International Journal of Fisheries and Aquatic Studies 2018; 6(2): 202-206

E-ISSN: 2347-5129
P-ISSN: 2394-0506
(ICV-Poland) Impact Value: 5.62
(GIF) Impact Factor: 0.549
IJFAS 2018; 6(2): 202-206

www.fisheriesjournal.com
Received: 17-01-2018
Accepted: 19-02-2018

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Parasitic study of Indian major carp, *Catla catla* (Hamilton, 1822) from Bheries in West Bengal, India

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Abstract
The present work was conducted to isolate and identify different parasites from Indian Major Carp (IMC) *Catla catla*. Parasitic Frequency Index (PFI, %) and Severity of infection were also measured for each parasite from Bheries (Saline soils) of Garia, Bantala, Bamanghata, Gangajuara of South 24 Parganas District, Naihati of North 24 Parganas District, Memari of Burdwan District of West Bengal, India. Approximately 300 fishes were observed in between April 2012 to March 2013. The infested fishes suffered mainly from respiratory manifestations, blackness of the skin and mortalities. The parasitic infestations were found to be the most important problem in Bheries (Saline soils) of selected districts in West Bengal. A large amount of parasitic disease causative agents were isolated and those were *Myxobolus* sp., *Thelohanellus* sp., *Trichodina* sp., *Dactylogyrus* sp., *Gyrodactylus* sp., Nematodes, *Argulus* sp., *Lernea*sp., *Chilodonella* sp., The prevalence of *Myxobolus* sp., *Dactylogyrus* sp. and *Gyrodactylus* sp. were highest in October to January (winter months, 73%, 56% and 13.3% respectively). Remaining all parasites were found more in February to March (spring season months).

Keywords: Catla catla, parasites, isolation, identification, PFI (%), severity, West Bengal

1. Introduction
India's aquaculture production can be classified into freshwater and brackish water production. The development support provided by the Indian Government through a network of 429 Fish Farmers Development Agencies (FFDA) and 39 Brackish Fish Farmers Development Agencies (BFDAs) [1]. These two Agencies are the vehicles for the development and growth of freshwater and coastal aquaculture. The three Indian major carps, namely Catla (*Catla catla*), Rohu (*Labeo rohita*) and Mrigal (*Cirrhinus mrigala*) contribute 87% of total fresh water aquaculture production [3]. West Bengal being a ‘rice-fish society’ the state is highly significant historically, geographically and strategically since long past [17]. According to annual report, 2016-17, the state has 37% of pond resources in India of which 70% are utilized for fish culture producing 1 to 3 million tonnes of freshwater fin- fish per year of the total 2.76 lakh hectares of impounded water area, about 70% to 79% is presently under fish culture [1].
Catla, *Catla catla* is one of the most important species of inland fisheries of West Bengal. The species is widely cultured all over the West Bengal and in India [12]. The production from culture system is hampered by the infestation of various fish parasites. The importance of fish parasite is related directly to the importance of fish health [11]. Fish parasites are the primary damaging agents during stress conditions followed by bacterial and fungal infection. The fish parasites directly feed on the digested contents of the host’s intestine or host tissues. The parasites multiply rapidly during stress condition or under favorable conditions (Dogiel, 1956), and cause economic loss, regularly cause high mortality [23]. In this study, we examined the prevalence of parasites found in Labeo rohita for understanding the relationship and severity with the fish size from different selected districts of West Bengal. Further studies needed on the life cycle, mode of infection, host-pathogen relationships and pathogenicity of parasites of carps with more emphasis on parasites.
The objectives of the present study were to isolate and identify different parasites from Indian Major Carp (IMC) *Catla catla* and to find out Parasitic Frequency Index (PFI, %) at month wise from different selected districts of West Bengal.

2. Materials and Methods

The present study about prevalence of parasitic infestation on Indian Major Carps was carried out for a period of 12 months from April 2012 to March 2013. The samples were collected from Garia, Bantala, Bamanghata, Gangajuara of South 24 Parganas District, Naihati of North 24 Parganas District, Memari of Burdwan District of West Bengal, India. The locations were selected in such a way that these units at different locations represent the concern district. The samples were collected on a regular basis once in every month. The fishes were brought to the laboratory in live condition with water filled buckets and the total lengths, body weight of fishes were taken. The vital organs like skin, intestine, kidney and gills were examined for the presence of different parasites.

