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Icthyo parasitic fauna and its prevalence in Khyber Pakhtunkhwa, Pakistan

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Diseases caused by parasites in fish species got least attention from farmers especially in Khyber-Pakhtunkhwa Pakistan because of which they faced high mortality and huge economic losses. For this purpose, in total 76 fish specimens of 17 ponds were investigated (from September to November 2021), to explore their ectoparasitic fauna, in selected districts of Khyber-Pakhtunkhwa (Peshawar, Charsadda, Mardan and Swabi) Pakistan. The results revealed that 59 fish samples were infested by the parasites including *Lernaea sps* [prevalence 39.47%], *Gyrodactylus sp,* [prevalence 19.73%], *Dactylogyrus sp,* [prevalence 17.10%], *Icthyopthyrius multifilis sp* [prevalence 13.15%], *Thelohanellus sps* [prevalence 11.84%], *Piscinoodinium pillulare* [prevalence 7.89%], *Loxophyllum sps* [prevalence 10.52%]. Most prevailed parasites reported in the current studies were *Lernaea sps,* which are the highest mortality causative agent of ichthyofauna, followed by *Gyrodactylus sp,* and less prevailed parasites reported in our studies was *Piscinoodinium pillulare.* All these mentioned parasites were reported for the first time from the study area.

Keywords: Fish, Ichthyofauna, Ectoparasites, Economical loss, prevalence, Mortality

INTRODUCTION

Aquaculture is one of the world's most commercially important sector, and fishes are one of humanity's most valuable and nutritional resource (Dar et al., 2014). With the ever-increasing demand for lower-cost protein sources, fish is receiving more and more attention. Therefore, fish must be free of all the diseases in order to produce healthy and high-quality fish meat (Bash et al., 2001). Parasite is an organism that resides in/on the body of a larger species during its life cycle and causing a variety of problems (Llewellyn et al., 2014). Infections caused by parasites has been discovered to be quite fatal on occasion which has the potential to increase the mortality of aquatic fauna (Ahmed, 1996). Fish ectoparasites are one of the most serious issues in pond fish farming (Yin et al., 1963). The presence of parasites in aquatic species indicates that aquatic habitats are degrading (Rueckert et al., 2009). Infectious parasites infecting fish have a direct or indirect impact on human health. Living parasites as well pathogens that have already died present in fish tissue can cause allergic reactions in humans (Buchmann et al., 2001). Pakistan has 193 freshwater fish species and out of which30 spp are commercially very important (Ali, 1993).The parasites assault, producing vast destruction and catastrophic diseases in fish, eventually leading to extinction, as well as being damaging to human health (through food chain). The goal beyond this study was to find out how common ectoparasites are in fish fingerlings from the *Cyprinidae* family, which have a high market value in Pakistan (Habib et al., 2019). Protozoa, monogeneans

(Platyhelminthes), and crustaceans (Arthropoda) are the most common ectoparasitic groups and protozoans are mostly free-living or commensal on fish. Although some of them can cause significant diseases that may eventually results in mortality. The ectoparasites ciliates are the most common (Buchmann et al., 2001). Lernaea sp, Argulus sp, Dactylogyrus sp, and Monogenea sp, Lernaeosis are prevalent coincidental agents of parasite infection that have been found to inflict considerable economic losses in Pakistan's fisheries sector (daulat & khan, 1976).

MATERIALS AND METHODS

Study area;
In total, 76 fish samples were collected from 13 ponds of zone 2, Peshawar, Charsadda, Mardan, Swabi. Khyber-Pakhtunkhwa Pakistan. District Peshawar lies between 34.0151° N, Latitude and 71.5249° E longitude and is the capital city of Khyber-Pakhtunkhwa province with total area of 215 km². District Chasradda lies between 34.1682° N latitude and 71.7504° E longitude with total area of 996 km². District Mardan lies between 34.1986° N, latitude and 72.0404° E longitude with total area of 1632 km². While District Swabi lies between 34.1241° N latitude and 72.4613° E longitude with total area of 1,543 km².

