Proc. Natl. Acad. Sci., India, Sect. B Biol. Sci. (July–Sept 2021) 91(3):635–641
https://doi.org/10.1007/s40011-021-01253-y

RESEARCH ARTICLE

Prevalence of Myxozoan Parasites of Riverine Fishes of Jalpaiguri District, West Bengal, India

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Received: 26 March 2020 / Revised: 26 March 2020 / Accepted: 24 February 2021 / Published online: 29 April 2021

Abstract A survey on prevalence of myxozoan (Myxozoa: Myxosporea: Bivalvulida) parasites was conducted during the 2014–2017 season from two major rivers, Mahananda and Teesta of Jalpaiguri district. 1874 fishes under twenty different genera of eight families were examined, among which 108 (5.76%) fishes of six different genera of four families were found to be infected. The myxozoans were recorded from the gills and tailfins of the host fishes. Intensity of infections fluctuated according to season. Infection rate was lower in monsoon all through the survey period. Maximum infections were recorded in *Chanda nama* (33%) and *Channa punctatus* (33%) during summer of 2014–15 and winter of 2014–15, respectively. Six host species were infected with six different myxozoan species. Among the six myxozoan parasites, three have been identified as *Myxobolus shantipuri*, *Myxobolus koli*, and *Thelohanellus qadrii*. Rest of the three species have been identified up to generic level and they are designated as *Thelohanellus* sp. I, *Henneguya* sp. I and *Henneguya* sp. II in this communication.

Keywords Myxozoan · *Myxobolus* · *Thelohanellus* · *Henneguya*

Significance statement Myxozoans are responsible for doing considerable harm to the pisciculture. Prevalence study of myxozoans helps to reveal the status of fish health of that particular area which is important both to fish farmers and common people as well.

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Introduction

Jalpaiguri, one of the northern most districts of West Bengal, lies at the center of vast fertile plains of Terai at southern Himalayan foothills. It is watered by innumerable rivers and rivulets like Mahananda, Neora, Jaldhaka, Teesta, Torsa, etc.; and hilly jhoras rising from and flowing down the Himalayas and enrich the district both economically and naturally. The climate has undergone a remarkable change since the past few years, with the soaring temperature and the decreasing rainfall each year [1]. The water qualities of the river Mahananda and Teesta, the lifelines of north Bengal, are also degrading due to developmental pressure like deforestation, indiscriminate constructions at the riversides of Teesta, overpopulation on the riversides of Mahananda and Teesta, series of dams on Teesta, dumping of sewage via open drains into Mahananda and stagnation of water [2]. All these factors have a great impact on ecological, social, cultural, religious, aesthetic, tourism-related and economic aspects of these rivers [3]. In the near past, the region was well known for its rich fish diversity [4] and there has been no record of serious disease infestation. In the changing scenario, a preliminary survey on prevalence of myxozoan (Myxozoa: Myxosporea: Bivalvulida) parasites was conducted during the 2014–2017 season from two major rivers of Jalpaiguri district to evaluate the threat imposed by the developmental activities on fishes.

Material and Methods

The fish specimens were collected from the two sample collection sites of Jalpaiguri district for three consecutive years; i.e., from March 2014 to February 2017. Sample

Table 1 Fishes examined for myxozoan infection from the rivers of Jalpaiguri district during the survey period 2014–17