The methods for collection and preservation of the samples for parasitic examination were followed as described by [20] and [16]. Phenotypic characterization of all protozoans, monogenians, digeneans, and nematode parasites were studied as described by [21]. Photomicrographs were taken using a Motic BA400 phase contrast microscope with in-build digital camera.

2.1 Determination of Parasitic Frequency Index (PFI)

The parasitic frequency index (PFI) was calculated by taking the percentage of the number of hosts infected by an individual parasite species against the total number of hosts examined in a particular area under investigation.

$$\text{Prevalence}(\%) = \frac{\text{Total number of infected fishes}}{\text{Total number of fish host examined}} \times 100$$

The frequency index were further classified into rare (0.1 – 9.9%), occasional (10-29.9%), common (30 – 69.9%) and abundant (70-100%) as per [22].

2.2 Determination of severity of infection/ infestation

In order to assigning numerical qualitative value to severity grade of infections surface infestation and disease syndrome severity, the generalize scheme by [14] was followed.

2.2.1 Statistical Analysis

Two way ANOVA was done to determine the significance of differences in Parasitic Frequency Index (PFI) of parasites among different seasons as well as different months and different length groups. It was also followed to determine the significance of differences in prevalence of parasites in different organs of the fishes as well as length groups of fishes. The critical difference (CD) was calculated to examine which of the source and month differed significantly [19].

3. Results and Discussion

Month wise distribution of parasites in *Catla catla* is presented in Table 1. Severity of infestation of parasites also studied in IMC during the study period (Table 1).

Parasitic Frequency Index (PFI) of *Myxobolus* sp. were highest during November, December and January (PFI, 84.61%, 86.95%and 95%), which were stated as ‘abundant’. These results are supported by Basu *et al.,* [5] who have recorded high prevalence of myxozoan parasites during August to January when the ambient temperature was 25°C and lowest prevalence in the month of July (16.66%). In the present study no infestation was recorded in the months of May, but Kaur *et al.,* [12] reported 100% prevalence of infection in the month of May and the probable reason might be the different in geographical area. Also in March the PFI of *Myxobolus* sp. were more (PFI, 86.36%) which was also stated as’ abundant’. In rest of the months April, July, August and October the conditions were ‘occasional’ and remaining months June, September and February the prevalence was ‘common’.

PFI of *Thelohanellus* sp. varied from 4.76% to 40.9%. The occurrence were high during February and March (PFI, 38.46% and 40.9%) and this indicates that the prevalence of these parasite were ‘common’. This could be due to high stocking density, water depth, temperature along with other physiochemical parameters and management practices maintained in culture systems these results strongly supported with results of Banu *et al.,* [4]. During August and October the occurrence were lowest (PFI, 7.4% and 4.76%), and the condition was ‘rare’. In rest of the months the prevalence of this parasite was ‘occasional’.

PFI of *Trichodina* sp. varied from 8.69% to 70.37%. The occurrences were highest in the month of August (PFI, 70.37%) and prevalence of these parasites was ‘abundant’ during this month. It was lowest in the month of December (PFI, 8.69%), and the condition was stated as ‘rare’. In rest of the months April, May, June, February and March the prevalence of these parasites were ‘common’. Fish confined to ponds, tanks and aquaria, trichodiniasis is a frequent problem Arthur and Lom [2]. Trichodinds are opportunistic parasites which become pathogenic under stress full conditions, the present findings were corroborated with works of Mc Ardle [15] and Eisa *et al.,* [9].

PFI of *Gyrodactylus* sp. were highest in the month of August (PFI, 37.03%), and the condition were ‘common’. and lowest in the month of September (PFI, 9.52%). In July, October, December, January and February the condition were ‘occasional’ and in rest of the months they were not found.

PFI of *Dactylogyrus* sp. was highest in the month of January and August (PFI, 90% and 88.88%) which was ‘abundant’ condition. Lowest prevalence recorded in the month of June (PFI, 4%) the condition was stated as ‘rare’. In July, September, December and February the condition was ‘common’. Only one month in November prevalence recorded as ‘occasional’. In rest of the months April, May and March they were not found. The probable reason for the high prevalence of monogeneans in winter may be the fact that monogeneans have a direct life cycle, i.e. without involvement of any intermediate hosts. So, the fall in temperature might be the reason for prevalence of monogeneans. Higher temperature seems to be un favourable for these parasites, as they were found to occur less during summer and monsoon than in winter which corroborates with the findings of Ghosh *et al.,* [10] and Kim *et al.,* [13]. The result of the present study corroborates with that of Ghosh *et al.,* [10] who observed highest infestation of *Dactylogyrus* sp. in *Catla catla* during December to February.

