



Population dynamics of crustacean parasites and their effects on the health status of fresh water cultivable fishes

Sadguru Prakash¹, Ashok Kumar Verma^{2*}

¹Department of Zoology, M.L.K.P.G. College, Balrampur, Uttar Pradesh, India

²Department of Zoology, Govt. PG College, Saidabad, Prayagraj, Uttar Pradesh, India

Abstract

An investigation was carried out on the population dynamics of crustacean parasites of Indian major carps and Exotic carps and the effect of these parasites on the erythrocytes, thrombocytes and leucocytes counts, as well as serum glucose and protein levels in Cultivable freshwater species. Out of 150 examined fishes 35 fishes were found infected with total 82 crustacean ectoparasites. The prevalence, intensity and abundance of crustacean parasites were high in Indian major carps than the Exotic carps. The result revealed that exotic carps, *Hypophthalmichthys molitrix* was more susceptible and followed by *Cirrhinus mrigala*, *Catla catla*, *Labeo rohita* and *Cyprinus carpio* to crustacean parasites. Four genera of crustacean parasites were identified as copepods (*Ergasilus*, *Lerneae* and *Lamproglena*) and Branchiura (*Argulus*) from gills, skin and fin bases of infected fishes during study period. Results indicated that presence of these four species of crustacean parasites caused alterations in various haematological indices of both indigenous and exotic carps. Haemoglobin (Hb), Total Erythrocytes Count (TEC), Packed Cell Volume (PCV), Mean Corpuscular Volume (MCV), Mean Corpuscular Haemoglobin (MCH), Lymphocytes, Eosinophil and serum protein level were found decreased in the fishes infested with parasites as compared to those which were devoid of any parasitic infestation. Similarly, Total Leucocyte Count (TLC), Monocytes, Neutrophil, Basophil and Serum Glucose were found increased in parasitized fishes than non-parasitized ones. The results indicated that fishes suffer anemia, loss of appetite and reduced growth when affected with parasites.

Keywords: crustacean parasites, population dynamics, haemato-biochemical indices

Introduction

Crustacean constitutes near about 4200 species and it is the second largest class of the phylum Arthropoda, the largest phylum of kingdom Animalia (Verma and Prakash, 2020) [19]. Most of the crustaceans are free living, some are sessile and few are parasitic (Ashok, 2017; Prakash *et al.*, 2021a; 2021b) [3,15, 16]. Like other crustaceans parasitic crustaceans also have segmented body, covered with exoskeleton and jointed appendages and worldwide distributed in fresh and brackish water bodies. There are three parasitic groups comprising mainly external parasites in cultivable finfish in the class Crustacea; the Branchiura, Copepoda and Isopoda. Members of the Branchiura and Isopoda are relatively large and both sexes are parasitic, while Copepods (the most common crustacean parasites) are generally small even microscopic with both free living and parasitic stages in their life cycle. Male parasitic copepod dies after copulation in pre adult stage hence only mature female attached to fish. Usually these crustacean parasites cause only minor harm to their hosts when present in small numbers. However, in case of heavy infections severe damage to skin, muscles, and gills tissue accompanied with secondary infections can occur. Cultivable freshwater species as well as other wild species have been found to be seriously affected by the outbreak, resulting in severe ulcerations and large scale mortality. Infestation of fishes by crustacean parasites (Fish lice, *Argulus* sp, Gill lice, *Ergasilus* sp, anchor worm, *Lerneae* sp and *Lamproglena*) in natural waters does not manifest in serious disease outbreak but infestation in confined waters very often cause serious disease outbreaks. Clinical signs may vary from minor skin irritation, lethargy,

localized ulceration, reduced appetite and heavy loss. Both Indian major carps, *Catla catla*, *Labeo rohita* and *Cirrhinus mrigala* as well as exotic carps, *Cyprinus carpio*, *Hypophthalmichthys molitrix*, and *Ctenopharyngodon idella* are most severely affected freshwater species are. Fishes of all sizes were equally affected.