Sl no	Fish family	Local name	Scientific name	No. of fish examined	Source of fish collection	Infected/ Not infected
1	Cyprinidae	Chela	*Salmophasia bacaila* Hamilton, 1822	96	Mahananda Bridge	Not infected
2	,,	Mourola	*Amblypharyngodon mola* Hamilton, 1822	122	Mahananda Bridge and Teesta barrage	Infected
3	,,	Darkina	*Rasbora daniconius* Hamilton, 1822	102	Bridge and Teesta barrage	Infected
4	,,	Koksa	*Barilius shacra* Hamilton, 1822	100	Teesta barrage	Not infected
5	,,	Chebli	*Devario aequipinnatus* McClelland, 1839	95	Teesta barrage	Not infected
6	,,	Punti	*Puntius sophore* Hamilton, 1822	109	Mahananda Bridge and Teesta barrage	Infected
7	,,	Boreli	*Barilius vagra* Hamilton, 1822	105	Mahananda Bridge and Teesta barrage	Not infected
8	Cobitidae	Gutum	*Lepidocephalichthys annandalei* Chaudhuri, 1912	88	Mahananda Bridge	Not infected
9	,,	Gunte	*Lepidocephalichthys guntea* Hamilton, 1822	89	Mahananda Bridge	Not infected
10	,,	Bilturi	*Acanthocobitis botia* Hamilton, 1822	102	Teesta barrage	Not infected
11	,,	Nodiari	*Somileptes gongota* Hamilton, 1822	103	Teesta barrage	Not infected
12	,,	Bou Mach	*Botia dayi* Hora, 1932	84	Teesta barrage	Not infected
13	Siluridae	Pabda	*Ompok pabda* Hamilton, 1822	79	Mahananda Bridge	Not infected
14	,,	Madhu pabda	*Ompok pabo* Hamilton, 1822	94	Teesta barrage	Not infected
15	Bagridae	Tangra	*Mystus vittatus* Bloch, 1794	89	Mahananda Bridge and Teesta barrage	Infected
16	Ambassidae	Rup chanda	*Chanda ranga* Hamilton, 1822	90	Mahananda Bridge and Teesta barrage	Not infected
17	,,	Tak chanda	*Chanda nama* Hamilton, 1822	88	Mahananda Bridge and Teesta barrage	Infected
18	Badidae	Bot koi	*Badis badis* Hamilton, 1822	58	Teesta barrage	Not infected
19	Ophiocephalidae	Lata	*Channa punctatus* Bloch, 1794	84	Mahananda Bridge and Teesta barrage	Infected
20	Gobiidae	Bele	*Glossogobius giuris* Hamilton, 1822	97	Teesta barrage	Not infected

Table 2 Percentage and site of infection of myxozoans, in different host fishes of Mahananda and Teesta River during the survey period 2014–2017

Host fishes	Site of infection	Infecting myxozoan species	No. of fish examined	No. of fish infected	Percentage of infection (%) in three different seasons during the survey								
					2014–15			**2015–16**			**2016–17**		
					S	**M**	**W**	**S**	**M**	**W**	**S**	**M**	**W**
Rasbora daniconius Hamilton, 1822	Gills	*Myxobolus shantipuri*	102	22	25	08	25	29	09	18	27	18	38
Amblypharyngodon mola Hamilton, 1822	Gills	*Thelohanellus qadri*	122	23	25	12	19	29	17	13	20	08	33
Puntius sophore Hamilton, 1822	Tailfin	*Myxobolus koli*	109	19	29	08	17	29	11	29	25	10	10
Channa punctatus Bloch, 1794	Tailfin	*Henneguya* sp. I	84	14	22	13	33	11	08	20	09	20	17
Chanda nama Hamilton, 1822	Gills	*Thelohanellus* sp. I	88	15	33	10	08	27	11	14	29	13	20
Mystus vittatus Bloch, 1794	Gills	*Henneguya* sp. II	89	15	25	08	25	22	13	27	30	10	09

