

**EPIDEMIOLOGICAL STUDIES ON HELMINTH
PARASITES IN CATFISHES OF GOMTI RIVER,
LUCKNOW**

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BY

KM. RESHU

Enrollment No. 953/17

Supervisor

DR. NEESHMA JAISWAL

Assistant Professor

DEPARTMENT OF ZOOLOGY
SCHOOL OF LIFE SCIENCES

BABASAHEB BHIMRAO AMBEDKAR UNIVERSITY
(A CENTRAL UNIVERSITY)

VIDYA VIHAR, RAEBARELI ROAD, LUCKNOW- 226025 (U.P.), INDIA

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SUMMARY

India is the third largest fish-producing country in the world. As of fiscal year 2022-23, the anticipated total fish production is 16.25 million metric tons (MMT), with 12.12 MMT originating from the inland sector and 4.13 MMT from the marine sector. The fish industry demonstrated an impressive annual average growth rate of 7% over the preceding five years. Contributing approximately Rs. 1,37,716 Crores to the total Gross Value Added (GVA) at constant prices for the fiscal year 2022-2023, the fishing industry plays a substantial role in the economy as a whole.

In terms of nutritional security and economic stability, fish is essential for a significant portion of the human population. Belton and Thilsted (2014) describe fish meat as the most cost-effective animal protein available to the impoverished, as well as a high-protein and low-fat food.

Aquaculture is a sector of the food industry that is expanding at a rapid pace, and the fish meat that is produced by aquaculture is in high demand due to the excellent nutritional value that it possesses. (Tidwell and Allan, 2001). Ornamental fish are kept as pets and for recreational purposes. The issue of parasites and their significance in fish health, productivity, hygiene, and behavioral change has been brought to the forefront by the expansion and growth of fishery and aquaculture in recent decades (Rogers, 1978).

Furthermore, parasite infections not only have a negative impact on their hosts by modifying their physiology, behavior, nutritional status, and injuries. (Ljubojevic et al., 2015) but also found to deteriorate the reproductive system of the host fishes (Rogers, 1978). The food value of the majority of fish is diminished and mortality may occur as a result of the excessive parasite infection. Furthermore, the transmission of certain zoonotic helminth parasites to humans is exclusively by means of fish (Khanum et al., 2008). Additionally, individuals may develop catastrophic secondary infections because of the severe weakness that their infections induce. (Panayotova-Pencheva, 2013; Rondon et al., 2017).

Nematodes are recognized as highly detrimental parasites for fish and are deemed the most lethal among helminths (Yasmin and Bilqees, 2007). A comprehensive study on parasitic nematodes in freshwater fishes of Europe contributes contemporary insights into the taxonomy, biology, and ecology of these parasites (Moravec, 1995).

Fish-infecting trematodes are found in the subclasses Aspidogastrea and Digenea under the phylum Platyhelminthes. Also referred to as flukes (Petty et al., 2022). Trematodes mostly endoparasites are a group of fish parasites, that inhabit various fish species in larval or adult stages. Fishes infected with digenetic trematodes show decreased growth rate, increased mortality, and reduced market value.

Cestodes are prevalent in newly imported or wild-caught fish, exhibiting an indirect life cycle where fish serve as both intermediate and final hosts. Diphylobothrium, a type of tapeworm, has a larval stage that can be fatal to fish and infected to humans. Tapeworms infect various parts of the fish, including the alimentary tract, muscles or other internal organs, often manifesting in symptoms such as swollen abdomen, general unthriftiness. and wasting. Swimming difficulties may also occur in infected fish. Cestodes prevail as the primary endohelminths in elasmobranchs and exhibit limited noteworthy variations in bony fishes, specifically within Caryophyllidea, Proteocephalidea, and Pseudophyllidea (Cribb et al. 2002)

Parasitic infestations of fish in tropical and subtropical regions pose a significant challenge to aquaculture, as they result in substantial economic losses, whether directly or indirectly. The issue of parasitic infestation doesn't just pertain to the economic losses in the country but also affects human health as certain human zoonotic diseases are caused by a variety of significant helminth parasites, which are transmitted to humans through the consumption of raw or partially cooked fish.

