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Original Research Article

A New Species of the Genus *Dicauda, Dicauda manipurensis* sp. nov. (Cnidaria: Myxosporea: Myxozoa) from fresh water fish *Esomus danrica* (Hamilton, 1822) from Manipur, India

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ABSTRACT:
Myxozoa, a clade of cnidarian, are diversified endoparasite occasionally highly problematic parasites of fish. A new myxozoan parasite belonging to the genus *Dicauda* have observed from Manipur, a region of Indo-Burma Biodiversity Hotspot. Morphologically and morphometrically the species is different from the known species so far described all over the world. The present species was identified from gill lamellae and fin of fresh water fish *Esomus danrica* collected from Thoubal "24˚64' 03.43" N & 93˚98' 25.24" E", Manipur, India and described as *Dicauda manipurensis* sp. nov.

Keywords: Cnidaria, Myxozoa, Endoparasite, *Dicauda*, India-Burma Biodiversity Hotspot, *Esomus danrica*, Manipur, India.

INTRODUCTION

The State Manipur is situated in the extreme North Eastern border of India and is surrounded by lofty hills on all its sides with distinct geographical entity. The state is considered to be the meeting place of Eastern Himalaya and the Burmese region thus the fish fauna of Manipur is of special interest in the fact that it is drained by two major rivers system of the world; The Barak drainage system and The Chindwin-Irrawaddy system. The varied water bodies like lakes, streams, rivers etc. are also available which give rise to a rich fish fauna diversity with different morphological adaptations and colourations. Such is the richness of the water body of Manipur which one would love to called a little paradise on earth that one would discovered a gold mine of fishes. Fish is an important source of protein for the people of Manipur (Manipuries). Hora, 1921 while reporting on the fish and fisheries of the state remarked "owing to religious tenets the Manipuries are forbidden any kind of animal food except fish, which thus form a very important item in the diet". But these fishes are not free from parasites. Myxozoa (Grassé, 1970) is an economically and ecologically important group of Cniderian parasite parasitize to aquatic invertebrates, fishes, reptiles, birds, mammals including human worldwide (Fiala *et. al.*, 2015). Hence study on Myxozoan parasite of fishes

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became utmost important in the state in view of human health care as the people of Manipur have the habit of eating fish as a staple food item. So far more than 16 species of Myxozoan (a clade of cnidarian, Okamura *et.al.,* 2015) parasites are known to infect fresh water fishes from Manipur belonging to the genus *Myxobolus, Thelohanellus, Henneguya, Myxidium* (Soni *et.al.,* 2018). In this present communication a new species of Myxozoa belonging to the genus *Dicauda* infecting the fresh water fish *Esomus danrica* from Thoubal, Manipur, India is described. Complete morphology, morphometry with illustrations and photomicrographs of the new species *Dicauda manipurensis* sp.n is presented.

MATERIAL AND METHODS

Host fishes of about 6-7 cm in length were collected alive from a pond of Thoubal, brought to the laboratory for examination of myxozoan parasites. Most of the body parts were examined viz. body surface, scale, fins, gills, intestine, airbladder etc. under stereoscopic microscope for plasmodia (cyst). Sporogonic plasmodia when found, were carefully removed with the help of sterile forceps, teased on a clean grease free slides with 0.5% NaCl solution (physiological saline), covered with coverslip and examined under the oil immersion lens of Olympus CX41phase contrast microscope otherwise fresh smear of every parts of the body were prepared for examination. Some slides containing myxozoans were treated with Indian Ink for detection of mucus envelop, some were treated with Lugol's Iodine solution for detection of iodinophilous vacuoles and some were treated with KOH solution for extrusion of polar filament. For permanent slide preparation, some of the smeared slides were air dried, fixed in acetone free absolute methanol, stained with Giemsa and mounted with DPX. Measurements (in micrometre) were taken with the aid of a calibrated ocular micrometre. Photographs were taken using an Olympus CX41 Phase contrast microscope with an attached Olympus digital camera.

