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PARASITIC STUDY OF *Cirrhinus mrigala* (HAMILTON, 1822) IN SELECTED DISTRICTS OF WEST BENGAL, INDIA.

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ABSTRACT

The dissertation work was carried out to isolate and identify different parasites from Cirrhinus mrigala, severity of infestation and to find out Parasitic Frequency Index (PFI, %) months, seasons and lenghth groups wise from different selected districts of West Bengal. An investigation was made on Cirrhinus mrigala, such way that the selected districts were had more potential fishery resources, easy to collect samples from those districts and easy transportation of collected samples to laboratory. 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 major problem and the most prevalent disease causative agents among cultured fish spp. The isolated parasites were Myxobolus sp., Thelohanellus sp., Trichodina sp., Dactylogyrus sp., Gyrodactylus sp., Nematodes, Argulus sp., Lernea sp., Chilodonella sp., Development stage, Ichthyoptherius multifilus, Cosia sp. and unidentified Crustaceans. During study period Myxobolus sp. and Thelohanellus sp. prevalence were highest in winter and spring seasons (81% and 51% respectively). Trichodina sp. prevalence were more in summer i.e. 47%. During study period Dactylogyrus sp., Gyrodactylus sp. prevalence were highest in rainy season 65% and 6.9% respectively. Ichthyoptherius multifilus and Cosia sp. were recorded only in spring season. 1cm to 30cm length group fishes were more infested with the parasites compare to 30.5 cm to 40cm length groups.

Key words: Cirrhinus mrigala, Parasitic study, Severity of infestation, PFI (%), Seasons, Length groups, Selected districts, West Bengal.

I. INTRODUCTION

The aquaculture, in recent decades is considered to be one of the important factors of the world economy. Other than marketing concern, the biggest challenges that face the fish farmers is to control many biotic and abiotic factors, influencing fish rearing and aquaculture operations [1]. Freshwater aquaculture depends mainly on carp culture practices that account for around 80% of the total inland fish production and have proved sustainable at different levels of production over the years. Though the country possesses a large number of potential cultivable carp species, it is only the three Indian Major Carps *viz*., Catla (*Catla catla*), Rohu (*Labeo rohita*) and Mrigal (*Cirrhinus mrigala*), that contribute a lion's share with production of over two million tonnes. Production comes from over

2.25 million ha of tanks or ponds, 1.3 million ha of oxbow lakes, 3 million ha of reservoirs and 1.2 million ha of coastal brackish water area [2].

West Bengal being a 'rice-fish society', the State is highly significant historically, geographically and strategically since long past. The State has 37% of pond resources in India of which 70% are being 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. In other water bodies the utilization is much lower except for sewage fed fisheries [3]. The state of West Bengal has always attracted attention for being the highest producer of freshwater table fish and fish seed in the country together with the unique distinction of having the maximum water area under traditional shrimp farming. Everyday about 250 million litre of waste water along with solid waste is discharged here though different canals. Maximum Bheries of fish farmers using this canal water (Kolkata sewage water) for culture. Huge amounts of hazardous substances, heavy metal, sulphide, grease, oil originated from different industries of the surroundings, domestic waste water and industrial effluent from Kolkata city and pollute the aquatic environment. That can leads to stress on the fishes, under the stress condition fish can prone to so many parasitic diseases [4]. It is important to mention here that the parasitic infestations are reportedly playing a major role in disease occurrences (78%) in Indian freshwater aquaculture [5]. The results corroborate the observations of [6], who reported that ectoparasitic diseases are the main problem in freshwater fish farms of Andhra Pradesh, India which caused an annual loss of US$ 1 million due to disease-induced mortality and impaired growth. Further, the freshwater fish farmers of Andhra Pradesh, India [7] and West Bengal, India [8] were estimated to produce about 21% and 26% less, respectively than the expected production due to diseases, poor farm management practices and impaired growth. The normal growth is affected by parasite that lives on the fish if highly infested. Ectoparasites not only harm the fish directly but also render the fish for grown, reduce host population and induce mortalities [9,1]. [10] discussed that gill myxoboliosis was the most widely distributed disease infecting various species of carps in many states in India and also reported heavy mortality in Andhra Pradesh during November and December, 2000. In cultured fish population, the parasites may involve in the serious outbreak of disease [11]. They have been receiving considerable scientific attention due to serious damage to fishery resources by them [12]. In the high stocking conditions, particularly if the fishes are stressed, the parasites multiply rapidly. The temperature and slow water flow rate may also increase the parasitic infestation [13].The incidence and intensity of parasite also varied with season [14]. Young fishes are more prone to infection than older one [15]. The stoking density and water quality parameters correlate with the incidence of fish parasites [16]. In confined water bodies of Pakistan, the ectoparasites are one of great problem to fresh water carps [17]. In order to increase profitability, health care based on the knowledge of organisms, their ecology, and application of the knowledge in the control of diseases is essential [18,19]. Therefore, contribution to the knowledge of fish parasites is a pre-requisite for the rapid and correct diagnosis of the disease. Early diagnosis can lead to preventive measures which is the best way to reduce outbreak of disease [19-21]. Despite parasitic infestations, neither fish mortalities nor major disease outbreaks were recorded in the present study.

II. MATERIALS AND METHODS

The present study on the prevalence of parasitic infestation in freshwater carps was carried out for a period of 12 months between 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 concerned district. The samples were collected on a regular basis once in every month. In each sampling about 20-30 fishes were collected in live condition. The fishes were brought to the laboratory in live condition with water filled buckets and the total length, 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 [22]. External parasites from body surface, fin and gills were removed by scrapping the slime with a sharp scalpel it was mixed with a drop of physiological saline and was spread on a clean dry glass slide with coverslip on top of it. The gill arches were removed and macerated on slides and examined under a compound binocular and trinocular microscope. In case of monogeneans the gills were removed into petridishes containing physiological saline water and gently scrapped to dislodge monogeneans. The monogeneans were removed on to clear slides with a fine pipette in a drop of water and covered with cover slip. For endoparasites fishes were dissected out ventrally by a sharp scalpel to observe parasites inside buccal cavity stomach and intestine. The whole gut was removed in a watch glass containing 0.9% physiological saline and was cleaned several times with tap water to free it from any unwanted materials. Small worms were searched initially with the help of magnifying glass by scrapping out mucus. [23]. Phenotypic characterization of all protozoans, monogeneans, digeneans, and nematode parasites were studied as described by [24]. Photomicrographs were taken using a Motic BA400 phase contrast microscope with in-built 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 [4, 25]

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 [4, 26] was followed.

