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Epidemiology and management of parasitic infestations in Indian major carps in extensive culture systems of Andhra Pradesh, India

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ABSTRACT

The occurrence of parasitic infestations in Indian major carps (IMCs) culture systems are seen regularly and is regarded as one the major bottle neck in fresh water fish culture practices causing significant loss to farmers. This study aimed to isolate and identify parasitic threats affecting IMCs in community and panchayat tanks, by characterizing the severity of infestations. The investigation spanned four distinct sites, viz. Srikakulam, Vizianagaram, East Godavari, and West Godavari districts in Andhra Pradesh, India. Approximately, 80-100 infected fish were examined per site, with documentation of gross clinical signs and parasitic burden. *Dactylogyrus* spp., *Gyrodactylus* spp., *Myxobolus* spp., and *Thelohanellus* spp. were identified as the primary parasitic agents affecting the carp populations. Infested fish exhibited characteristic symptoms, including respiratory distress, skin darkening, and increased mortality rates. The findings confirmed parasitic infestations as a major challenge in cultivable fish ponds. Notably, *Catla catla* and *Labeo rohita* displayed higher susceptibility to *Dactylogyrus* spp. and myxozoans (*Myxobolus* spp. and *Thelohanellus* spp.), respectively. For effective management of parasitic infestation in carp culture, adoption of standard disease management procedures such as applying 25 kg/ha of common salt and 1 L/ha of formalin resulted in lower mortality (1-4%) in experimental tanks in comparison with control tanks (5-13.5%). This study enhances our comprehension of management and preventive approaches for reducing diseases caused by myxozoan and monogenean parasites in aquaculture systems.

Keywords: Andhra Pradesh, Carps, Disease management, Epidemiology, Fish culture, Fish parasites, Monogeneans, Myxosporean

The aquaculture sector is gaining momentum as a key source of animal protein, livelihood for millions of poor people and supports the economy. As per FAO (2020), it has been regarded as the fastest growing food production segment over the last decades with higher growth rate of 4.5%, in comparison with other food sectors. The state of Andhra Pradesh had emerged as front runner in aquaculture production over several past decades compared to other states in India. Indian major carps (IMC) contribute about 75% of freshwater fish production in the nation with higher genetic diversity across major riverine systems (Behara *et al.* 2022). However, various disease problems have been noticed in aquaculture not only because of intensification of farming practices, but also due to the movement of different stages of fish such as spawn, fry, fingerlings and brood stock, poor biosecurity measures, pathogenic microbial load and changing climatic conditions (Bondad-Reantaso *et al*. 2005). Also from a general perspective, parasites cause diseases in natural water bodies (Devi *et al.* 2023). The aquaculture sector faces catastrophic socio-economic losses and ecological consequences due to parasitic infections with a high share (46%) which has emerged as a major bottleneck to freshwater aquaculture in India (Mishra *et al*. 2017)**.**

The influence of seasonality on parasitic prevalence evidenced that higher infestation was noticed during pre-monsoon and lower during monsoon season. These infections set in when the host prevails in unfavourable environmental conditions, especially when the water quality is poor (Devnath *et al*. 2024). Small scale farmers are suffering with huge losses because of inadequate knowledge and skills in the disease prevention and control during disease outbreaks (Bagum *et al*. 2013).

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Indian traditional, extensive culture systems with parasitic infections have received particular attention for lack of proper documentation. However, accurate and rapid disease control measures are also ecologically viable and critical for sustainable farming. On the other hand, application of pesticides and other drugs are considered to be significantly toxic both to fish, and other organisms in the ecosystem. Limited studies have been carried out in the state of Andhra Pradesh on the prevalence of fish parasites such as Myxosporean and Monogeneans and their frequent invasion into polycultures was often challenging during winter (Sundeep *et al*. 2016). Prevalence studies help in devising disease control programs and monitoring of the impact of diseases in the food production systems to reduce the infestation levels for a disease over time. With this background, the present study aimed to examine the prevalence and control of selected parasites in traditional fish culture tanks of Andhra Pradesh, India.

MATERIALS AND METHODS

Collection of fishes: In the present study, fish samples were collected from year-round extensive IMCs rearing farms located in Srikakulam, Vizianagaram, East Godavari, and West Godavari districts during the period 2016 to 2019 for the presence of myxobolus and other external parasitic infections during winter season (Supplementary Table 1). The fish samples included fingerlings of Indian major carps (IMC), namely, *Catla calta (vern. Krishna boche), Labeo rohita (vern. ragandi),* and *Cirrhinus mrigala (vern. Erra mila).* The stocking density of each Grama panchayat tank was 2500-3500/ha of 40-50 g of IMC in polyculture model during July and August months of each year. In the winter months (November-January), the disease outbreak was noticed using a cast net during sampling of fishes. A random sampling strategy was followed from four directions of the culture tank. A total of 80-100 fish were collected from each tank for observation and the data on the parasitic prevalence was recorded. At the time of collection, the average temperature of the culture water was around 27-31°C for all tanks.