The present research and thesis work deals with the Morphological and molecular identification of helminth parasite of catfishes inhabiting the two main sites of Gomti river, Lucknow, Uttar Pradesh, India. Additionally, prevalence and population dynamics of helminth parasite and the effect of different abiotic factors and seasonal variation on helminth parasite in cat fishes is studied so as to complete the research work.

Lucknow, which stands at an elevation of approximately 123 meters (404 ft) above sea level. Lucknow district covers an area of 2,528 square kilometers (976 sq mi) (Lucknow District Population Census, 2011; Lucknow Population statistics, map, and location 2014), bounded on the east by Barabanki, on the west by Unnao, on the south by Raebareli and in the north by Sitapur. The city Lucknow along with 15 prominent cities situated on the banks of river Gomti pour untreated sewage, domestic wastewater, industrial waste, and effluent from sugar factories and distilleries with negative effects on the ecosystem, threatening particularly ichthyo-fauna. The

river Gomti enters Lucknow after covering 240 km of its journey from its origin. Kathna, Reth, Sarayan, Luni, Kalyani, and Sai rivers are the tributaries of river Gomti which serves as a domestic water supply to different geographical areas of the country. (Nagpure et al., 2001; Sarkar et al., 2010).

Gomti river Located in the wooded Pilibhit district of Uttar Pradesh, India, the natural lake known as "Gomat Taal" (at an elevation of around 200 m; North latitude 28°34' and East longitude 80°07') is the headwater source of the Gomti River, a significant tributary of the Ganga. The Gomti passes through Lucknow after a distance of 190 kilometers (120 mi), supplying the city with water for approximately 30 kilometers (19 mi) (Bano and Serajuddin, 2016). The Gomti River is home to a rich fish biodiversity, consisting of 56 species distributed across 41 genera, 9 orders, and 21 families. Following Order Cypriniformes (33.91%), which contained the greatest number of species, the following orders were identified: Perciformes (17.85%), Ophiocephaliformes (5.37%), Mastacembeliformes (3.59%), Mugiliformes (1.79%), Beloniformes (1.79%), and Tetraodontiformes (1.79%) (Bano and Serajuddin, 2016).

The study is focused on the parasitological investigations of the two edible catfishes *Rita rita* and *Wallago attu* for the period of two consecutive years i.e. from July 2021 to June 2023 at two different sites of the Gomti River, Lucknow named Kudiya ghat (latitude 26.874857°, longitude 80.911069°) and Pipraghat (latitude 26.83315°, longitude 80.968279°).

Every year 420 fish samples per species were brought from every single sampling site. For the duration of two years, from July 2021 to June 2023 the total number of dissected fish samples of one species from a single site is 840. So, the total number of dissected fish of both species of *Rita rita* and *Wallago attu* from both the sampling sites for the complete duration of the study is 3360.

Fresh and preferably live specimens of two common edible catfishes i.e. *Rita rita* and *Wallago attu* of all sizes and weights were collected with the help of local fishermen from both the sampling sites i.e. Kudiya ghat and Pipra ghat of Gomti river at Lucknow for the duration July 2021 to June 2023. All specimens of fishes were transported to the parasitology laboratory, BBA University, Lucknow then the fishes were identified using standard taxonomic keys (Talwar and Jhingaran, 1991) and were subjected to parasitological investigations.

The standard length (from the tip of the snout to the end of the base of the caudal peduncle) in centimeters (cm) and the weight of each fish was measured using an SF 400 digital weighing scale in grams (g).

The host fish was anesthetized by clove oil or by cutting at the neck region. The fishes were dissected along the midventral line and the gastrointestinal tract was examined so as to check the presence of helminth parasites. The isolated helminth parasites which include nematodes, trematodes and cestodes were washed with the help of normal saline (Kumari et al., 2018) then processed for morphological analysis. The collected parasites were quantified, documented and processed for further identification according to the standard techniques (Eiras *et al.*, 2006).