III. RESULTS AND DISCUSSION

3.1. Monthly prevalence of different parasites in in *Cirrhinus mrigala*

Monthly distribution of parasites in *Cirrhinus mrigala* is presented in **Table-2** and **Figure-1.** Parasitic Frequency Index (PFI) of *Myxobolus* sp. (**Figure-4, 5**) were highest in November (PFI, 92%) stated as ‘abundant’ and lowest in the month of July (PFI, 6.66%) stated as rare. They were ‘abundantly’ distributed in the months of August, November, December and January (PFI, 88%, 92%, 82.6% and 85.71% respectively). After February they decreased (PFI, 19.47%**)** and become ‘occasional’ in this month. A slight increase again appears in the month of March (PFI, 65%) and were stated as common. June, September, October and March they were found as ‘common’. They were found as ‘occasional’ in the months of April

and February (PFI, 12% and 19.47%). In May month they were not found. Present study supported by [55], during his study period at Rajshahi, Bangladesh, the highest number of parasites reported in December month and lowest in February. [27] recorded high prevalence of myxozoan parasites during August to January when the ambient temperature was 25ºC. These results were strongly supported by [28]. Present findings were quite similar with results of the above authors. Similar findings regarding incidence of diseases in fish during winter months showed by. [29, 30]. PFI of *Thelohanellus* sp. (**Figure-6**) reaches to the peak in the month of November (PFI, 84%) stated as 'abundant' and lowest in the month of June (PFI, 8%) rare. These parasites were found in the months of November, December, January and February stated as abundant, occasional, common and abundant respectively. *Thelohanellus* sp. not found in the months of April, May and July. As the water quality parameters fluctuate very quickly during winter and summer season, fish becomes affected with diseases in these two seasons. The parasitic infection is greatly influenced by the season, which basically interferes with ecology and physiology of the fish. During the breeding season of fish lesser number of parasites invade the host because of the presence of the oestrogen these were supported by [31]. During the study period *Trichodina* sp. (**Figure-7**) were found as 'common' in the months of April, May and June (PFI, 44%, 50% and 48%). and these were not found in the month of July. These parasites reaches to the peak in the month of December (PFI, 91.3%) stated as 'abundant' and lowest in the month of October (PFI, 19.04%) as 'occasional' and these were found throughout the year except July and November. It may due to high stocking density is being maintained during carp nursery operations, and this density induces bio-ecological stress to fry and make the fry more susceptible to the parasitic infection. It is evident from the available literature that parasitic diseases caused significant damage in nursery systems of carp, catfish and shellfish of Srilanka, Malaysia, Indonesia, Taiwan and India [32]. Various physicochemical factors such as water and atmospheric temperature, pH, hardness of water, dissolved oxygen, biological oxygen demand (BOD) have strong impacts on fish health and their resistance to attack by the causative agents. Present study strogly supported by [33, 34]. *Gyrodactylus* sp. (**Figure-13**) were found only in the month of August (PFI, 20%) as 'occasional', rest of the months were not observed.

During the study period *Dactylogyrus* sp. (**Figure-12**) were not found in the summer months i.e. April, May and June. These were reached peak condition in the month of January (PFI, 80.95%) as 'abundant' and lowest in the month of March (PFI, 15%) stated as occasional. These were observed 'common' in condition, in the months July and February during the study period and abundant in the months of August, September and January (PFI, 80%, 70% and 80.95**%**). October, November and March these parasites found occasionally (PFI, 16%, 15% and 28.57%). *Gyrodactylus* sp. were found only in August (PFI, 20%), 'occasinal'. The results of this study are similar with that the results of [35, 36].

Some found that *Dactylogyrus vastator* as a common parasite of carps and the infection rates were higher during summer [35]. In general, the abundance of *Gyrodactylus salaris* to increase during the summer and only early autumn and to decrease during the winter and early spring studied the life cycle of *Dactylogyrus vastator*. During May and June the life span at 24-28^{o} C was estimated to be 11-13 days. The suitable temperature for development of life cycle and reproduction of ectoparasites is called optimum temperature [13] which was agreement with the present findings. Adult worms lived only 5 days. Thus in summer (110-130 days) there is potential for

ten parasite generations to be completed in carp ponds. That is why intensity of infection was higher in the summer. [36] reported that prevalence of *Gyrodactylus crysoleucas* increased in late spring and summer. Nematodes (*Capillaria* sp.) (Plate-2, Figure-12) were found only in the months of April, June, July, December and January (PFI, 12%, 36%, 23.33%, 8.69% and 4.76%)respectively. In rest of the months not found. PFI (%) of Nematodes reached peak stage in the month of June (PFI, 36%), lowest in the month of August (PFI, 4.76%). The results of the present study satisfied all the criteria as reported by [37] who opinioned the month of August was observed as most suitable for proliferation of nematode parasites in *Catla catla* and *Cirrihinus mrigala. Argulus* sp. (Plate-2, Figure-8) found only in the months of October and March (PFI, 4.76% and 5%) stated as 'rare and occasionsl', rest of the months not found, present study supported by [38], who reported on *Argulus* sp. in IMC.*Lernea* sp. were not found during the study period. *Chilodonella* sp. (**Figure-9**) were found only in the month of September (PFI, 10%), rest of the months absent. [39] reported *Chilodonella* glides over the fish's gill and skin surfaces which supported the present findings. *Chilodonella* sp. live on the skin and gills of fish. Two species of *Chilodonella* occur on freshwater fishes, *Chilodonella cyprini* occurring on the skin and gills of carp *Cyprinus carpio* (L) and *C. hexasticha* on the skin and gills of tench (*Tinca tinca*) which also corroborated the present observation. *Chilodonella hexasticha* from the gills of tropical ornamental *Symphysodon discus*, cichlids (*Oreochromis mossambicus, Oreochromis niloticus, Oreochromis aureus*), and coldwater cyprinids (*Abramis brama, Abramis ballerus, Blicca bjoerkna, Cyprinus carpio*).