Fish collection and processing;
The collected fish samples from the study site were were brought immediately in oxygenated bags to the lab, department of Zoology Bacha Khan University Charsadda. Morphometric measurements were done by taking total length [TL] fork length [FL] standard length [SL] anal length [AL] preorbital length [POL] post orbital length [POL] head length [HL] and body depth [BD]. The fishwere weighed by using digital scale and species identification was done by using method of Hoffman, 2019. Identified species were *C.carpio, H.molitrix, L.rohita, C.catla, C.idella, O.niloticus,* which belongs to 2 families, Cyprinidae and Cichlidae.

Parasitological study
For parasites collection and identification, the fish samples were deeply examined with naked eyes after that it was examined with hand lens, naked eye parasites including lernaea sps were removed by using forceps and preserved in 70% ethanol. For microscopic ectoparasite, examination was conducted using the method of scraping (Kabata, 1985). For further studies the isolated parasites were collected, counted, and processed following Gussev (1986).

RESULTS AND DISCUSSION

The results indicated that 59 fish species were infested by different parasites include *Lernaea sps* [crustaceans]. *Gyrodactylus* sp and *Dactylogyrus* sp [trematodes]. *Icthyophthirius multifiliis*, *Thelohanellus* sps, *Piscinoodinium pillulare and Loxophyllum helus,* [protozoans] as shown in Figure 1. These parasites were reported for the first time from these districts, After observation and identification, analysis of the parasites was done by calculating their prevalence and intensity by equation 1 and 2. Parasitic species, their natural hosts, prevalence and intensity have been shown in Table 1.

$$\text{Prevalence} = \frac{\text{number of infested fishes}}{\text{number of fishes examined}} \text{ X } 100\% \quad (1)$$

$$\text{Intensity} = \frac{\text{number of parasites}}{\text{number of fishes examined}} \quad (2)$$

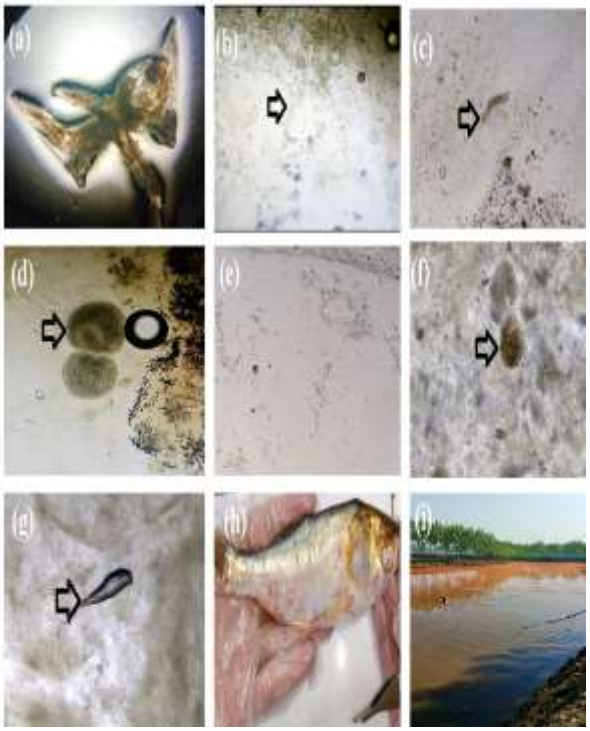

Figure :1 (a) *lernaea sps,* (b) *Gyrodactylus sp (c) Dactylogyrus* sp, (d) *Icthyopthyrius multifilis* sp, (e) *Thelohanellus* sps (f) *Piscinoodinium pillulare* (g) *Loxophyllum* sps (h)Fish intensely infested by *Lernaea* sps, (i) Polluted water pond at Charsadda,