RESULTS

Cyst: Not found

Spore: The mature spores are elongated, ellipsoidal with broadly rounded anterior end. The posterior end is somewhat narrower than the anterior end. There is a distinct sutural ridge which is slightly curved in sutural view. In morphometry, the length of the spore is 12.19 – 15.24 (13.56 ± 1.03) µm and 7.11 –7.62 (7.26 ± 0.33) µm in breadth. The thickness of the spore is 6.10 – 6.60 (6.17 ± 0.18) µm.

Two polar capsules, spheroidal in shape having neck, occupy the anterior region of the spores. The polar capsules are arranged parallel and open anteriorly at two different opening. The capsule which are pointed anteriorly and broadly rounded posteriorly have the capsular dimension of 3.05 – 4.06 (3.50 ± 0.50) µm in length and 2.03 – 2.54 (2.31 ± 0.25) µm in breadth. The polar capsule houses the polar filament but its turn inside the polar capsule is indistinct. Three-fourth of the extra-capsular region is occupied by homogenous granules of sporoplasm. Sporoplasmic nucleus and iodinophilous vacuoles are not observed in the sporoplasm.

At the posterior part of the spore two caudal appendages are observed extending in opposite direction. These two appendages are not a continuation of the smooth shell valves but appear to adhere to the shell valves along a distinct boundary.

Intra-specific Variation

Intra-specific variations among the spores are observed distinctly. Some spores have polar capsules at different level while some have more spherical polar capsules. Some also have cylindrical spore having closer and smaller caudal appendages. In some other spores the caudal appendages are extra-large. All these variations are regarded as intra specific variations.

Seasonal Intensity

The present species was first collected in the month of April, 2014 from Thoubal Ningombam, (Lat. 24˚64′03.43′′N & Long. 93˚98′25.24′′E). During the study period, the species was found mostly in summer and rainy season. Out of 40 host fish were examined 20 (50%) host fishes were infested.

Taxonomic Affinities

The present myxozoan with two caudal appendages extending in opposing direction with two polar capsules at the anterior end of the spore agree with the diagnostic characters of the genus *Dicauda* eracted by Hoffman and Walker, 1978. *Dicauda atherinoidi* Hoffman and Walker, 1978 was reported from subcutaneous tissue of yearlings of *Notropis atherinoides* in the USA. The spore's body were almost circular having spored dimension 10.3µm in length and 9.3 µm in breadth. The two polar capsules were pyriform in shape having spore dimension 5.2 µm in length and 3.3 µm in width with 3 to 6 filament turns. The appendages were about 50 µm in length. But the present species have elongated, ellipsoidal spores with rounded anterior end having spore dimension 12.19 – 15.24 (13.56 ± 1.03) µm in length and 7.11 –7.62 (7.26 ± 0.33) µm in breadth. Two polar capsules were spheroidal in shape, having 3.05 – 4.06 (3.50 ± 0.50) µm in length and 2.03 – 2.54 (2.31 ± 0.25) µm in breadth. The appendages was about 8.13 – 12.19 (9.50 ± 1.41) µm in length. When the present species is closely compared morphologically with the species *Dicauda atherinoidi* as cited above, it can be confirmed that the present species is new to science and designated as *Dicauda manipurensis* sp. nov. This is communicated for the first time from India and most probably will be the second species of the genus *Dicauda* after its introduction by Hoffman and Walker in 1978.

Diagnostic Character of the Genus *Dicauda*:

Spore ellipsoidal, ovoid or rounded in valvular view and biconvex in sutural view. Shell valves smooth. Two polar capsules found mostly pyriform exceptionally. One is extensively missing. Posteriorly, the sutural ridge may extend into a crescentic edge. Binucleate sporoplasm, often with the iodinophilous vacuole. Spores with two caudal appendages, extending in opposing direction, which is not a continuation of the smooth shell valves, but a structure made up from a different material and adhering to the shell valves along a distinct boundary.

Table 1: Morphometric Data in µm of 20 fresh spores of *Dicauda manipurensis* sp. nov.