During the study period *Vorticella* sp. (**Figure-8**) and Crustaceans were found in the month of July (PFI, 16.66% and 20%), stated as 'occasional', rest of the months absent. A great number of Vorticellids on the skin of debilited, moribund fish and prey on the body surface of the fishes and feed on the tissues, which supports the present findings. Ectoparasitic protozoa attack the fish and cause massive destruction of the skin and gill epithelium [40, 41] which also approved the present findings. Intestinal flukes were recorded only in the month of July (PFI, 12%) stated as 'occasional', rest of the months absent. Developmental stages were reached peak stage in the month of November (PFI, 24%), lowest in the month of June (PFI, 4%) stated as 'occasional' and 'rare' respectively. During the study period *Ichthyoptherius multifilus* (**Figure-10**) were found only in the month of February (PFI, 9.52%) stated as 'rare' but digeneans were also found 'rare' in the months of January, February (PFI, 9.52%) and *Costia* sp. were recorded only in the month of March (PFI, 5%) stated as 'rare', rest of the months not found. The ciliated protozoan, *Ichthyophthirius multifiliis*, the causative agent of ichthyophthiriasis or Ich, is one of the most important pathogenic parasites of cultured fish present study strongly supported by [42]. The PFI of digeneans (Plate-2, Figure-11) were more in the intestine compared to gills and stomach of the fishes. This observation derives support from that of [43] who observed maximum incidence of *Opecoelus sphaericus* in intestine of greenlings (*Hexagrammos otakii*). Higher incidence of digeneans in intestine may be attributed to the endoparasitic nature of this group. [44] reported digeneans from fish muscle, body cavity, mesenteries, liver and swimbladder. Easy availability of nutrients also might contribute to aggregation of the parasites in the intestine.

Statistical analysis (**Table-1**) revealed that there was no significant difference ($P>0.05$, $df=11$) in monthly PFI values. However there was significant difference ($P<0.05$,$df=15$) in PFI values among the parasites. Similarly there was significant difference ($P<0.05$,$df=15$) in PFI values among the *Myxobolus* sp.,

Trichodina sp., *Dactylogyrus* sp. and *Thelohanellus* sp., however there was significant difference (P<0.05,df=15) in PFI values among the *Myxobolus* sp., Nematodes, Developmental stage, *Gyrodactylus* sp., Crustaceans, *Argulus* sp., Digeneans, *Vorticella* sp., Intestinal flukes, *Chilodonella* sp. and *Ichthyophthirius multifilis.* However there was no significant difference (P>0.05, df=11) in PFI values all parasites except *Myxobolus* sp.

3.2. Occurrence of parasites in different seasons

Occurrence of parasites in different seasons is given in **Table-4** and Figure-2. The influence of parasites in relation to the seasons has been described by many workers [14, 28] who worked on seasonal variations. The total study period was divided into four seasons; i.e. Summer (April - June); rainy season (July-September) or Monsoon Winter (October-January) and Spring (February- March).

The occurrence *Myxobolus* sp. were found in all seasons, but it reached peak during the winter season (PFI, 81%) as 'abundant', low in summer (PFI, 22%) which was stated as 'occasional'. They were also 'common' in rainy season and most abundant during winter and spring season. These results were similar with the works of [28], in *L. rohita*. Similar findings were showed by [29, 30] also reported more incidence of diseases in fish during winter months. *Thelohanellus* sp. presence were constantly increased from summer to spring season (PFI, 2.7% to 51%), these parasites reached peak condition in spring (PFI: 51%) as 'common', lowest in summer (PFI, 2.7%) which stated as 'rare'. These parasites 'commonly' found in winter and spring seasons (PFI, 38% and 51% respectively). The occurrence of *Trichodina* sp. 'occasional' in rainy season and spring season (PFI, 12% and 12%), which was common in summer and winter (PFI, 47% and 44% respectively). The probable reason for the higher prevalence of *Trichodina sp* in fishes in winter may be that they have a direct life cycle, i.e. without involvement of any intermediate hosts. So the temperature may be the main factor of their prevalence. On the other hand in winter the fishes become stressed which might also be the reason for their highest prevalence as it was reported that Trichodinds are opportunistic parasites which become pathogenic under stress full conditions [45]. These parasites were found high in summer season (PFI, 47%). The reason may also be the overcrowding of fish in ponds and deteriorating water quality due to sudden low temperature as suggested that the intensification of fish culture creates disease problems that originate from overcrowdings. Deteriorating water quality such as unsuitable water temperature is a reason for trichodiniasis. The trichodiniasis is caused by *Trichodina* sp.; the infection being stimulated by the high density of fish in ponds.

Prevalence of *Dactylogyrus* sp. decreased from rainy season to spring season (PFI, 65% to 31%) which were common to occasional. These parasites were not found in summer season (PFI, 0%). *Gyrodactylus* sp. were 'rare' in rainy season (PFI, 6.6%). These parasites were not found in rest of the seasons probable reason may be higher temperature was ambient for the proliferation of Monogeneans, these results supported by [37]. The cestodes attain their sexual maturity in summer. So their presence was observed in summer season only this were also strongly supported by [37]. Occurrence of nematodes were gradually decreased from summer to winter season (PFI, 16% to 1.1% respectively) and these parasites were not found in spring season. Nematodes were rarely found in rainy and winter season, occasionally found in summer season. During the study period *Argulus* sp. were 'rare' in winter and spring seasons (PFI, 1.1%, 7.3% respectively). Rest of the seasons these were not found, these findings were corroborated with the worlyk of other authors [46, 47]. *Chilodonella* sp. and *Vorticella* sp. were found 'rarely' in rainy season (PFI, 2.6%), rest of the season these were not found. Low water temperatures being

more optimal for reproduction of *Chilodonella piscicola* and massive infections with *Chilodonella sp* occur in low (12–17°C) ambient temperatures in South Africa and Israel which also supports the present findings and also corbborated with results of [48], who studied on exotic carps. Epizootics caused by *C. hexasticha* in Australia occurred during the winter months and the optimum temperature for *C. cyprini* is 5-10°C. The occurrence of intestinal flukes and Crustaceans were rare in summer and rainy season respectively. The occurrence of developmental stages of eggs/parasites were highest in spring (12.19%), lowest in summer (PFI, 1.38%), which were 'occasional' and 'rare' respectively. These parasites were also rare in winter (PFI, 7.7%) and not found in rainy season. Low temperatures being more congineal for reproduction of *Chilodonella piscicola* and some trichodinids. The probable reason for the availability of developmental stages more in winter may be due to the delaying of developmental process at low temperature as suggested, at a temperature of 15–17°C, the process of division of developmental stages of *Ichthyothirius sp* is lengthened. The temperatures below 15°C will delay development of asian tapeworm to 6–8 months. The low temperatures seem to delay or even interrupt development and consequently completion of the life cycle of tapeworms. At 28–30°C, 77% of the eggs hatched in the first day after release, the remainder during the following five days, where as at 14–15°C, the incubation period extended to 10–28 days and for all practical purposes interrupted below 12°C which may also be the reason for the availability of developmental stages of eggs more in winter.