S = Summer, M = Monsoon, W = Winter

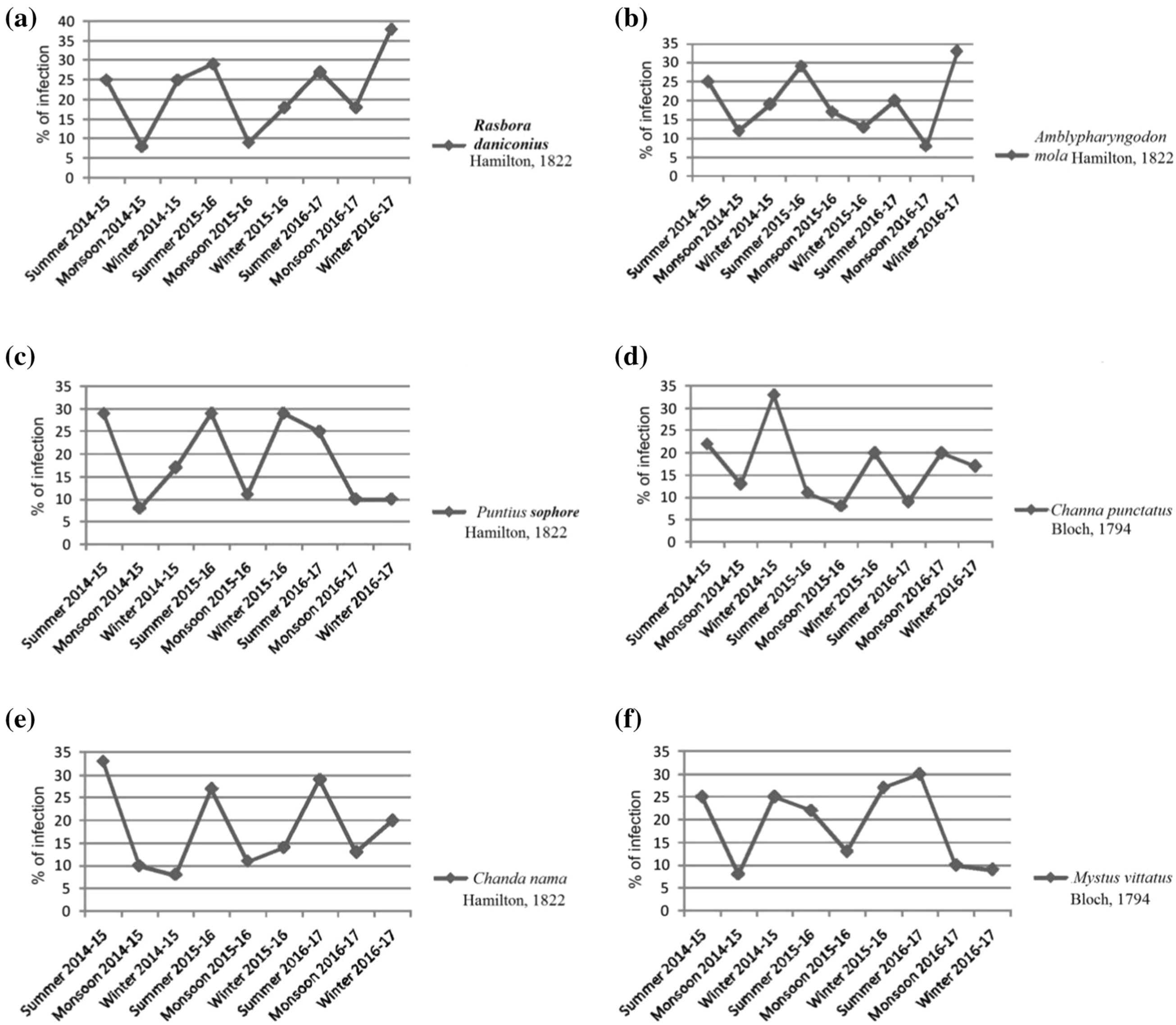

Fig. 1 Seasonal fluctuations of myxozoan infections of six host fishes of Jalpaiguri district during the survey period 2014–17 (**a** – *Rasbora daniconius,* **b**—*Amblypharyngodon mola,* **c** – *Puntius sophore,* **d**—*Channa punctatus,* **e**—*Chanda nama* and **f**—*Mystus vittatus*)