Lernaeid copepods are among the most dangerous

parasites of freshwater fish fauna particularly juvenile fish, which can be killed by a single infestation (Reed et al., 2012). The life cycle of Lernaea species includes three free-living naupliar stages, five copepod phases, and one adult stage. Male die after mating, while females transform and insert their anterior bodies into the host tissue before producing eggs (Dove & Ernst, 1998). Gyrodactylus sp. body shape was discovered to be fusiform. There were no eyespots that have two backers and 16 marginal hooks. Marginal hooks are utilized for attachment to the fish (Host), while anchors are used to aid the marginal attachment hooks (Sharma et al., 2011) . Parasites of fish, Gyrodactylus sp. on the skin causes superficial epidermal lesions by attaching and feeding on the epidermis, producing an increase in mucus production that disrupts the skin's respiratory function (Ferguson, 1989). Dactylogyrus is one of the largest genera of parasitic helminthes, approximately 95% of them are parasites of the gills of fishes in the family Cyprinidae (Francis-Floyd & Reed, 1991). The precise observation of copulatory components of diverse shapes and sizes in various Dactylogyrus species is directly dependent on morphological identification. Because Dactylogyrus is a small parasite (typically less than 1 mm), distinguishing between species can be challenging, especially when flattened marginal hooks appear in viscous gill fluid during the mounting step (Hoffman, 2019). I. multifiliis is round or oval, measuring ± 50-100 µm, with a macronucleus, and micronucleus (Kudo, 1933). This parasite has a transparent macronucleus in the shape of a horseshoe, as well as a micronucleus attached to the macronucleus (Zhang et al., 2013). *Thelohanellus* species were basically placed in the genus *Myxobolus* Bütschli, 1882 due due the precence of iodinophilous vacuole in the sporoplasm, and other general features of the spore (pyriform, two valves, no tails) (Carreon et al., 2021). Subsequently, there are 108 nominal species of Thelohanellus and majority of which are plasmodia-forming parasites that infect various tissues of freshwater fishes, and some of which are pathogenic, inflicting economic losses in aquaculture (Kabata, 1985).

Table 1: Parasitic specie, their natural host, prevalence (%) and intensity

No	Parasitic species	No of samples	No of fish infested	No of ectoparasite	Natural host	Prevalence in %	Intensity
1	*lernaea sps*	76	30	93	*C.carpio H.molitrix L.rohita C.catla*	39.47	1.22
2	*Gyrodactylus sp,*	76	15	22	*C.carpio H.molitrix L.rohita*	19.73	0.28
3	*Dactylogyrus sp,*	76	13	19	*C.carpio C.idella O.niloticus*	17.10	0.25
4	*Icthyopthyrius multifilis sp,*	76	10	15	*C.idella L.rohita, O.niloticus*	13.15	0.19
5	*Thelohanellus sps*	76	9	88	*L.rohita H.molitrix C.idella*	11.84	1.15
6	*Piscinoodinium pillulare*	76	6	13	*C.catla L.rohita*,	7.89	0.17
7	*Loxophyllum sps*	76	8	7	*C.carpio L.rohita*	10.52	0.09

Piscinoodinium pillulare is a common dinoflagellate protozoan found in tropical freshwater fishthat became famous for causing diseases known as “rust disease” or “velvet disease. It parasitizes the gills and the surface of the fish. Rhizocysts are structures that help the parasite attach to the host by penetrating the host cells, producing structural alterations and eventually cause cell death (Nagasawa et al., 2007). The ciliate species of the pleurostomatid genus Loxophyllum live in both marine and freshwater environments and can be found all over the world,[26] Loxophyllum sps body is elongated, with cells measuring 100–300 40–70 m, with the majority measuring 200–250m, with a

beak-like anterior end and a bluntly pointed posterior end, the body is slightly contractile (Lin et al., 2008).

CONCLUSION

In conclusion, the ponds of selected districts of Khyber-Pakhtunkhwa (Peshawar, Charsadda, Mardan and Swabi) Pakistan are more exposed to ectoparasites. Among these parasites *lernaea* sps were more prevailed with highest percentage 39.47. During our studies it was also observed that those fish species which were effected with lerneosis had retarded growth. Serious attention is needed from fisheries department Khyber-Pakhtunkhwa Pakistan for the management of fish parasite. Furthermore research on their biological control/chemical control must be initiated on emergency basis in order to make sure a hygienic supply of fish to market, to prevent economical loss and to maintain the natural taste of fish (as parasitic infestation alters the natural taste of fish meat). First author is thankful to Mrs. Nazia Ehsan, Department of Zoology wildlife and Fisheries, University of Agriculture, Faisalabad, her immense guidance, friendly behavior and further attitude always inspired us to do our best in the research work.

CONFLICT OF INTEREST

The authors declared that present study was performed in absence of any conflict of interest.

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AUTHOR CONTRIBUTIONS

AK presented the idea, IU and BJ have supervised and wrote the manuscript, SU and AUJ did the experiment. SU, MS, ZU and HU reviewed and reform the manuscript. All authors read and approved the final version.

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