The occurrence of digeneans (**Figure-14**) gradually increased from winter to spring (PFI, 2.2% to 4.2%). The observations of the present study are in agreement with that of [43] who reported lowest prevalence of digeneans in greenlings (*Hexagrammos otakii*) during May - July and highest prevalence in October. The reason for higher occurrence of digeneans in winter may be the optimum temperature for their growth lies in lower range which prevails in the winter season. Higher occurrence of *Aprocotyle simplex* in December 6ºC. Another alternative reason may be availability of intermediate hosts (snails) to complete their life cycle and seasonal fluctuation in prevalence of intermediate hosts may determine occurrence of digeneans in fish. *Costia* sp. and *Ichthyoptherius multifilus* were found in only spring season (PFI, 2.43% and 4.87% respectively). These were not found in rest of the seasons. Various physicochemical factors such as water and atmospheric temperature, pH, hardness of water, dissolved oxygen, biological oxygen demand (BOD) have strong impacts on fish health and their resistance to attack by the causative agents [33, 34].

Statistical analysis (**Table-3**) showed that there was no significant difference ($P>0.05$, df=3) in PFI values among the seasons. However, there was significant difference ($P<0.05$, df=14) among the parasites. There was no significant difference ($P>0.05$, df=14) in PFI values among *Myxobolus* sp., *Trichodina* sp., *Dactylogyrus* sp., *Thelohanellus* sp. However, the PFI values differ significantly ($P<0.05$, df=14) between *Myxobolus* sp., *Trichodina* sp., there was significant difference between *Myxobolus* sp. and rest of the parasites analyzed. Similarly there was no significant difference among *Trichodina* sp., *Dactylogyrus* sp., *Thelohanellus* sp., Nematodes, and Development stages of parasites.

However, the PFI values differ significantly ($P<0.05$, df=14) in *Trichodina* sp. and rest of the parasites. Except Myxobolus sp. and Trichodina sp. there was no significant difference ($P>0.05$, df=14) in PFI values among the other parasites.

3.3. Prevalence of parasites in different length groups of *Cirrhinus mrigala*

The influence of parasites in relation to the length of fishes has been described by many

workers [49-52]. The size of the fishes may affect prevalence and the diversity of parasitic fauna, so the Indian Major Carps examined were divided into different length groups and the results were noted and discussed made accordingly.

The distribution of different parasites in different length groups of *Cirrhinus mrigala* is represented in **Table-6** and Figure-3. The fishes were grouped into 1-10 to 40.5-50 cm length groups.

During the study period occurrences of *Myxobolus* sp. were 'common' in 1cm to 30 cm length groups, not found in rest of the length groups. *Myxobolus* sp. were reached peak stage in 10.5 to 20 cm length group, lowest found in 20.5 to 30 cm length group. Occurrences of *Thelohanellus* sp. were 'common' (PFI, 35.2%) in 10.5 to 20 cm length group, 'occasional' in 1-10 and 20.5 to 30 cm length groups. *Trichodina* sp. were found more in 1-10 cm length group (PFI, 60.7%), which was 'common' and lowest in 20.5-30 cm (PFI, 17.4%) as 'occasional'. *Gyrodactylus* sp. were found 'rare' from 1-30 cm length groups, not found in rest of the length groups. Occurrences of *Dactylogyrus* sp. were 'occasional' in 10.5-20 and 20.5-30 cm length groups and 'rarely' found in 1-10 cm length group. The results revealed that the prevalence of monogenean were more in the large size groups of the hosts than the smaller size groups. [53] provided evidence of a positive correlation between host size and monogenean parasitization level. [43] also showed a weak positive correlation between intensity of monogeneans and body length of greenlings. A large gill surface and an increased volume of water passing over the gills in larger fish might increase the probability of contact with parasite oncomiracidia and resulted in a higher infection level. Nematodes (**Figure-15**) and Development stages of parasites were found 'rarely' (PFI, 1.7-8.8%) in 1-20 cm length groups and occasional in 20.5-30 cm length groups. This result showed that the nematodes infection were more in smaller group than the larger size group of fishes. The changes in food habits in different stage of the hosts might be the reason for fluctuation in the infestation by the nematodes in different groups of fishes. [54] also found a similar result. He reported the abundance of *Hysterothylacium aduncum* in the little trout is higher than that of bigger ones. Occurrences of *Argulus* sp. were found in 1-10 and 20.5-30 cm length groups, which was stated as 'rare'. Prevalence of *Chilodonella* sp., *Ichthyophthirius multifiliis*, Digenian parasites, Intestinal flukes and Ciliophoran parasites were 'rare' in 10.5 to 20 cm length group. These parasites not found in rest of the length groups. Crustacean's parasites and *Vorticella* sp. were only found in 20.5 to 30 cm length groups, these were not found in rest of the length groups but the condition was 'occasional' and 'rare' respectively. [49] has observed varying results in the parasitic abundance in different length groups of fish, which he attributed to the changes in the feeding at different ages of the host. From the aforesaid observations we can say that the prevalent of parasite in different length groups were varying may be due to the changes in the feeding habit of fishes.

Statistical analysis (**Table-5**) revealed that there was significant differences ($P<0.05$, df=4) in PFI values among the all length groups and ($P<0.05$, df=15) all parasites. However *Myxobolus* sp. differ significantly ($P<0.05$, df=15) in PFI values to all other parasites. Similarly there was significant difference ($P<0.05$, df=15) in in PFI values among the *Myxobolus* sp., *Trichodina* sp., *Dactylogyrus* sp., *Thelohanellus* sp., Nematodes, Development stages of parasites, *Costia* sp., Crustaceans, *Gyrodactylus* sp. and *Argulus* sp., however there was no significant difference ($P>0.05$, df=15) in all parasites except *Myxobolus* sp.

