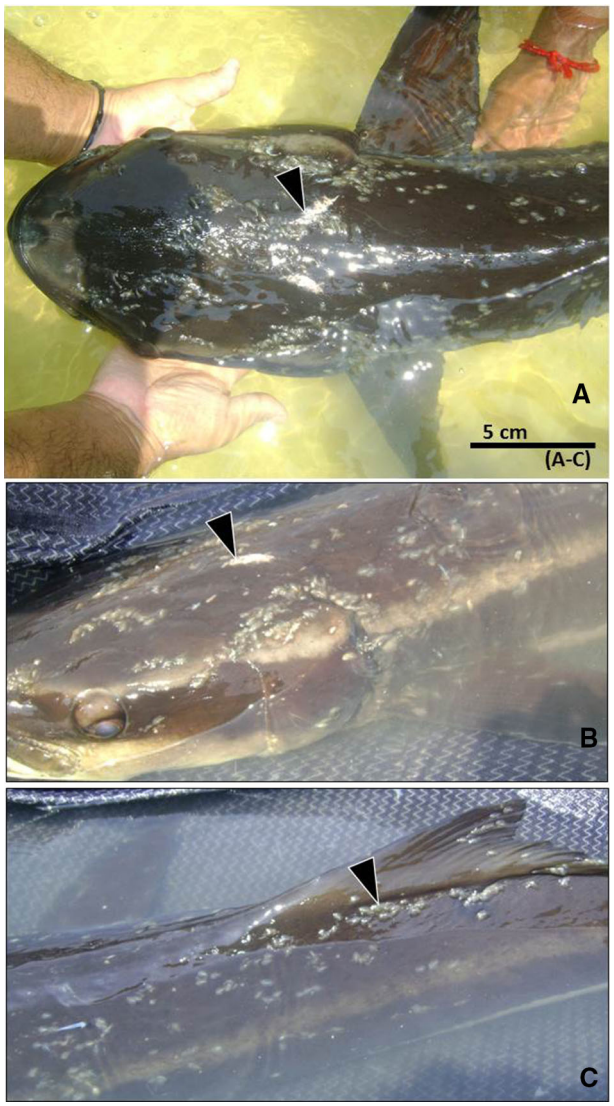

Fig. 1 Infection of fish lice on the body surface of *Rachycentron canadum* broodstock maintained at hatchery, head (**a**, **b**) and tail (**c**) regions. Arrows (**a**, **b**) indicated lesions on body and arrow in C indicates bunch of argulids

A total of 30 specimens of fish cobia *Rachycentron canadum* (Total Length = 45–120 cm, Weight = 3.0–25 kg) were procured from wild facility near Muttukkadu, Mahabalipuram (12°46′56.21N; 80°15′36.99E) and transported in Fiberglass Reinforced Plastic tanks with medical grade oxygen bubbling to fish hatchery located at the Muttukkadu Experimental Station of Central Institute of Brakishwater Aquaculture (CIBA), Chennai, India (12°48′40.17N; 80°14′37.34E).

Fishes were acclimatized, quarantined [10] and stocked at the density of 1 kg/m^3 in the polythene lined earthen pond (size 45 × 15 m) having the average water depth of 1.5 m. Fishes were fed with forage fishes like tilapia (*Oreochromis mossambicus*) and oil sardine (*Sardinella* sp.) @ 5% of the average body weight once a day. After the 3 months of stocking parasitic infection was noticed on the body of some fishes (Fig. 1a, b). These ectoparasites were removed carefully with forceps and fixed in 70% ethanol. To find the percentage distribution of ectoparasites on different parts of body, fishes (n = 30) were sampled at fortnight interval for a year.

To identify the parasites and also to understand the systematics of parasite was, it examined under compound phase contrast microscope (Leica M205C, Seoul) and scanning electron microscope (SEM). Ethanol fixed parasites were observed under light microscope. For SEM, the specimens were dehydrated through a graded series of ethanol (90, 99.5 and 100%) and finally by isoamyl alcohol. The samples were critical point-dried using CO_2 gas and ion-sputtered for observation under SEM (model: JSM6510-LV, JEOL, Tokyo). Mean values of parasite distribution were subjected to one-way ANOVA analysis and post hoc test were carried out using Duncan'smultiple range tests to determine the significant differences if any between the mean values. Comparisons were made at 5% probability level. All statistical analyses were performed with the software SPSS 17.0 version for Microsoft Windows. Specimens are deposited at the Zoological Survey of India Museum, Kolkata.