collection sites of Mahananda and Teesta rivers were located at Mahananda Bridge (Latitude 26°71′N, Longitude 88°41′E), Siliguri and Teesta barrage, Gajoldoba (Latitude 26°75′N, Longitude 88°58′E), respectively. The fishes were examined thoroughly for myxozoan parasites and were examined at least twice in every season during the study period to record the seasonal fluctuations of infection. Plasmodia, when found, were carefully removed from the host body parts with the help of a sterile forceps and smeared on clean grease-free slides with drops of 0.5% NaCl solution. Some of the fresh spores were treated with 5% of KOH solution for the extrusion of polar filaments. The spores were fixed in absolute methanol. For examination of the spores, the Giemsa-stained slides were placed under the oil immersion lens (Leica DM 2500 Light microscope). Mucus envelopes of spores were observed by following the method of Lom and Vavrá [5]. Measurements (based on fifty fresh spores treated with Lugol's iodine) were taken with the aid of a calibrated ocular micrometer. Myxozoans are identified from standard literature [6–8]. The fish samples were identified after Dey [9] and Talwar and Jhingran [10]. Classification of fishes was followed on lines of Jayaram [11]. All the ecological terms used in this communication are in conformity with the Margolis [12]. The prevalence is expressed in percentage and is calculated by total number of infected fishes divided by total number of host fishes examined [13].

Prevalence of infection
= (Total number of infected fishes /
Total number of host fishes examined) × 100

Results and Discussion

During the course of the survey from March 2014 to February 2017, 20 species of fishes under 8 different families were examined, out of which 6 species of fishes under 4 different families, viz., Cyprinidae, Ophiocephalidae, Ambassidae and Bagridae were found infected with 6 different myxozoan species (Table 1). However, 3 host fishes out of 6 belonged to family Cyprinidae.

Altogether, 108 fishes out of 1874 fishes examined were found to be infected with myxozoan parasites, i.e., 5.76% of fishes were found infected during the survey period. The average percentage of infection recorded highest in *Rasbora daniconius* (21.6%) and lowest in *Channa punctatus* (16.7%). Infections were recorded only from gills and tail fins (Table 2). The percentage of infection of the host fishes in different seasons was also calculated during the three years survey period (Table 2). The overall rate of infections was recorded high during summer (March to June) and winter (November to February) and comparatively low in Monsoon (July–October) throughout the study period (Fig. 1). These kinds of observations could be due to apparent scarcity and abundance of water at the respective seasons. The highest seasonal incidences were recorded in *Chanda nama* (33%)

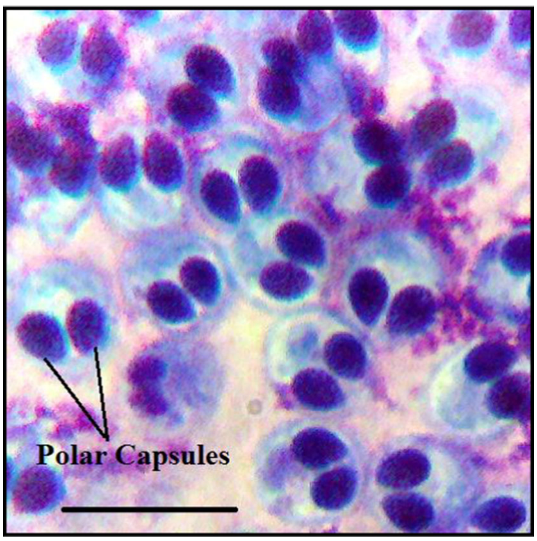

Fig. 2 Giemsa-stained spores of *Myxobolus shantipuri* Basu and Haldar, 2002 (Bar = 10 μm)

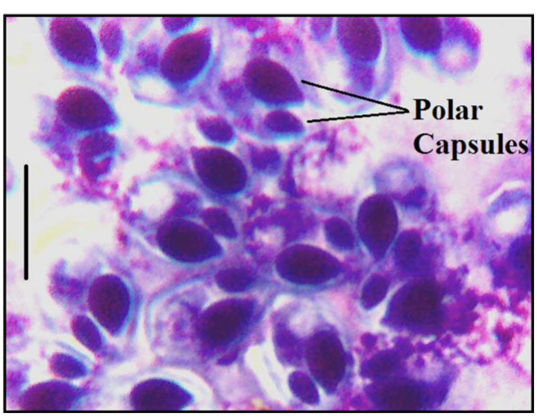

Fig. 3 Giemsa-stained spores of *Myxobolus koli* Lalitha Kumari, 1969 (Bar = 10 μm)