IV. CONCLUSION

The present study also brings about the conclusion that the *Cirrhinus mrigala* were 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 developmental stages of parasites or eggs. Winter was the most vulnerable period to get parasitic infestation. During this period the water quality get deteriorates and the fishes were in stressed condition which favours the parasites to infest. Some parasites were found more during summer which favours their reproduction due to the availability of their intermediate hosts. In monsoon the temperature fluctuates which also favours growth of some parasites. These parasites were found mostly on gills and skin of the fishes. The small and medium size fishes were found to be more vulnerable due to their poor immunity power and wide spread surface area respectively, which favours more colonization of parasites.

Avoid overcrowding in the pond to maintain proper health of the fishes. Water quality should be maintained during winter which can prevent parasite infestation to a greater extent. Establishment of strong quarantine system can prevent the entry of exotic parasites. To improve the fish's own resistance during the winter a good feed to be fed to increase the immunity of the fishes. If parasites infestation occurs then some antiparasitic drugs can used such copper sulphate, ferrous sulphate, iodine, potassium permanganate to eradicate the parasites.

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Table-1. Two way ANOVA of PFI values for *Cirrhinus mrigala* from April-2012 to March-2013.

Source of Variation	*SS*	*df*	*MS*	*F*	*P-value*	*F crit*
Parasites	49503.47	15	3300.232	12.55947	1.89E-20	1.727388
Months	3441.585	11	312.8714	1.190674	0.297074	1.847078
Error	43356.77	165	262.7683			
Total	96301.83	191				

Table -2. Prevalence (PFI %) and Severity of infection of parasites in *Cirrhinus mrigala* of West Bengal during the period between April 2012 and March 2013.

Month	No. of Fish investigated	Parasitic data				
		Parasites present	No. of Infected fishes	PFI (%)	Site of infection	Severity of infection
April	25	*Myxobolus* sp.	3	12.00	Gill	0.5
		Trichodina sp.	11	44.00	Gill	0.5
		Intestinal flukes	3	12.00	Intestine	0.5
		Nematodes	3	12.00	Intestine	0.5
May	22	*Trichodina* sp.	11	50.00	Gill	0.5
June	25	*Myxobolus* sp.	13	52.00	Gill	1
		Thelohanellus sp.	2	8.00	Gill	0.5
		Trichodina sp.	12	48.00	Gill	0.5
		Nematodes	9	36.00	Gill	1
		Developmental stage	1	4.00	Gill	0.5
July	30	*Myxobolus* sp.	2	6.66	Gill	0.5
		Vorticella sp.	5	16.66	Gill	0.5
		Dactylogyrus sp.	15	50.00	Gill	2
		Nematode	7	23.33	Gill	1
		Crustaceans	6	20.00	Body	0.5
August	25	*Myxobolus* sp.	22	88.00	Gill	2
		Thelohanellus sp.	9	36.00	Gill	1
		Trichodina sp.	5	20.00	Gill	0.5
		Dactylogyrus sp.	20	80.00	Gill	3
		Gyrodactylus sp.	5	20.00	Fin	0.5
September	20	*Myxobolus* sp.	12	60.00	Gill	0.5
		Thelohanellus sp.	2	10	Gill	0.5
		Trichodina sp.	15	75.00	Gill	1
		Chilodonella sp.	2	10	Gill	0.5

		Dactylogyrus sp.	14	70.00	Gill	2
October	21	*Myxobolus* sp.	13	61.90	Gill	0.5
		Trichodina sp.	4	19.04	Gill	0.5
		Dactylogyrus sp.	6	28.57	Gill	0.5
		Argulus sp.	1	4.76	Body	0.5
November	25	*Myxobolus* sp.	23	92.00	Gill	2
		Thelohanellus sp.	21	84.00	Gill	2
		Trichodina sp.	3	12.00	Gill	0.5
		Dactylogyrus sp.	4	16.00	Gill	1
		Developmental stage	6	24.00	Gill	1
December	23	*Myxobolus* sp.	19	82.60	Gill	1
		Thelohanellus sp.	6	26.08	Gill	0.5
		Trichodina sp.	21	91.30	Gill	0.5
		Nematodes	2	8.69	Gill	0.5
		Developmental stage	1	4.34	Gill	0.5
January	21	*Myxobolus* sp.	18	85.71	Gill	2
		Thelohanellus sp.	8	38.09	Gill & Fin	1
		Trichodina sp.	12	57.14	Gill	1
		Dactylogyrus sp.	17	80.95	Gill	0.5
		Nematode	1	4.76	Intestine	0.5
		Digenean	2	9.52	Intestine	0.5
February	21	*Myxobolus* sp.	19	19.47	Gill	3
		Thelohanellus sp.	17	80.95	Gill	3
		Trichodina sp.	5	23.80	Gill	0.5
		Ichthyoptherius multifilus	2	9.52	Body	1
		Dactylogyrus sp.`	10	47.61	Gill	1
		Digenean	2	9.52	Intestine	0.5
		Developmental stage	5	23.80	Gill	0.5
March	20	*Myxobolus* sp.	13	65	Gill	1
		Thelohanellus sp.	4	20	Gill	1
		Trichodina sp.	6	30	Gill	1
		Argulus sp.	3	15	Body	1
		Dactylogyrus sp.	3	15	Gill	0.5
		Costia sp.	1	5	Body	0.5

Table -3. Two way ANOVA of PFI values for *Cirrhinus mrigala* from April-2012 to March-2013 in different seasons.

Source of Variation	*SS*	*df*	*MS*	*F*	*P-value*	*F crit*
Seasons	645.7793	3	215.2598	1.482761	0.233011	2.827049
Parasites	17532.31	14	1252.308	8.626197	2.41E-08	1.935009
Error	6097.349	42	145.175			
Total	24275.44	59				

Table- 5. Two way ANOVA of PFI values for *Cirrhinus mrigala* from April-2012 to March-2013 in different length groups.

Source of Variation	*SS*	*df*	*MS*	*F*	*P-value*	*F crit*
Length groups	2207.282	4	551.8205	4.969807	0.001582	2.525215
Parasites	6399.692	15	426.6461	3.842461	9.4E-05	1.836437
Error	6662.076	60	111.0346			
Total	15269.05	79				

Table- 4. Prevalence (PFI, %) of parasites in *Cirrhinus mrigala* in different seasons from April 2012 to March 2013.