Character	Range	Mean	S.D.	S.E.	C.V.
Spore Length	12.19 -15.24	13.56	1.03	0.23	23.02
Length of Caudal Prolongation	8.13 -12.19	9.50	1.41	0.31	31.54
Spore Wide	7.11 - 7.62	7.26	0.33	0.05	5.20
Spore Thickness	6.09 - 6.60	6.17	0.18	0.04	4.05
Length of Polar Capsule	3.05 - 4.06	3.50	0.50	0.11	11.30
Wide of Polar Capsule	2.03 - 2.54	2.31	0.25	0.056	5.65

Taxonomic Summary

Type Host : *Esomus danrica* (Hamilton, 1822)
Type Locality : Thoubal Ningombam, (Lat. 24˚64´03.43´´N & Long. 93˚98´25.24´´E)
Infestation site : Gill lamellae, Fins.
Prevalence : 20/40 (50%).
Intensity rate : High.
Type Material : Slide containing holotype ($MU/LSD/P/D_{i-a}$) and paratype ($MU/LSD/P/D_{i(b-e)}$) deposited in Parasitology Section, Department of Life Sciences, Manipur University.
Etymology : Species name derived from Manipur, name of the state from where the species collected.

DISCUSSION

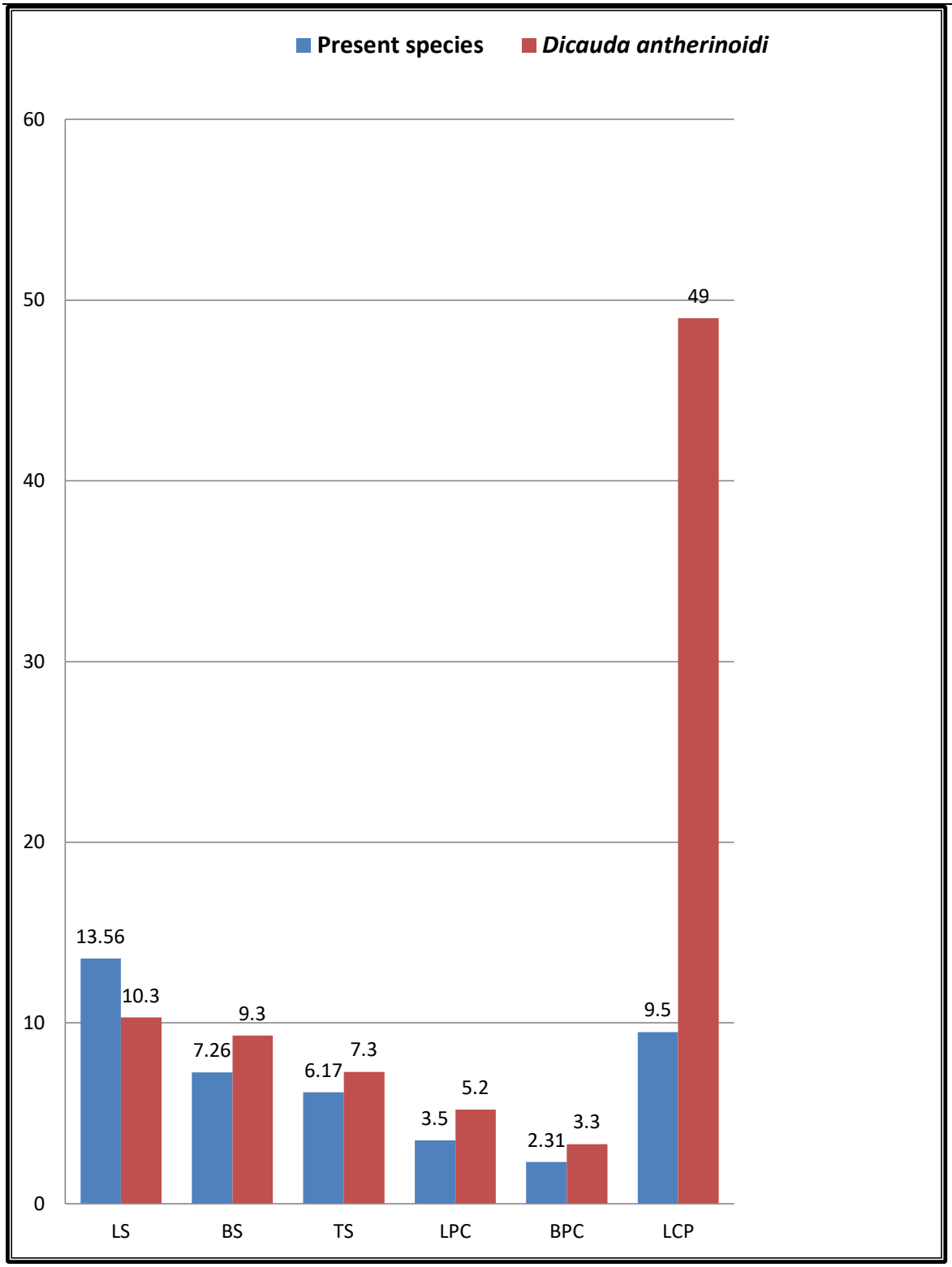
The genus *Dicauda* was erected by Hoffman and Walker (1978). When they introduced the genus, there was a single species, *Dicauda atherinoidi*. They differentiated the genus from *Unicauda* in having two caudal appendages, extending in opposing direction instead of having a single caudal appendage as in *Unicauda*, a structure made up from a different material and adhering to the shell valves along a distinct boundary.