Parasitic diseases are the limiting factors in fish culture, because of increased density of fish in lentic water bodies where the fish pathogens can easily transmit from one fish to another. These parasites deteriorate the normal health condition of fish and also affect the reproductive potential of host fishes as well as reduces the fish production, profitability, and their nutritive value, thus results great economic loss of fish farmers (Prakash and Verma, 2017; Prakash and Verma, 2020; Prakash and Singh, 2020) [11,12,10]. Moreover, parasites can have a variety of negative effects on their host, including consuming the host's food, destroying the host's tissues, abnormal growth, mechanical implications, biological effects, different types of tissue reactions, and the release of toxins or poisons. Fish are afflicted by illnesses and parasite infestations much like people and other animals are. Fish have both specific and non-specific defenses against disease. Skin, scales, and the mucus layer released by the epidermis traps pathogens and suppressed the host's growth are examples of non-specific defenses (Singh and Prakash, 2022) [14].

Blood is an efficient bio-indicator of the health of an organism. Haemato-biochemical indices are an important tool to diagnose and monitor the response of organisms to stressors and thus it reveals the health status under such adverse conditions. These haemato-biochemical variables

relate to metabolic levels (energetic), respiration (haemoglobin), and defense mechanisms, acting as physiological indicators to changing external conditions (Parmar and Prakash, 2022) [9]. Being a good pathological reflector of the whole body, haemato-biochemical indices are important in diagnosing the functional status of the fish (host) infested by parasites and also to rise the physiological condition and nutritional status of fish (Tabasum and Dixit, 2021) [17]. Considering the above facts, the present study was carried out to investigate the impact of crustacean parasites on the haematological and serum biochemical parameters of fresh water fishes collected from the lentic water body of district Balrampur, U.P., India.

Materials and Methods

The live freshwater carp (Catla, *Catla catla*; Rohu, *Labeo rohita*; Nanni, *Cirrhinus mrigala*; Silver carp, *Hypophthalmichthys molitrix* and Common carp, *Cyprinus carpio*) were collected from lentic waterbodies of Balrampur with the help of fisherman and transported to the Ichthyology lab, Department of Zoology, M.L.K.P.G. College, Balrampur. The fishes were examined immediately after collection for investigation of crustacean parasites. The external surface such as scales, fins, skin and fin base of host fishes were examined under a magnifying glass for crustacean parasites or any kind of lesions. Then scrapping of the skin was done by a scalpel to collect the mucus in a petri dish for microscopic examination. After that gills were removed from the bronchial cavity and placed on the glass slide for microscopic examination. From these crustacean parasites were collected by a hairbrush then fixed according to method suggested by standard method (Lucky, 1971) [6] and identified according to 'Parasitic copepod and Branchiura' of Fishes by Yamagutti (1963) [21]. Prevalence, Intensity and Abundance of parasitic infection was calculated by following the formula (Margolis *et al.*,1982) [7].

$$\text{Prevalence \%} = \frac{\text{Total no. of infected fishes}}{\text{Total no. of fish examined}} \times 100$$

$$\text{Intensity of Infection} = \frac{\text{Total no. of parasites collected}}{\text{Total no. of infected fish examined}}$$

$$\text{Abundance (\%)} = \frac{\text{Total No. of Parasites collected}}{\text{Total No. fish (Host) Examined}}$$

Blood was collected from the caudal peduncle and heart of infected and uninfected live fishes using 2 ml plastic syringe and needle treated and preserved with EDTA (ethylene diamine tetra acetate, an anticoagulant). The blood parameters like total RBC, WBC, Hb% and PCV were analyzed according to the methods of Dacie and Lewis (1977) [4]. The heparinized blood samples were centrifuged at 300 rpm for 10 minutes and the serum collected for biochemical analysis. Glucose and protein were analyzed according to the method of Mendel *et al.* (1954) [8]. and Lowry's methods (1951) [5]. The data obtained were subjected to statistical analysis using SPSS software.

Results and Discussion

Total 150 fresh water fishes (90 Indian major carps and 60 Exotic carps) were collected randomly from different water bodies of Balrampur district. Out of 150 examined fishes 35 fishes were found infected with total 82 crustacean ectoparasites (Table 1).