Cirrhinus mrigala		*Myxobolus* sp.		*Thelohanellus* sp.		*Trichodina* sp.		*Dactylogyrus* sp.		*Gyrodactylus* sp.		Nematodes		*Argulus* sp.	
Period	Total no of fish examined	No. of infected fish	PFI (%)	No. of infected fish	PFI (%)	No. of infected fish	PFI (%)	No. of infected fish	PFI (%)	No. of infected fish	PFI (%)	No. of infected fish	PFI (%)	No. of infected fish	PFI (%)
Summer (April - June)	72	16	22[b]	2	2.7[a]	34	47[c]	0	0	0	0	12	16[b]	0	0
Rainy season (July-September) or Monsoon	75	36	48[c]	11	14[b]	20	26[b]	49	65[c]	5	6.6[a]	7	9.3[a]	0	0
Winter (October-January)	90	73	81[d]	35	38[c]	40	44[c]	29	32[c]	0	0	1	1.1[a]	1	1.1[a]
Spring (February- March)	41	32	78[d]	21	51[c]	11	26[b]	13	31[c]	0	0	0	0	3	7.3[a]

Conti…

Chilodonella sp.		*Vorticella* sp.		Intestinal flukes		Crustaceans		Development stage		Digeneans		*Costia* sp.		*Ichthyoptherius multifilus*	
No. of infected fish	PFI (%)	No. of infected fish	PFI (%)	No. of infected fish	PFI (%)	No. of infected fish	PFI (%)	No. of infected fish	PFI (%)	No. of infected fish	PFI (%)	No. of infected fish	PFI (%)	No. of infected fish	PFI (%)
0	0	0	0	3	4.16[a]	0	0	1	1.38[a]	0	0	0	0	0	0
2	2.6[a]	2	2.66[a]	0	0	6	8[a]	0	0	0	0	0	0	0	0
0	0	0	0	0	0	0	0	7	7.77[a]	2	2.22[a]	0	0	0	0
0	0	0	0	0	0	0	0	5	12.19[b]	2	4.87[a]	1	2.43[a]	2	4.878[a]

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 -6. Prevalence (PFI, %) of parasites in different length groups of *Cirrhinus mrigala* from April 2012 to March 2013.

Parasites		*Myxobolus* sp.		*Thelohanellus* sp.		*Trichodina* sp.		*Gyrodactylus* sp.		*Dactylogyrus* sp.		Nematodes		*Argulus* sp.	
Length (cm)	Total no of fish examined	No. of infected fish	PFI (%)	No. of infected fish	PFI (%)	No. of infected fish	PFI (%)	No. of infected fish	PFI (%)	No. of infected fish	PFI (%)	No. of infected fish	PFI (%)	No. of infected fish	PFI (%)
1 to 10	56	25	44.6[c]	9	16.0[b]	34	60.7[c]	1	1.7[a]	4	7.1[a]	2	3.5[a]	1	1.7[a]
10.5 to 20	159	106	66.6[c]	56	35.2[c]	41	25.7[b]	5	3.1[a]	54	33.9[b]	14	8.8[a]	0	0
20.5 to 30	63	26	41.2[c]	16	25.3[b]	11	17.4[b]	2	3.1[a]	25	39.6[b]	7	11.1[b]	2	3.1[a]
30.5 to 40	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0
40.5 to 50	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0

Cont…

Development stage		*Chilodonella* sp.		Crustaceans		*Ichthyophthirius multifiliis*		Digenian parasites		*Costia* sp.		*Vorticella* sp.		Intestinal flukes		Ciliophoran parasites	
No. of infected fish	PFI (%)	No. of infected fish	PFI (%)	No. of infected fish	PFI (%)	No. of infected fish	PFI (%)	No. of infected fish	PFI (%)	No. of infected fish	PFI (%)	No. of infected fish	PFI (%)	No. of infected fish	PFI (%)	No. of infected fish	PFI (%)
1	1.7[a]	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0
11	6.9[a]	2	1.2[a]	0	0	3	1.8[a]	2	1.2[a]	1	0.6[a]	0	0	3	1.8[a]	2	1.2[a]
8	12.6[b]	0	0	7	11.1[b]	0	0	0	0	7	11.1[b]	1	1.5[a]	0	0	0	0
0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0
0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0

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

Figure-1. Prevalence of different parasites (PFI, %) in Mrigala (*Cirrhinus mrigala*) from April 2012 to March 2013

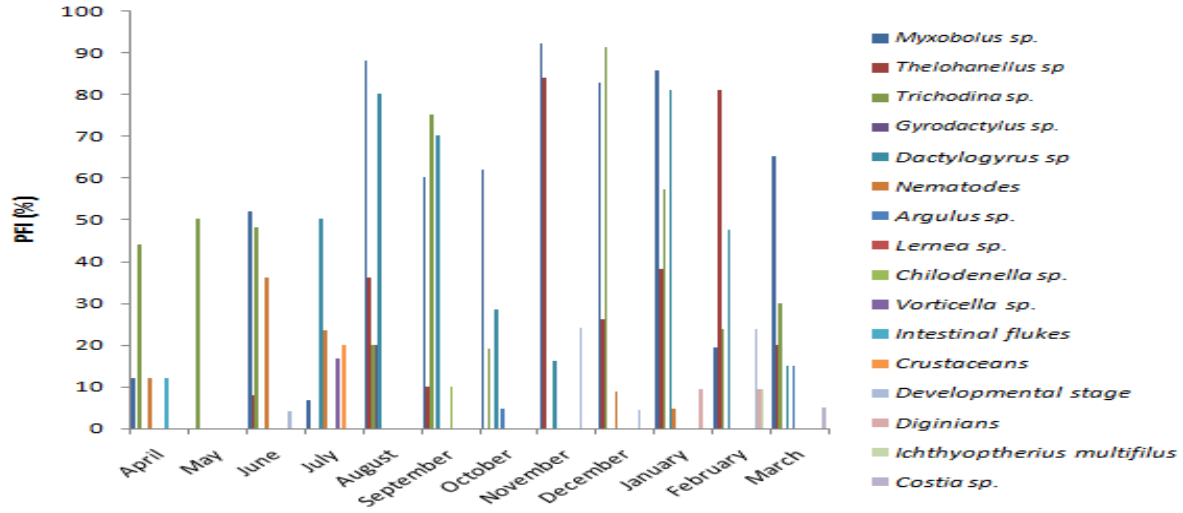


Figure-2. Prevalence of parasites (PFI, %) in Mrigala (*Cirrhinus mrigala*) in different seasons from April 2012 to march 2013