PFI of Nematodes varied from 5% to 22.22% throughout the year. In August, they were recorded highest (PFI: 22.22%) followed by a decrease up to January (PFI, 5%). In the months of July, August and March condition were ‘occasional’. February, May, June, September, October and November they were not found. The results of the present study satisfied all the criteria as reported by Dash *et al.,* [7]. Month of August was observed as most suitable for proliferation of nematode parasites in *Catla catla.* The results of present study showed that nematodes occurrence were more in August than other months. The probable reasons behind this might be due to the environmental conditions which were conducive to these parasites, presence of first intermediate hosts (copepod) and host specificity of parasites. The present study were similar with the works of Kim *et al.*, [13], who had reported that seasonal distribution of nematodes may be related to the fluctuation in temperature, presence of intermediate hosts and feeding habits of the hosts.

PFI of *Argulus* sp. were only found in the months of October and March (PFI, 4.76% and 4.545%), and in rest of the months they were not found. PFI of developmental stages of eggs/ parasites varied from 4.76% to 23.07%. They were highest during February (PFI, 23.07%) which was stated as 'occasional' and lowest in the month of February (PFI, 4.76%) considered as 'rare'. These were found only in September, October, January and February and in rest of the months they were not found. *Argulus*is an ectoparasite, generally found in the skin and gill portion of the fish body. Large individuals preferred the skin behind the base of the pelvic and pectoral fins and lesser extends in adipose fins. Mouth, base of the dorsal fin and ventral fins of the hosts were found to be the best suited place for the attachment of the parasite. The present study corroborated with the findings of earlier authors Chowdhury [6], Shella *et al.*, [18]. and Samir [17] as these were found to attack mainly on external organs. Ectoparasites were not host specific and can live on variety of host species and have good potential to spread.

PFI of *Chilodonella* sp., *Learnea* sp. and unidentified crustaceans were only found in the month of February (PFI, 7.60%, 7.69% and 15.38% respectively) condition was 'rare and occasional' respectively. In rest of the months they were not found. Probable reason might be the abundance of intermediate hosts and sudden fluctuation of water quality parameters, these results strongly supported by Dash *et al.*, [7]. and Kim *et al.*, [13]..

Statistical analysis revealed that there was no significant difference ($P<0.05$, df=11) in PFI values among all the months (Table 2). However, there was significance difference ($P>0.05$, df=10) in PFI values among the parasites.

Table 1: Monthly distribution of parasites and severity of infection in Catla catla