Argulids are economically important pests of fish in temperate and tropical regions [1, 5]. Report on *Argulus* spp. in marine/brackishwater is relatively fewer than fresh water. In the study, we report ectoparasites from the pond cultured fish cobia. Based on the previous report on *Argulus* [10], the present species seems to be *Argulus quadristriatus* Devaraj and Ameer Hamsa, 1977 (Crustacea: Branchiura: Argulidae) (Fig. 2a, b) since, four longitudinal stripes are found on the dorsal side of body (Fig. 2a). The following description of an adult female *A. quadristriatus* is considered as its taxonomical features.

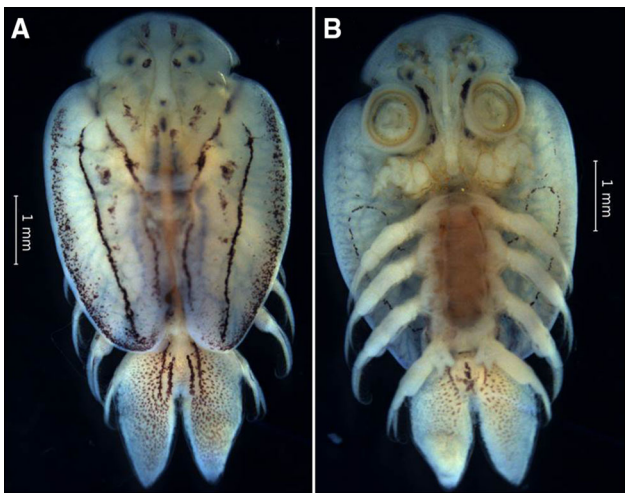

Fig. 2 Infected fish lice *Argulus quadristriatus* (Crustacea, Branchiura): dorsal (**a**) and ventral (**b**)

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Body 6.59–10.88 mm long (n = 10), consists of cephalon, four thoracic somites and abdomen. Cephalon longer than wide, having 2 compound eyes and nauplius eye. Compound eyes long, with medial cleft on middle of abdomen and 2 carapace lobes; ventral surface covered with pointed spinules on frontal region and lateral margins. First to fourth thoracic somites densely ornamented with spinules on ventral surface; fourth thoracic somite bearing pair of posterolateral lobes dorsally. Abdomen bilobate, longer than wide, comprising caudal rami. Caudal rami long, bearing five setae each.

Scanning electron micrographs revealed the sucking discs (Fig. 3a) (maxillules), consisting of 50–60 ribs and, at the base three concentric layers of muscle fibers was observed. Number of sclerites in normal rods (ribs) ranges from eight to ten. The maxillipeds (Fig. 3b, c) are five segmented/podomere, the basal segment bearing three stout spines (arrowed). Four pairs of biramous swimming legs (Fig. 3b, d) were present in the thoracic region. All features observed in the present study were similar to the previous report from India [11] and Japan [12].

Argulus quadristriatus was first described based on a single adult female specimen from the body surface of the seaperch *Psammoperca waigiensis* (Cuvier, 1828) (Perciformes: Latidae) collected along the Palk Bay off Mandapam, south east coast of India [11]. The morphological features are closely similar to the present study. Recently the same species *A. quadristriatus* has also been reported from three species of teleost host species including two species of goatfish, *Upeneus tragula* Richardson, 1846 and *Parupeneus ciliatus* (Lacepède, 1802) (Perciformes: Mullidae), and a species of goby, *Favonigobius reichei* (Bleeker, 1854) (Perciformes: Gobiidae) off Japanese waters [12]. All reported species of *A. quadristriatus* have been reported from different Perciformes of five different families of five different fishes [11, 12, present study].

The redescribed Japanese specimens [12] are closely similar to Indian specimens [11, present study], it is evident through our SEM study. In the present study, maximum percentage distribution of *A. quadristriatus* was noticed on head and operculum ($p < 0.05$) (Fig. 1 and Table 1). Site of infection revealed different size of argulids with excess mucus secretion. Pathological symptoms of infected fishes were observed as erratic swimming, rubbing against substrate in the pond, red spots on abdomen, lesions of epithelial tissues on the infested regions and discoloration of skin. Like other argulids, *A. quadristriatus* attached to fish skin by means of suckers, repeated penetration to the skin of the fish to feed on blood, causing wounds and lead to ulcer and secondary infection [13]. While severe infection occurs on the host, it becomes lethargic, non-feeding, discoloration of body and stress [13, 14]. In the present study, we have observed high number of argulids on the fish body in summer months from April to June. Results in the present study are similar to that of some previous studies mentioned that the intensity of argulid infection increased with rise in temperature [1, 5, 15] and the degree of infection on host depends upon the number of parasites and the size of the host fishes [15]. When temperature increased, hatching of *Argulus* commenced and population becomes increased in high temperature. It is common on parasite reproduction capabilities [5].