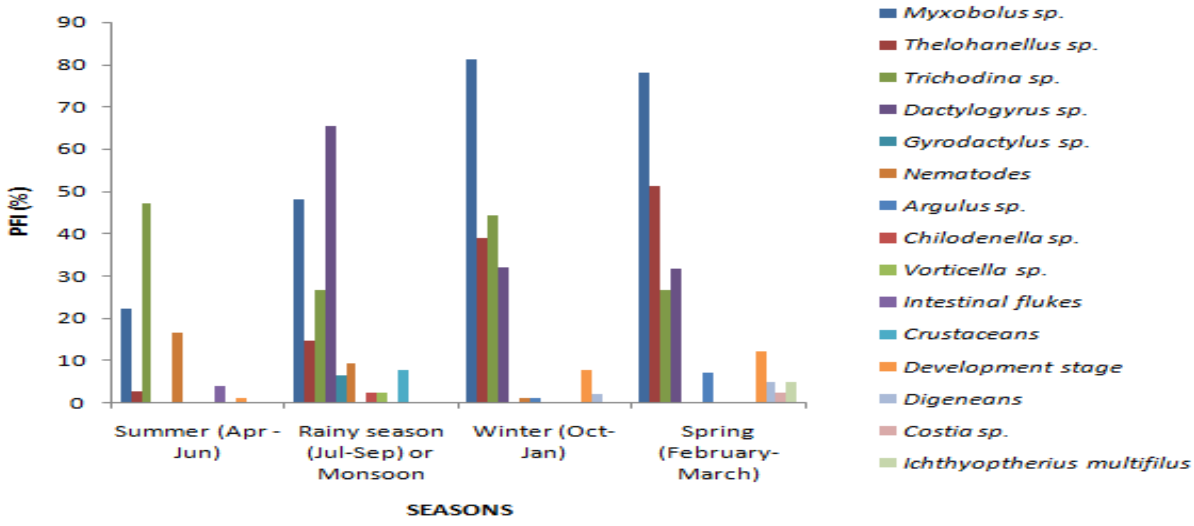
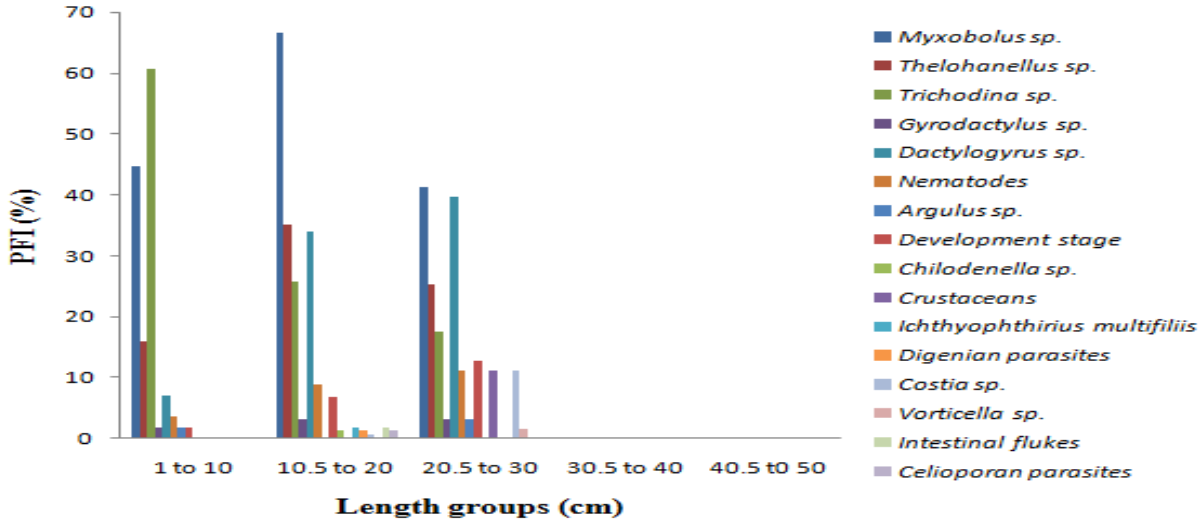


Figure-3. Prevalence of parasites (PFI, %) in different length groups of Cirrhinus mrigala from April 2012 to March 2013



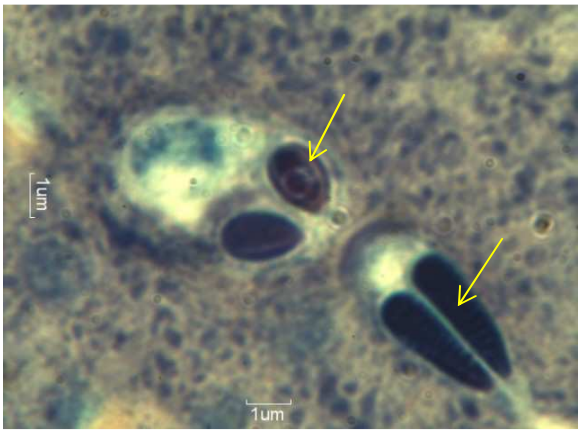


Figure-4. Spore of *Myxobolus* sp. with distinct polar capsules (arrows) from *Cirrhinus mrigala* (Giemsa stained, 1000x)

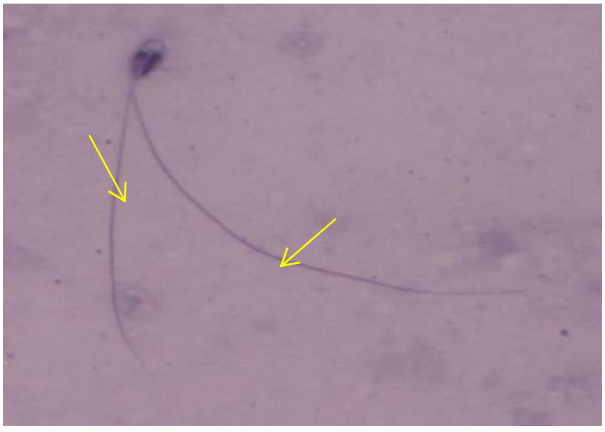


Figure-5.*Myxobolus* sp. with two distinct polar filaments (arrow) attached to the gills of *Cirrhinus mrigala* (Giemsa stained, 200x)

Figure-6.*Thelohanellus* sp. with distinct single polar filament (arrow) present on the gills of *Cirrhinus mrigala* (Giemsa stained, 200x)