Examination of fish and data collection on parasitic prevalence: Examination was carried out when the fish attained 3-4 months age and weighed around 200-500 g. Gross observations included a close note of the fish's health status, appearance, and external symptoms for each collected specimen. External parasitic assessments were carried out thoroughly from the fish gills, fins, scales, and operculum by using microscope. The gills and body surface were thoroughly examined for parasites. Furthermore, gross clinical signs and other symptoms were observed and meticulously recorded.

The collected data were analyzed by using formula (Margolis *et al*. 1982), to calculate the prevalence of parasites known to cause infection in fishes. The formula can be referred to as follows for a particular infection site:

$$\text{Prevalence} = \frac{\text{Number of infected fish}}{\text{Number of examined fishes}} \times 100$$

Chemicals used for control of parasitic infestations: In the post-infected farms, application of different chemicals was noticed to control the parasitic infestations. The data was collected from the farmers on the usage of different chemicals in the culture tanks.

RESULTS AND DISCUSSION

The present study focussed on fish parasites, their prevalence, and economic evaluation in polyculture tanks. The outbreak of parasites in extensive ponds of Srikakulam district of Andhra Pradesh was found to be of *Dactylogyrus* spp., *Gyrodactylus* spp., *Myxobolus* spp. and *Thelohanellus* spp. found on carp gills (Figs. 1 and 2) and body surface (Supplementary Figs. 1 and 2). Most of the Monogenea classes were infected the Cyprinids which have high economic value in fisheries of South East Asia (Abidi *et al.* 2011). Prevalence of infection was found to be 21.34% (Table 1).

Clinical sign/symptoms: The fishes were highly symptomatic, lethargic, especially in the early morning hours (6-10 AM). They formed groups and moved nearby

Fig. 1. Gill myxoboliosis in *L. rohita* having pallor gills with severely necrotized and inflamed gill filaments; Gill filaments are uneven or torn showing creamy white gill patches over the entire gills; Massive tissue liquefaction (yellow arrow); Entire gills and body is mucus laden (Black arrow).

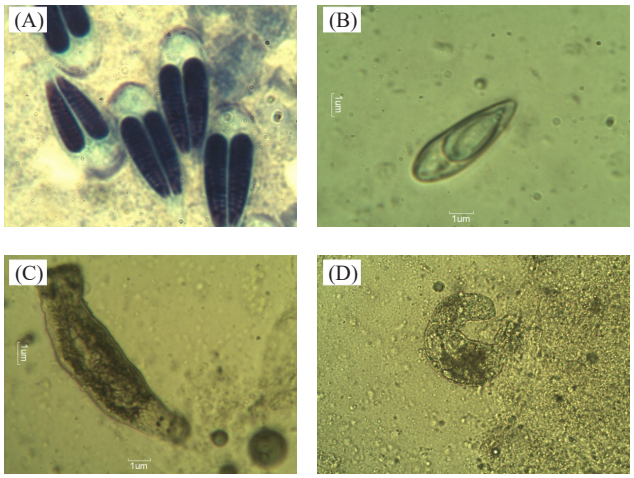

Fig. 2. (A) Spore of *Myxobolus* sp. with distinct polar capsules (arrows) from *Cirrhinus mrigala* (Giemsa stained, 1000×); (B) *Thelohanellus* sp. with distinct single polar filament (arrow) present on the gills of *Cirrhinus mrigala* (400×); (C) *Gyrodactylus* sp. with clear attachment site (arrow) and attached to the body of *Cirrhinus mrigala* (Wet mount, 100×); (D) *Dactylogyrus* sp. with distinct lobes (arrow) present in the gills of *Cirrhinus mrigala* (Wet mount, 200×).

Table 1. Summarized data of parasitic infestation and prevalence at four farm site (2016-19)

Cultured fish	No. of affected fishes	No. of fish examined	Prevalence (%)	Prevalence (%) in IMC
		Myxosporidians		
Catla catla	104	448	23.21	23.66
Labeo rohita	132	420	31.4	
Cirrhinus mrigala	48	240	20.0	
Cyprinus carpio	0	160	0.0	
Total	284	1268	21.4.0	
		Skin Fluke		
Catla catla	112	360	31.1	19.3
Labeo rohita	76	380	20.0	
Cirrhinus mrigala	44	240	18.3	
Cyprinus carpio	0	220	0.0	
Total	232	1200	32.8	
		Gill Fluke		
Catla catla	128	412	31.06	21.1
Labeo rohita	100	432	23.14	
Cirrhinus mrigala	32	200	16.0	
Cyprinus carpio	0	184	0.0	
Total	260	1228	26.03	

tank dyke, or on the surface water, portraying sluggish movement followed by turning towards the left/right side of the body; gills were pale, inflammed, haemorrhagic and necrotized; mucous was laden all over the body and gills; scale loss; fins with haemorrhages and erosion of caudal fin. The observed clinical signs recorded in this study corresponded with the previous reports in *C. catla* by Kumar *et al*. (2018) and Mamun *et al*. (2020).