Dicauda atherinoidi (Hoffman and Walker, 1978) was reported from subcutaneous tissue of yearlings of Notropis atherinoides in the USA. The spores body were almost circular having spore dimension 10.3µm in length and 9.3 µm in breadth. The two polar capsules were pyriform in shape having spore dimension 5.2µm in length and 3.3 µm in width with 3 to 6 filament turns. The appendages were about 50 µm in length.

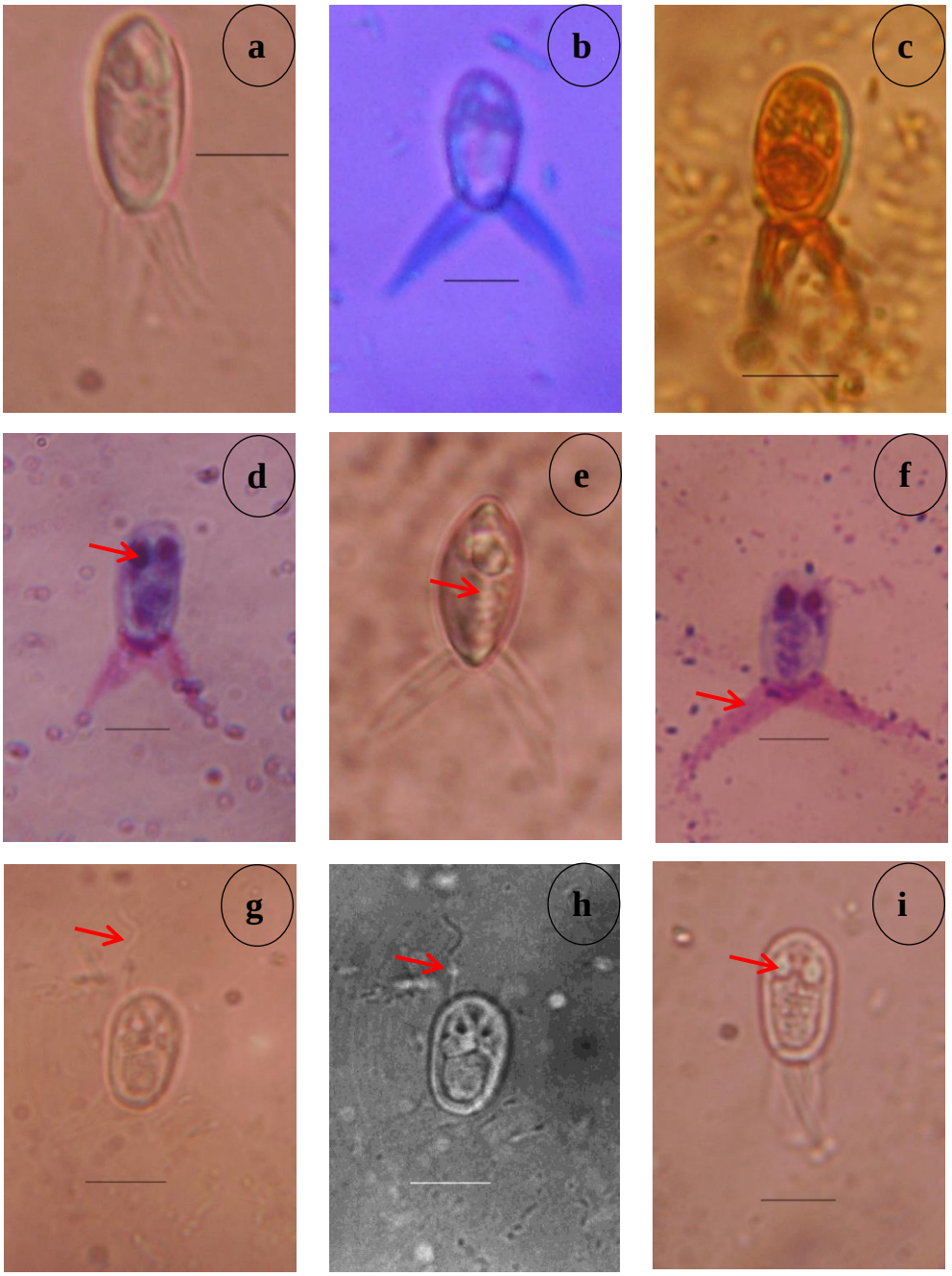
As far as literature was concerned Loch *et al.,* (2017) the description of *Dicauda atherinoidi* (Bivalvulida: Myxobolidae) with respect to morphological and histopathological characters had updated and host range expansion were also supplemented with the first molecular data and phylogenetic analyses of the genus. Interestingly a species of this genus was encountered during the survey of Myxozoan parasites in the ornamental fishes of Manipur. The species was closely compared morphologically with the available species of the genus *Dicauda* and confirmed that the present species belong to genus *Dicauda*. This had been reported for the first time from India and as the second species of the genus *Dicauda* after its introduction by Hoffman and Walker in 1978.