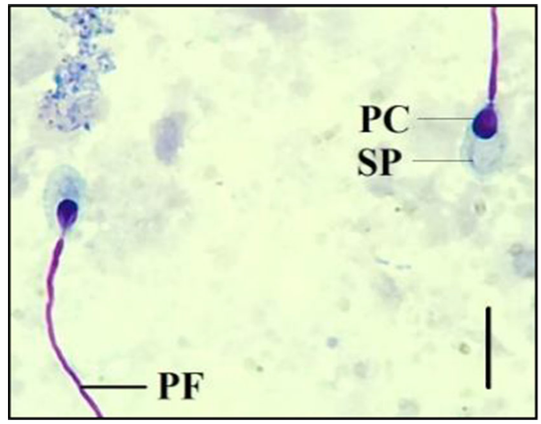

Fig. 4 Giemsa-stained spores of *Thelohanellus qadrii* Lalitha Kumari, 1969 [PC-Polar Capsule, PF-Polar Filament, SP-Sporoplasm] (Bar = 10 μm)

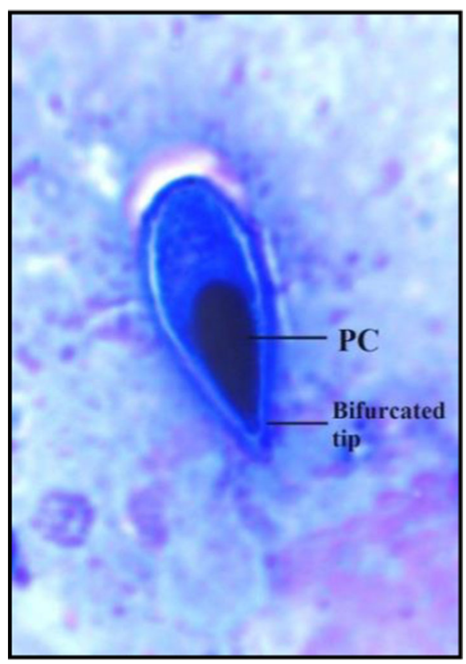

Fig. 5 Giemsa-stained spores of *Thelohanellus* sp. I [PC-Polar Capsule] (Bar = 10 μm)

Table 3 Comparative characters of six myxozoans collected in Jalpaiguri district during the survey period 2014–2017