Parasitic species often exhibit a heterogeneous infestation pattern and are highly dispersed in host population. In the present study the prevalence of crustacean parasites was high in Indian major carps than the Exotic carps but intensity of these parasites was high in Exotic carp than Indian major carp. In Indian major carp, prevalence of infection was recorded 23.33% in which maximum incidence of infection or prevalence was recorded in *Cirrihinus mrigala* (26.66%) followed by *Labeo rohita* (23.33%) and *Catla catla* (20.00%) where as in Exotic carp, prevalence of infection was recorded 46.66% in which maximum incidence of infection or prevalence was recorded in *Hypophthalmichthys molitrix* (30.00%) and minimum in *Cyprinus carpio* (16.66%) (Table 1).

Table 1: Population Dynamics of Crustacean Parasites from Fresh water Fishes

Name of Fish	No. of Fishes		No. of Parasites Collected	Prevalence (%)	Intensity	Abundance
	Examined	Infected				
Indian major carps						
<i>C.catla</i>	30	06	13	20.00	2.17	0.43
<i>L.rohita</i>	30	07	17	23.33	2.43	0.56
<i>C.mrigala</i>	30	08	19	26.66	2.38	0.63
Total	90	21	49	23.33	2.33	0.54
Exotic carps						
<i>H.molitrix</i>	30	09	21	30.00	2.33	0.70
<i>C.carpio</i>	30	05	12	16.66	2.40	0.40
Total	60	14	33	46.66	2.36	0.55

The average intensity of infection in Indian major carp was observed 2.33 in which maximum intensity was recorded in *Labeo rohita* (2.43) followed by *Cirrihinus mrigala* (2.38) and *Catla catla* (2.17) where as the average intensity of infection in Exotic carp was 2.36 in which maximum intensity was observed in *Cyprinus carpio* (2.40) and minimum in *Hypophthalmichthys molitrix* (2.33) (Table 1). The average abundance of infection in Indian major carp was observed 0.54 in which maximum abundance was recorded in *Cirrihinus mrigala* (0.63) followed by *Labeo rohita* (0.56) and *Catla catla* (0.43) where as the average abundance of infection in Exotic carp was 0.55 in which maximum abundance was observed in *Hypophthalmichthys molitrix* (0.70) and minimum in *Cyprinus carpio* (0.40) (Table 1).

Among the collected parasites, 04 genera of crustacean parasites were identified. Out of four parasites, three were copepod parasites and rest one was Branchiura. Copepod include genus *Ergasilus*, *Lernead* and *Lamproglena* collected from gills and skin; while Branchiura include only genus *Argulus* was collected from skin and fin (Table 2). Presence of crustacean ectoparasites on gills, skin and fin bases of host fishes were also reported by some researchers in Indian major carps (Prakash and Verma, 2020; Prakash *et al.* 2021a) [12,15]. The skin and gill are the main target organs for ectoparasitic infection and local or systemic inflammation is a factor that regulates susceptibility or resistance to infection, and a large number of host molecules involved in immunity modulate this inflammatory responses or process either directly or indirectly (Amriana *et al.*, 2021) [2].

Table 2: Distribution Pattern of Crustacean ectoparasites

Crustacean	Name of Parasites	Site of Infection
Copepod	<i>Ergasilus</i> (Gill Lice)	Gill
	<i>Lerne</i> (Anchor worm)	Skin
	<i>Lamproglena</i>	Gill
Branchiura	<i>Argulus</i> (Fish Lice)	Skin, Fin base

Table 3: Blood Parameters of infected and non-infected fishes collected from lentic waterbody