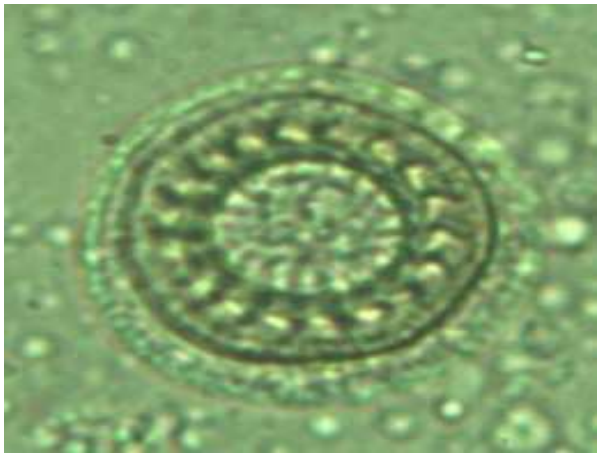


Figure-7. *Trichodina* sp. attached to the skin of *Cirrhinus mrigala* (Wet mount,200x).

Figure-8. *Vorticella* sp. with lengthy stalk (arrow) attached to the skin of *Cirrhinus mrigala* (Wet mount, 200x).

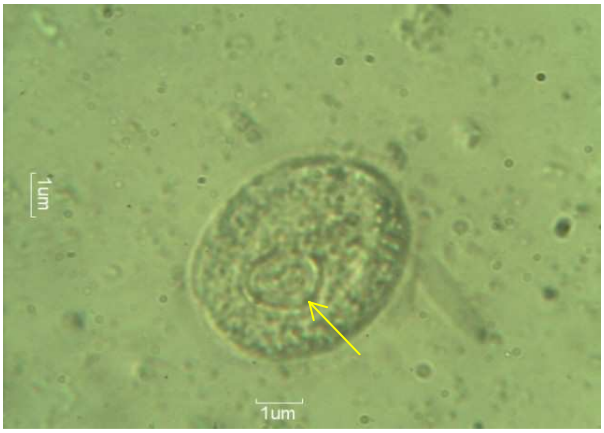


Figure-9. *Chilodenella* sp. with distinct nucleus (arrow) present in the gills of *Cirrhinus mrigala* (Wet mount, 200x).

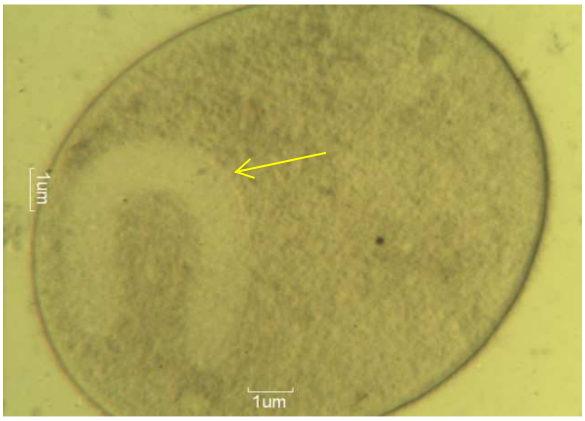


Figure-10. *Ichthyoptherius multifilus* with distinct nucleus (arrow) present in the gills of *Cirrhinus mrigala* (Wet mount, 200x).

Figure-11. *Argulus* sp. with clear egg pouch (arrow) attached to the dorsal fin of *Cirrhinus mrigala* ((Wet mount, 40x).

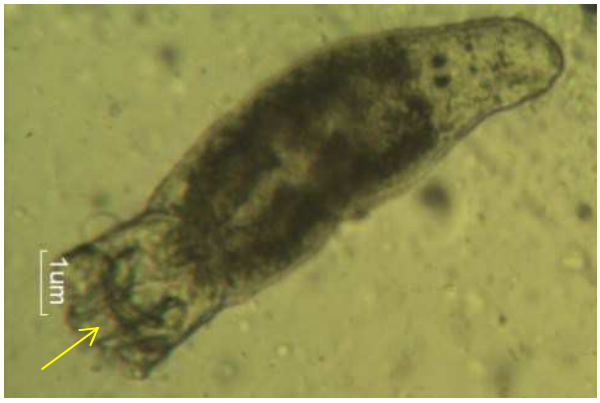


Figure-12. *Dactylogyrus* sp. with distinct lobes (arrow) present in the gills of *Cirrhinus mrigala* (Wet mount, 200x).

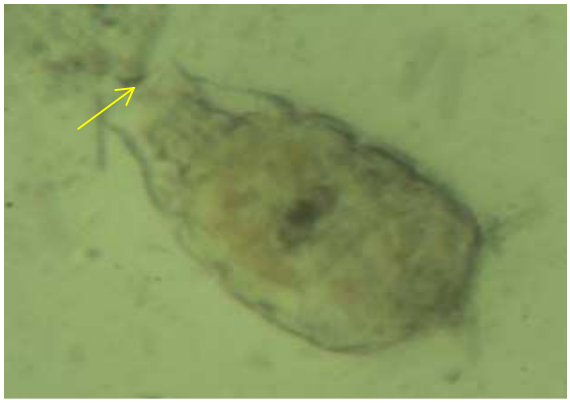


Figure-13. *Gyrodactylus* sp. with clear attachment site (arrow) and attached to the body of *Cirrhinus mrigala* (Wet mount, 100x).

Figure-14. Metacercaria stage of digenean parasites with distinct internal organs (arrow) present in intestine of *Cirrhinus mrigala* (Wet mount, 200x).

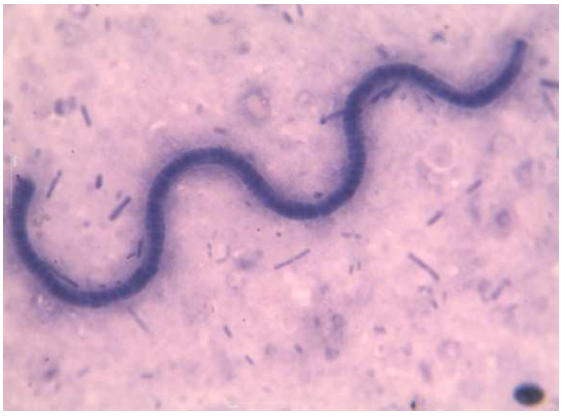


Figure-15. *Capillaria* sp. present in gills of *Cirrhinus mrigala* (Giemsa stained, 1000x).