Table 2: Showing morphometrical differences between *Dicauda atherinoidi* Hoffman and Walker, 1978 and present species

Character	***Dicauda atherinoidi***	***Present species***
Length of Spore (LS)	10.3 (8.5 –12.5)	12.19 -15.24 (13.56 ±1.03)
Breadth of Spore (BS)	9.3 (8.5 –11.0)	7.11 – 7.62 (7.26 ± 0.33)
Thickness of Spore (TS)	7.3 (7.0 – 9.0)	6.10 – 6.60 (6.17 ± 0.18)
Length of Polar Capsule (LPC)	5.2 (4.0 - 7.0)	3.05 – 4.06 (3.50 ± 0.50)
Breadth of Polar Capsule (BPC)	3.3 (2.5 – 4.0)	2.03 – 2.54 (2.31 ± 0.25)
Length of Caudal Prolongation (LCP)	49 (25 – 73)	8.13 – 12.19 (9.50 ± 1.41)
Angle length(º)	70 - 170	30 - 90
Site of Infection	Subcutaneous tissue	Gills & Fins
Host	*Notropis antherinoides*	*Esomus danrica*
Reference	Hoffman and Walker, 1978**.**	Present communication



Graph 1: showing comparative morphometrics differences between *Dicauda antherinoidi* and the present species.



Figures 1 (a-i): Photomicrographs of *Dicauda manipurensis* sp. nov. Bar –5.54 µm.

a: Mature spore in fresh condition.
b: Spore in stain using Indian-Ink Technique.
c: Spore in Lugol's Iodine solution.
d: Mature spore in valvular view showing polar capsule in Giemsa stain.
e: Mature spore in sutural view.
f: Spore showing caudal extension.
g & h: Spores in showing protruded polar filaments.
i: Mature spore showing polar capsule in fresh condition.

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