Blood Parameters	<i>Catla catla</i>		<i>Labeo rohita</i>		<i>Cirrhinus mrigala</i>		<i>Cyprinus carpio</i>		<i>Hypophthalmichthys molitrix</i>	
	Uninfected	Infected	Uninfected	Infected	Uninfected	Infected	Uninfected	Infected	Uninfected	Infected
RBC (x10 ⁶ /mm ³)	1.48 ±0.12	1.12 ±0.14	1.43 ±0.20	1.18 ±0.17	1.48 ±0.28	1.20 ±0.13	1.50 ±0.01	1.23 ±0.07	1.65 ±0.72	1.21 ±0.19
Hb%	9.08 ±0.42	6.18 ±0.32	8.64 ±0.41	6.02 ±0.13	9.68 ±0.91	7.11 ±0.23	7.08 ±1.5	5.12 ±0.12	9.18 ±1.8	6.02 ±0.18
PCV %	30.49 ±0.25	22.12 ±0.17	23.46 ±1.21	18.21 ±0.76	23.02 ±0.18	19.03 ±0.32	22.4 ±1.41	17.65 ±0.19	34.5 ±1.28	21.48 ±1.27
MCV (µm ³)	164.80 ±1.24	122.11 ±1.71	184.98 ±2.47	139.12 ±2.16	165.91 ±1.46	125.13 ±1.24	148.15 ±4.57	157.11 ±1.21	160.30 ±3.52	118.12 ±0.98
MCH (pg)	57.55 ±6.35	37.23 ±2.45	60.62 ±2.41	41.12 ±1.36	56.34 ±7.16	37.12 ±4.21	44.97 ±1.89	27.45 ±2.14	52.18 ±1.72	31.14 ±1.43
MCHC (%)	45.88 ±1.56	52.15 ±1.50	36.85 ±1.22	42.12 ±1.35	35.50 ±2.01	41.8 ±1.43	31.40 ±1.44	37.87 ±2.01	29.32 ±1.28	35.38 ±1.38
TLC (x10 ³ /mm ³)	1.77 ±2.14	2.21 ±1.21	1.11 ±3.21	2.01 ±2.12	1.14 ±4.46	2.31 ±2.12	1.09 ±1.06	1.89 ±1.17	1.02 ±1.28	1.91 ±1.11
Lympho-cytes (%)	70.4 ±1.49	62.9 ±1.21	69.8 ±2.21	61.9 ±1.18	68.9 ±1.73	62.2 ±1.54	72.30 ±3.69	64.5 ±2.12	70.40 ±1.28	61.4 ±1.52
Neutrophil (%)	21.9 ±1.21	27.25 ±1.17	22.6 ±2.24	28.42 ±1.13	21.70 ±2.2	27.8 ±2.5	18.3 ±1.15	25.2 ±1.12	20.5 ±1.28	28.32 ±1.31
Monocytes (%)	4.6 ±0.48	6.5 ±0.21	4.2 ±0.40	5.9 ±0.34	4.4 ±1.01	5.8 ±0.47	6.5 ±3.21	7.7 ±2.42	6.4 ±0.62	7.6 ±0.52
Eosinophil (%)	1.5 0.06±	1.0 ±0.03	1.6 ±0.08	0.9 ±0.04	2.8 ±0.16	1.5 ±0.18	2.4 ±1.54	1.1 ±1.21	1.8 ±0.25	1.0 ±0.22
Basophil (%)	1.6 ±0.48	2.4 ±0.45	1.8 ±0.7	2.8 ±0.8	2.2 ±0.97	2.7 ±0.54	0.6 ±0.00	1.5 ±0.21	1.0 ±0.01	1.6 ±0.12
Glucose (mg/dL)	67.75 ±0.89	71.42 ±0.73	59.83 0.23±	65.95 ±0.55	46.90 ±0.78	55.58 ±0.79	53.75 ±0.81	58.12 ±0.57	69.21 ±0.44	74.23 ±0.45
Protein (mg/dL)	31.8 ±2.42	26.5 ±1.53	29.9 ±1.76	24.3 ±1.87	30.4 ±1.54	26.2 ±1.32	26.58 ±1.63	22.23 ±1.75	30.2 ±1.43	26.7 ±1.25

During this study the results revealed that ectoparasites induced severe haematological alterations in both indigenous and exotic carps. Haemoglobin (Hb), Total Erythrocytes Count (TEC), Packed Cell Volume (PCV), Mean Corpuscular Volume (MCV), Mean Corpuscular Haemoglobin (MCH) and Lymphocytes, Eosinophil were found decreased in the fishes infested with parasites as compared to those which were devoid of any parasitic infestation (Table 3). Similarly, Mean Corpuscular Haemoglobin concentration (MCHC), Total Leucocyte Count (TLC), Neutrophil, Monocytes, Basophil, Serum Glucose and protein level were found increased in parasitized fishes than non-parasitized ones (Table 3).