Characters	Myxozoan species I	Myxozoan species II	Myxozoan species III	Myxozoan species IV	Myxozoan species V	Myxozoan species VI
Host	*Rasbora daniconius* Hamilton, 1822	*Puntius sophore* Hamilton, 1822	*Amblypharyngodon mola* Hamilton, 1822	*Chanda nama* Hamilton, 1822	*Channa punctatus* Bloch, 1794	*Mystus vittatus* Bloch, 1794
Genus characters	i) Spores are pyriform and thick shell valves are present ii) Two polar capsules are present and might be equal or unequal The above characters show that the specimen belongs to the genus *Myxobolus*	i) Spores are pyriform and thick shell valves are present ii) Two polar capsules are present and might be equal or unequal The above characters show that the specimen belongs to the genus *Myxobolus*	i) Spores are pyriform or tear shaped ii) Shell valves are thin iii) Single pyriform polar capsule is present The above characters show that the specimen belongs to the genus *Thelohanellus*	i) Spores are pyriform or tear shaped ii) Shell valves are thin. iii) Single pyriform polar capsule is present. The above characters show that the specimen belongs to the genus *Thelohanellus*	i) Spores are spindle shaped in valvular view ii) Two caudal appendages are present, originated from the shell valves iii) Two elongated polar capsules are present and might be equal or unequal. The above characters show that the specimen belongs to the genus *Henneguya*	i) Spores are spindle shaped in valvular view. ii) Two caudal appendages are present, originated from the shell valves. iii)Two elongated polar capsules are present and might be equal or unequal The above characters show that the specimen belongs to the genus *Henneguya*
Polar capsule	Two equal polar capsules are pyriform with long neck. They occupy almost half of the spore. Inside the large polar capsule each polar filament makes 4–5 loose coils	Two unequal pyriform polar capsules are present. One is remarkably smaller than the other	Polar capsule is single, large, pear-shaped, terminal with broad rounded posterior region. Polar filament forms 10–14 coils while inside the capsule, long, whip-like when it is extended	One large polar capsule is pyriform in shape. The tip of the polar capsule is bifurcated. Polar capsule occupies almost half of the spore. Inside the polar capsule polar filament makes 6–8 coils	Two unequal polar capsules are pyriform in shape. Two polar capsules are differentiated markedly according to their length. Two polar capsules occupy almost half of the spore	Two unequal polar capsules are elongated and pointed tip placed at the anterior end of the spore. Almost half of the spore is occupied by two polar capsules
Spore morphometry	Spore: 7.9 (7.1–8.9) × 6.9 (6.5–7.4) Polar capsule: 3.9 (3.5–4.1) × 2.7 (2.4–3.1)	Spore: 9.8 (9.2–10.2) × 6.7 (5.9–7.3) Polar capsule (large): 6.7 (5.9–7.3) × 3.1 (2.7–3.5) Polar capsule (small): 3.1 (2.7–3.5) × 1.9 (1.8–2.1)	Spore: 11.9 (11.1–12.6) × 5.15 (4.93–5.39) Polar capsule: 6.66 (6.5–6.8) × 2.94 (2.9–2.98)	Spore: 24.5 (22.9–26.4) × 9.1 (8.0–10.8) Polar capsule: 12.2 (10.9–13.4) × 5.0 (4.2–5.9)	Spore: 11.5 (10–12.8) × 3.4 (3.1–4.2), Polar capsule (large): 6.3 (5.0–7.0) × 1.3 (0.9–1.5) Polar capsule (small): 5.6 (4.5–6.2) × 1.2 (0.9–1.5)	Spore: 16.2 (14.5–17.5) × 4.5 (4.0–5.2) Polar capsule (large): 8.3 (7.0–9.1) × 1.8 (1.43–2.1) Polar capsule (small): 7.4 (6.6–7.9) × 1.6 (1.42–1.91)

Table 3 continued

Characters	Myxozoan species I	Myxozoan species II	Myxozoan species III	Myxozoan species IV	Myxozoan species V	Myxozoan species VI
Species identification	The morphometric data of present specimen resembles totally with ***Myxobolus shantipuri*** Basu and Haldar, 2002. (Fig. 2)	The morphometric data of present specimen resembles totally with ***Myxobolus koli*** Lalitha Kumari, 1969 (Fig. 3)	The morphometric data of present specimen resembles totally with ***Thelohanellus qadrii*** Lalitha Kumari, 1969 (Fig. 4)	The morphometric data of present specimen does not matches with any species available in the literature. In present communication the specimen is considered as ***Thelohanellus*** sp. I (Fig. 5)	The morphometric data of present specimen does not matches with any species available in the literature. In this communication the specimen is considered as ***Henneguya*** sp. I (Fig. 6)	The morphometric data of present specimen does not matches with any species available in the literature. In present communication the specimen is considered as ***Henneguya.*** sp. II (Fig. 7)

(All measurements are in μm)

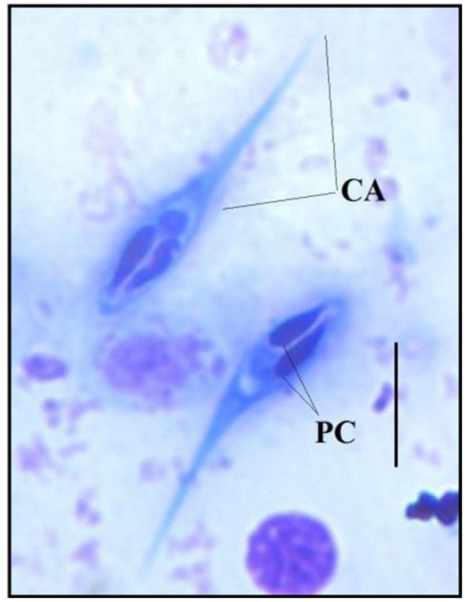

Fig. 6 Giemsa-stained spores of *Henneguya* sp. I [CA- Caudal Appendages, PC-Polar Capsule] (Bar = 10 μm)

and *Channa punctatus* (33%) during summer of 2014–15 and winter of 2014–15, respectively (Table 2).