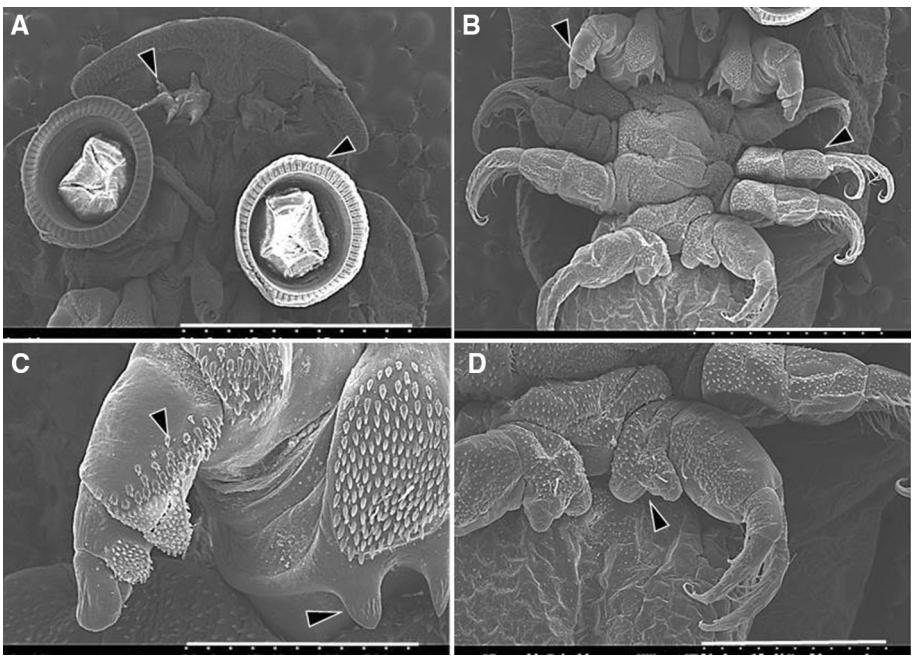

Fig. 3 Scanning electron microphotographs of *Argulus quadristriatus*: antenna (arrow) and ribs of the section cups with 9–11 imbricated plates (**a**); maxilliped and legs 1–4 (**b**); enlarged maxilliped showing spinules (upper arrow) and stout spines (lower arrow) (**c**); enlarged leg 4 showing spinules all over the region and natatory lobe (arrow) (**d**), and Scale bars: **a**, **b** 2 mm; **c** 0.5 mm; **d** 1 mm

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Table 1 The distribution of *Argulus quadristriatus* on different parts of body of fish cobia

Body parts	Numbers of parasites			
	Total	Dorsal	Ventral	Lateral
Head	$440^{Aa} \pm 4.00$	$350^{a} \pm 10$	$50^{c} \pm 18$	$120^{b} \pm 13$
		Dorsal	Ventral	Lateral
Trunk	$100^{Cb} \pm 2.50$	$90^{b} \pm 12$	$60^{c} \pm 11$	$140^{a} \pm 19$
		Dorsal	Caudal	Anal
Fins	$300^{Bb} \pm 1.61$	$450^{a} \pm 15$	$260^{b} \pm 34$	$170^{c} \pm 21$
		Left	Right	
Operculum	$390^{Aa} \pm 2.76$	$370^{b} \pm 24$	$400^{a} \pm 71$	

Mean value in the first column under each category with different superscripts (A, B and C) differ significantly ($p < 0.05$). Mean value in the row under each category with different superscripts (a, b, c and d) differ significantly ($p < 0.05$). Values are expressed as mean ± SE (n = 30)

Some of *Argulus* spp. are causing epizootic in freshwater fish farms and also in wild freshwater fishes all around the world [1, 5, 13, 15], but similar reports from marine fishes were not observed so far and hence the present investigation is a first report on marine argulid infection on fish cobia under pond culture system. We predict that introduction of argulid into the pond might have occurred during stocking itself, eggs or nauplii stages would have been carried over by the procured fish and it further suggests that argulids could appear on fish after few months of stocking. This case has not been reported so far in India, but the present investigation of unexpected infection gives a warning to fish farmers and considered as a serious pest for cobia culture. Since, cobia broodstock culture is rapidly expanding in India and other countries, the present report would give a new insight on the infection of this pest in marine fish and to initiate a study on control, it is one of our ongoing research activities.

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