In the present study the reduction in Hb%, RBC and PCV in infected fishes may leads to anemia because parasites act as stressors and through the primary stages of stress, the PCV are altered due to the release of catecholamine, an enzyme which activates the RBCs to swell as a consequence of fluid entry into the intracellular compartment (Ali *et al.*, 2010; Tabasum and Dixit, 2021) [1,17]. This lower RBC and haematocrit value (PCV) were caused by parasite feeding on the host's blood. The severe anemia leads to the reduction of RBC, PCV and Hb of the infected fishes (Parmar and Prakash, 2022) [9] and the anemia in infested fishes may be as a result of chronic liver inflammation which causes depression of erythropoiesis.

In the present study, the increase in total leucocytes count (TLC) in infested fishes occurred as a pathological response since WBC plays most important role during infestation by stimulating the haemopoietic tissues and the immune system by producing antibodies and chemical substances working as defense against infestation (Ali *et al.*, 2010) [1]. In the present study significant increase in WBC in infected fishes was due to response of cellular immune system to parasitic infection.

In the present study a decrease in the percentage of lymphocytes and eosinophils with significant increase in the percentage of granulocytes (Neutrophils and Basophils) and monocytes in the infested fish. It was reported that blood leucocytes, especially granulocytes and monocytes, could destroy pathogenic organism.

Fish parasites can cause alterations in hematological indices including anemia, leucocytosis, and lymphocytopenia, while *Argulus* parasite infestations can cause stress reactions with behavioral and physiological responses in host fish (Rocha *et al.*, 2018) [13]. For example, Ali *et al.* (2010) [1] demonstrated anemic conditions in infested fish, with a decrease in the number of erythrocytes and hematocrits in host blood due to parasite feeding activity as well as failure of host osmoregulation due to skin lesions.

In the present study the serum glucose level was significantly increased in the infected fish revealed that

parasites take glucose from the host and also during stress conditions fishes consume more energy to counteract the adverse effect of parasites. During stress conditions, epinephrine and cortisol activates increases that help in the breakdown of stored liver glycogen into glucose.

In the present investigation, the protein content in blood of infected fish was decreased. Similarly Upadhyay *et al.* (2009)^[18] also observed depletion in serum protein level in fresh water fish, *Channa striatus* infected with helminth parasites. Depletion of protein in infected fish tissues may be due to the conversion of protein resources for the release of energy to withstand the wear and tear created by the parasite (Yadav *et al.*, 2021)^[20].

Conclusions

Hematological parameters also showed significant differences between infested and non-infested fish, in particular an decrease in number of RBC, Hb% and PCV and also increase in the number of WBC and the percentage of granulocytes and monocyte cells in infested fish, which indicates an inflammatory response to parasitic crustacean infestation in fish. Thus, it can be concluded that haematological indices can be useful for the measurement of physiological disturbances in parasitized or stressed fishes and thus provide information about the level of damage in the host and the prognosis for the various diseases.