Microscopical observations of myxozoan species obtained from six different host fishes revealed that all the myxozoan species are different. Three of them are identified as *Myxobolus shantipuri*, *Myxobolus koli* [6] and *Thelohanellus qadrii* [6] (Figs. 2, 3, 4). As per literature available, rest of the species are identified up to generic level only and they are described here as *Thelohanellus* sp. I, *Henneguya* sp. I and *Henneguya* sp. II (Figs. 5, 6, 7). Detail taxonomic information is compiled in Table 3.

Conclusion

Incidences of heavy fish mortality or irreparable loss in fish industries due to protozoan infestation are very common [14]. The myxozoans are parasitic protozoans (recently Fiala et al.[15] placed it under phylum Cnidaria) that inhabit primarily in the tissues and organ cavities of cold-blooded (exothermic / poikilothermic) vertebrates, especially culturable fishes and as such they have an importance in ichthyopathology [14, 16, 17].

The present study records a considerable percentage of myxozoan infection and diversity in the fishes collected from Mahananda and Teesta River. Though, only 5.76% fishes are found infected, but in some host species, it may reach up to 33% in a particular season. Significant infection rate of fishes of these two rivers indicates depletion of water qualities in this region. North Bengal is well known for its icthyofaunal diversity with 141 fish species in the three districts of north Bengal, viz. Cooch

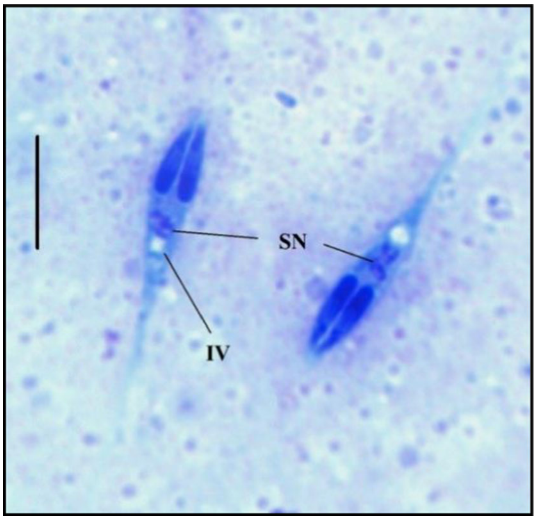

Fig. 7 Giemsa-stained spores of *Henneguya* sp. II [IV- Iodinophilous Vacuole, SN- Sporoplasmic Nuclei] (Bar = 10 µm)

Behar, Alipurduar and Jalpaiguri [4]. However, in the past few years, freshwater habitats of this region are drastically modified due to various anthropogenic activities, which have profound effect on fish health [18]. In the changing environment, fishes are stressed with suppressed immune system and they are easily getting infected [19]. So, there is an urgent need to plan comprehensively for management of these aquatic bodies for sustainable utilization of aquatic resources. Proper riverine environment is crucial to preserve the endemic and endangered species from becoming extinct due to anthropogenic stresses. However, before coming into final conclusion, further investigations are required in search of myxozoan as well as other fish diseases of other aquatic bodies of north Bengal and it should be correlated with other parts of West Bengal.

Acknowledgement First author would like to thank the Department of Zoology of Sidho-Kanho-Birsha University, Purulia for giving permission to do the work. Corresponding author is thankful to DST-FIST [No. SR/FST-LSI/2018/173(c) dated 18/09/2019 for infrastructural support. Authors are also like to express their sincere thanks to the fishermen of the district who generously spent time during the survey and shared their expertise in connection with the local fishes.

Declarations

Conflict of interest The authors declare that there is no conflict of interest.

Ethical Approval Not required as per the guidelines of Committee for the Purpose of Control and Supervision of Experiments on Animals (CPCSEA).

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