Months	Total number of fishes examined	Name of the parasites	No. of Infected fishes	PFI (%)	Site of infection	Severity of infection
April	24	*Myxobolus* sp.	5	20.83^{b}	Gill	0.5
		Trichodina sp.	15	62.50^{c}	Gill	1
		Nematodes	2	8.33^{a}	Intestine	0.5
May	25	*Trichodina* sp.	11	44.00^{c}	Gill	0.5
June	25	*Myxobolus* sp.	10	40.00^{c}	Gill	2
		Trichodina sp.	13	52.00^{c}	Gill	0.5
		Dactylogyrus sp.	1	4.00^{a}	Gill	0.5
July	30	*Myxobolus* sp.	5	16.66^{b}	Gill	1
		Thelohanellus sp.	3	10.00^{b}	Gill	0.5
		Dactylogyrus sp.	14	46.66^{c}	Gill	3
		Gyrodactylus sp.	4	13.33^{b}	Fin	0.5
		Nematodes	5	16.66^{b}	Intestine	Nematodes
August	27	*Myxobolus* sp.	5	18.51^{b}	Gill	2
		Thelohanellus sp.	2	7.40^{a}	Gill	0.5
		Trichodina sp.	19	70.37^{d}	Gill	0.5
		Dactylogyrus sp.	24	8.88^{a}	Gill	3
		Gyrodactylus sp.	10	37.03^{c}	Gill	0.5
		Nematodes	6	22.22^{b}	Intestine	0.5
September	21	*Myxobolus* sp.	13	61.90^{c}	Gill	2
		Trichodina sp.	6	28.57^{b}	Gill	0.5
		Dactylogyrus sp.	10	47.61^{c}	Gill	1
		Gyrodactylus sp.	2	9.52^{a}	Gill	0.5
		Development stage	2	9.52^{a}	Intestine	0.5
October	21	*Myxobolus* sp.	5	23.80^{b}	Gill	0.5
		Thelohanellus sp.	1	4.76^{a}	Gill	1
		Gyrodactylus sp.	4	19.40^{b}	Skin	0.5
		Dactylogyrus sp.	15	71.42^{d}	Gill	2
		Argulus sp.	1	4.76^{a}	Body	0.5
		Developmental stage	1	4.76^{a}	Gill	0.5
November	26	*Myxobolus* sp.	22	84.61^{d}	Gill	2
		Trichodina sp.	5	19.23^{b}	Gill	0.5
		Dactylogyrus sp.	3	11.53^{b}	Gill	0.5
Desember	23	*Myxobolus* sp.	20	86.95^{d}	Gill & Fin	1
		Thelohanellus sp.	8	34.78^{c}	Gill	0.5
		Trichodina sp.	2	8.69^{a}	Gill	0.5
		Gyrodactylus sp.	4	17.39^{b}	Skin	0.5
		Dactylogyrus sp.	15	65.21^{c}	Gill	1
January	20	*Myxobolus* sp.	19	95.00^{d}	Gill & Fin	1
		Thelohanellus sp.	4	20.00^{b}	Gill	0.5
		Gyrodactylus sp.	4	20.00^{b}	Skin	1
		Dactylogyrus sp.	18	90.00^{d}	Gill	1
		Nematodes	1	5.00^{a}	Intestine	0.5
		Digenean	1	5.00^{a}	Intestine	0.5
February	26	*Myxobolus* sp.	12	46.15^{c}	Gill	3
		Thelohanellus sp.	10	38.46^{c}	Gill	2
		Trichodina sp.	12	46.15^{c}	Gill	0.5
		Chilodenella sp.	2	7.69^{a}	Skin	3
		Gyrodactylus sp.	6	23.07^{b}	Body	0.5
		Dactylogyrus sp.	18	69.23^{c}	Gill	2

		Digenean	6	23.07^{b}	Intestine	0.5
		Learnea sp.	2	7.69^{a}	Body	0.5
		Crustaceans	4	15.38^{b}	Gill	0.5
March	22	*Myxobolus* sp.	19	86.36^{d}	Gill & Fins	2
		Thelohanellus sp.	9	40.90^{c}	Gill	2
		Trichodina sp.	10	45.45^{c}	Gill	1
		Argulus sp.	1	4.545^{a}	Body	0.5
		Nematodes	3	13.63^{b}	Intestine	0.5

PFI=Parasitic Frequency Index (%). a=rare (0.1 – 9.9%); b=occasional (10 – 29.9%); c = common (30 – 69.9%); d = abundant (70 – 100%);

Table 2: Two way ANOVA of PFI (%) values for Catla catla from April-2012 to March-2013.

Source of Variation	***SS***	***df***	***MS***	***F***	***P-value***	***F crit***
Parasites	37291.08	10	3729.108	11.8812	1.13E-13	1.917827
Months	5321.455	11	483.7687	1.541322	0.126932	1.876732
Error	34525.28	110	313.8662			
Total	77137.81	131				

4. Conclusion
The present study concluded that *Catla catla* was vulnerable to different parasites such as *Myxobolus* sp., *Thelohanellus* sp., *Trichodina* sp., *Gyrodactylus* sp., *Dactylogyrus* sp., Nematodes, *Argulus*sp *Lernea* sp., *Chilodonella* sp, unidentified crustaceans and dev

elopmental stages of parasites or eggs. Winter months (October to January) was the most susceptible period to get parasitic infestation on *Catla catla*. During this period the water quality get deteriorates and the fishes were in more stress in condition which favors the parasites to infest. Some parasites were found more during summer, this conditions favors their reproduction due to the availability of their intermediate hosts. In monsoon the temperature fluctuates which also favors the growth of some parasites. We also found that most of the parasites were found on gills and skin of the *Catla catla*. It is recommended to study the variation in intensity of infestation of parasites in fish with sex and tissue, with pollution state of the habitat and with the habitat type, health status of imported fish should be checked. Water quality should be maintained during winter season which can prevent parasite infestation to a greater extent. Establishment of quarantine system and development of specific pathogen resistant species for parasites both can prevent the entry parasites.

5. Acknowledgement
Authors thank to the Indian Council of Agricultural Research (ICAR), Government of India, and New Delhi for financial support under the Niche Area of Excellence mode.

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