Diagnosis and prevalence of parasites: The prevalence of various parasites observed from all tanks is mentioned in Supplementary Table 1. The parasites were broadly classified according to tissue specificity and gross clinical signs. Growth reduction and mucus accumulation were common in all parasite-infested fish, exceptionally high in *C. catla* infected with gill flukes (Fig. 1). Similarly, higher mucus aggregation and mortality of *C. catla* was observed in polyculture tanks (Kumar *et al*. 2020). At the same time, the head's size was comparatively on the higher side, while the posterior position, i.e. muscle mass, was emaciated. In technical terms, it is called to be Pinheads. In this study, this was named as "Reverse growth", as the growth of the fish gets reduced in upcoming days post-infestation. Cloudy appearance, creamy white patches of 5-10 mm diameter were observed on gill filaments. The fishes were observed with haemorrhaged fins and erosion of the caudal fin may be due to stress caused by parasites which, in turn, lead to secondary bacterial infections (Abdel-Latif *et al.* 2020). In the present study, the lower mortality rates (1-4%) was recorded in recommended treatment tanks during the study period from 2016-2019, whereas, in the farmers treated tanks, higher mortality rates (5-13.5%) were observed.

Control measures: In the initial days of disease outbreaks, farmers applied pesticides to prevent mortality, but they were ineffective in combating the parasites and which resulted in lower yields (Table 2). A dose of common salt @ 25 kg/ha and liquid formalin 1 L/ha were applied every month for the later cultivation cycle as recommended treatment. For secondary infection control, sanitizer Potassium Permanganate ($KMnO_4$) was applied at a rate of 400 g per acre (0.1 ppm) as prolonged dip treatment. Formalin is commonly used to control external protozoan, monogenetic trematodes, and *Saprolegnia spp.* in aquaculture (Rowlan *et al*. 2006). Common salt can effectively treat fishes infected by *Ichthyophthirius multifiliis* (Ich) within 3 days post-infection (Maceda and Cable 2014), while formalin was effective for continued bath treatment for 6 days to control *I. multifiliis.* Kumar *et al.* (2022) reported supplementation of turmeric oil at an optimum concentration induced anti-stress and anti-oxidative (Darwish *et al*. 2010) responses, and increased survival against parasitic-bacteria co-infections. Following the recommendations, which included using common salt and formalin, increased fish production with higher net returns and B:C ratio was observed due to reduced parasite load (Table 2). Similarly, the significant impact on growth and yield in carp farms was seen due to ectoparasites, protozoans and monogenean parasites (Hossain *et al.* 2022). The spread of infection, production losses, and economic losses were closely monitored in recommended tanks in comparison with farmer treated tanks (Table 2).

Often, infections due to these parasites have minimal effects on the host, unlike viral and bacterial diseases. Nevertheless, heavy parasitic infestations show deleterious effects on host. The fishes do have indirect effects like Reduced fish growth thereby affecting fish production drastically; Reduced feed conversion ratio (FCR), which is another kind of economic loss to farmers in the form of feed wastage on daily basis; and Secondary infections, which worsen the condition further. Even in the present study, mortality was not significant, but chronic losses

Table 2. Production and economic parameters in farmer treated tanks and recommended treatment (Demonstration) tanks

Technology Assessed	Mortality rate %	Production per hectare (tons)	Net return (Profit) in ₹/ha	B:C ratio
	2016			
T_1: FT: Using chemicals after noticing the incidence of disease	13.5	1.21	24200	1.25
T_2: RT: Application of salt @25 kg/ha + Formalin @ 1 L/ha for every month during culture period	3.5	1.56	39000	1.33
	2017			
T_1: FT: Using chemicals after noticing the incidence of disease	6.5	1.38	34500	1.31
T_2: RT: Application of salt @25 kg/ha + Formalin @ 1 L/ha for every month during culture period	2	1.72	60200	1.5
	2018			
T_1: FT:Using chemicals after noticing incidence of disease	9	1.83	51240	1.36
T_2: RT: Application of salt @25 kg/ha + Formalin @ 1 L/ha for every month during culture period	4	1.98	67320	1.48
	2019			
T_1: FT: Using chemicals after noticing incidence of disease	5	2.213	73029	1.43
T_2: RT: Application of salt @ 25 kg/ha + Formalin @1 L/ha for every month during culture period	1	2.403	93717	1.55

Note: FT, Farmer treated; RT, Recommended treatment.

were reported on an average between 5% and 10% across each consecutive year, which accounted for 21.3% of all parasitic infestations.