References

1. Ali H, Ansari KK, Prakash S. Haematological abnormalities in an exotic carp, *Hypophthalmichthys molitrix*, infested by a fish louse, *Argulus* sp. *Pollution Research*, 2010;29(4):593-595.
2. Amriana, Sari DK, Sriwulan, Anshary H. Prevalence of *Argulus indicus*, histopathology and hematological properties of infected wild fish in lake Towuti, Indonesia. *Biodiversitas*, 2021;22(8):3578-3584. [10.13057/biodiv/d220859](https://doi.org/10.13057/biodiv/d220859).
3. Ashok KV. A Handbook of Zoology. Shri Balaji Publications, Muzaffarnagar. 5th edn, 2017, 648.
4. Dacie JV, Lewis SM. Practical Hematology. Elsevier, 1977, 653.
5. Lowry OH, Rosebrough NJ, Farr AL, Randall RJ. Protein measurement with the folin phenol reagent. *J. Biol. Chem.*, 1951;193(1):265-275.
6. Lucky Z. Methods for the diagnosis of fish diseases. Amerno Publishing Co. Pvt. Ltd. New Delhi, Bombay, Calcutta and New York, 1977.
7. Margolis L, Esch GW, Holmer JC, Kuris AM, Schad, GA. The use of ecological terms in parasitology. *Journal of Parasitology*, 1982;68:131-133.
8. Mendel B, Kemp A, Mayes DK. A colorimetric micro method for the determination of glucose. *Biochem. J.*, 1954;56:639-645.
9. Parmar A, Prakash S. Impact of ectoparasites on Haemato-biochemical indices of snake headed fishes. *International Journal of Zoological Investigation*, 2022;8(2):264-268. <https://doi.org/10.33745/ijzi.2022.v08i02.057>
10. Prakash S, Singh D. Seasonal variation of helminth Parasites in fresh water fishes of Sauwan Nallah of Balrampur, U.P., India. *International Journal of Scientific Research in Biological Sciences*, 2020;7(4):26-29.
11. Prakash S, Verma AK. Incidence of parasites in *Labeo rohita* (Hamilton) at Balrampur (U.P.). *Life Science Bulletin*, 2017;14(2):181-183.
12. Prakash S, Verma AK. Seasonal variation in prevalence of ectoparasitic infection in Indian major carps at Balrampur, U.P., India. *Uttar Pradesh Journal of Zoology*, 2020;41(10):121-127.
13. Rocha MJS, Jerônimo GT, da Costa OTF, Malta JC de O, Martins ML, Maciel PO *et al.* Changes in hematological and biochemical parameters of Tambaqui (*Colossoma macropomum*) parasitized by metazoan species. *Rev Bras Parasitol Vet*, 2018;27(4):488-494. [10.1590/S1984-296120180073](https://doi.org/10.1590/S1984-296120180073).
14. Singh D, Prakash S. Alterations in organic reserves in *Channa punctatus* infected with ectoparasites. *International Journal of Zoological Investigation*, 2022;8(2):534-539. <https://doi.org/10.33745/ijzi.2022.v08i02.067>
15. Prakash S, Yadav DK, Jaiswal L. Population Dynamics of Crustacean parasites of freshwater fishes at Bahraich, U.P. *International Journal of Zoology and Applied Biosciences*, 2021a;6(1):15-18.
16. Prakash S, Yadav DK, Jaiswal L. Protozoan parasites of fishes from Chittorgarh reservoir, Balrampur district (U.P.). *International Journal of Zoology and Applied Biosciences*, 2021b;6(3):110-113.
17. Tabasum B, Dixit R. Hematological effects of ectoparasites in some catfishes of Indore, Madhya Pradesh. *Research Journal of Agriculture Sciences*, 2021;12(2):405-410.
18. Upadhyay N, Bansal A, Goyal RK. Effect of Helminth parasites on biochemical profile of freshwater fish, *Channa striatus*. *Asian Journal of Microbiology, Biotechnology, Biotechnology and Environmental Sciences*, 2009;11(4):847-849.
19. Verma AK, Prakash S. Status of Animal Phyla in different Kingdom Systems of Biological Classification. *Int. Journal of Biological Innovations*, 2020;2(2):149-154. <https://doi.org/10.46505/IJBI.2020.2211>
20. Yadav R, Narayan A, Singh D, Rajpoot V. Prevalence of Piscean cestode, *Mystoides chhaviensis* in freshwater fish from Bundelkhand region of Uttar Pradesh, India. *International Journal of Biological Innovations*, 2021; 3(2):407-410. <https://doi.org/10.46505/IJBI.2021.3223>
21. Yamagutti S. Parasitic Copepoda and Branchiura of Fishes. Interscience Publishers, John Wiley & Sons. New York, 1963, 1104.