Parasitic infections are the predominant cause of disease incidences in the freshwater aquaculture sector. A recent publication by Sahoo *et al.* (2020) showed parasitic infections in India are the dominant pathogens with 75% of all the disease incidences during 2016-19. In India, freshwater fishes are prone to various parasitic diseases throughout the culture period (Vineetha and Abraham 2009, Vijaysunderdeva *et al*. 2018). Among them, monogeneans and myxosporidians infestations showed a majority of the infestation rates of 25% and 9%, respectively (Mishra *et al.* 2017). According to Mishra *et al*. (2017), myxosporidians can affect all stages of fish in all seasons (summer, rainy and winter). Monogeneans and myxosporeans contribute about 19% and 13% of all the parasitic diseases, respectively, in India (Sahoo *et al.* 2020). The infections caused by these parasites in fish can cause severe economic loss for the aquaculture sector.

A sudden seasonal change of water temperature can provide opportunities for parasites to infect the host. In four years of repeated sampling in the winter season, the IMC's in the Srikakulam district, Andhra Pradesh, showed the presence of parasites and mortality. Winter was the most vulnerable season for parasitic infestation in fish due to low water temperatures combined with weakened immunity and poor water quality issues (Dash *et al.* 2015, Sahoo *et al.* 2020). In winter, the mortality rate was reported to be slightly high due to myxosporidian parasites, compared with infections by flukes. Previous reports indicate that myxosporidian infections are common and have a high prevalence during winters (Mukherjee *et al*. 2016, Ahmad and Kaur 2018, Sahoo *et al.* 2020). In support of this study, similar results were observed in carps that weights 200-500 g having higher disease incidences (Mishra *et al.* 2017, Sahoo *et al*. 2020).

In India, numerous myxosporean parasites are identified from various fishes; yet the majority of them have been recovered from cyprinids (Kaur and Singh 2012). Among IMCs, rohu (*L. rohita*) has shown a high prevalence rate of 31.4% in myxosporidian infestation; clarifying the sensitivity of rohu to gill myxoboliosis in the present study. *T. bifurcate* is the most pathogenic myxosporidians infecting the gills of *L. rohita*, causing mortalities up to 20-30% (Farhaduzzaman *et al*. 2010, Kaur and Katoch 2014). In the present study, 5-10% of mortalities were observed in myxozoans infested tanks. Apart from common carp, all IMCs were infected with a parasite, and may be due to host susceptibility. Kalavati and Nandi (2007) have documented that gill myxoboliosis was the most widely disseminated disease in many Indian states in aquatic habitats, and was reported in all three major carp species. Reports from Punjab also suggest that, gills are the most infested organ for numerous Myxozoan parasites (Ahmad and Kaur 2018).

Previous studies showed that in the month of December to February, the overall prevalence (51.66%) of infection has been recorded in myxozoans and less in skin flukes (17.5%). During the winter months (December to February), the highest prevalence of 37% has been reported (Banerjee and Bandopadhyay 2010). In this study, overall average of 21.3% prevalence of parasite was observed in IMC's, while the highest was in myxosporeans (23.66%), followed by gill fluke (21.1%) and skin fluke (19.3%). Among IMC's,

Catla fish are found to be very sensitive to both of the parasites. Khatri *et al.* (2009) also described the sensitivity of catla to infections. Sahoo *et al.* (2020) concluded that the reason for high susceptibity of catla to various diseases is that it is a surface feeder and is subjected to temperature fluctuations, resulting in weak immunity. Similarly, in present study significantly high loads of fluke infections was noticed, predominantly in the catla variety.

Kalavati and Narasimahamurti (1985), Adriano *et al.* (2009) and Kaur and Katoch (2014) all observed cyst inflammation and rupturing, which resulted in haemorrhages, and a significant loss of respiratory surface. These monogenetic trematodes can damage skin and gills (Petchimuthu 2018), causing irritation and destruction of gill tissues leading to impairment of breathing. Such damage to the respiratory organs and osmoregulation drastically resulted in suffocation of fish, finally ending up with morbidity and mortality.

In conclusion, the present study presents comprehensive information on the management and treatment of diseases caused by mixosporians and monogenean parasites in extensive culture systems. These parasites were obtained from gills and portions of skin tissues of Indian major carps. Particularly higher prevalence was found in catla and rohu. The findings of this study will help in implementing Best Management Practices (BMPs) essential to protect frequent prevalence of the disease and production losses in aquaculture. The control measures recommended in the study have reduced mortality, increased production and higher B:C ratio than the farmers treated groups. Further advancement of knowledge about the disease cycle, the host-pathogen and the interaction of the environment prior to disease incidence, is very vital for the future of scientific methods of disease control systems.

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