In the study of helminths, microscopy is being used more and more as a potent investigative tool. This has improved our understanding of the intricate life cycles and host-parasite connections. Its application in parasitology extends beyond taxonomy to include multidisciplinary research on parasites, including epidemiology, disease control, and functional studies (Halton, 2004). Accordingly, drug efficacy and drug resistance tests have shown the value of using scanning electron microscopy to comprehend the tegumental character of parasites (Roy et al., 2012; Giri and Roy, 2014). Helminth parasite identification is typically done using a light microscope, however as technology has advanced, SEM has been employed as a complimentary tool for morphological identification (Roy and Tandon, 1993). Recently, molecular technologies combined with various microscopical methods have been employed and widely recognized for determining the true taxonomic classification of parasites (Ghatani et al., 2014; Robles et al., 2014). Thus, for the structural characterization of helminth parasites, SEM analysis remains one of the most important methods in helminthology.

The scanning electron microscope (SEM) facilitates a thorough understanding of the surface architecture of helminths, which includes the head and tail regions such as the buccal cavity, teeth, lips, cuticular ridges or ornamentations, spines papillae, alae, vulva, bursa, and spicules in the case of nematodes; scolex, bothria, hooks arrangement, the nature of segmentation in cestodes and oral suckers, acetabulum, holdfast (metacercariae), haptors, sensillary organs, etc. in trematodes (Tandon and Roy, 2002; Halton, 2004). Numerous publications have demonstrated the use of SEM as a supplemental tool in the taxonomy of helminths, including fish parasites (In Camallanidae: Santos and Moravec, 2009; Chaudhary et al., 2017). Quiazon et al. (2008), Kanerek and Bohdanowicz (2009), and Gustinelli et al. (2010) in Anisakidae. Amin et al. (2016).

The identification of morphological features of parasites was carried out by following established standards in the scientific literature. This process referenced the works of (Yamaguti, 1959, 1961, 1963, 1971) and the comprehensive Keys to the Trematoda (Volumes 1-3) authored by (Gibson et al., 2002; Jones et al., 2005, and Bray et al., 2008). Additionally, the CIH Keys to the Cestode Parasites of Vertebrates, as compiled by (Khalil et al., 1994), and the CIH Keys to Nematodes (Nos. I-X), curated by (Anderson et al., 1974-1983), were consulted to ensure a thorough and accurate identification process.

Helminth parasites that had undergone morphological analysis by light and scanning electron microscopy revealed the presence of cephalic plates in the buccal cavity, amphid, phasmid, bifid mucron on posterior part, nerve ring, excretory pore, externally visible spicules, different caudal suckers and gubernaculum morphological characters lead to the characterization of nematode parasite as *Truttaedacnitis sandeepi* n.sp. in **Rita rita** fish whereas *Opisthorchis sp.* in *Rita rita* fish was characterized on the basis of the presence of minute serration all over the body, presence of oral sucker, ventral sucker, cirrus pouch, uterus filled with eggs, vitelline glands, testis, ovary and excretory pore. The morphological characterization of *Gangesia agransensis* in *Wallago attu* was based on the presence of scolex (wider than neck region), cone like rostellum with single row of rostellar hooks surrounded by retractor muscle, rostellar hooks are paired in groups of five, uninoculated suckers with small hooklets arranged in two rows. After the morphological analysis the isolated helminth parasites were also identified at the species level based on molecular techniques by using the genetic markers 18S, 28S, and ITS2. For molecular analysis the isolated gastrointestinal helminth parasites from host fishes *Rita rita* and *Wallago attu* were washed in PBS and then fixed in 100% molecular grade ethanol and further preserved at -20°C. Molecular techniques are employed with morphological techniques to ease the taxonomic identification process of the parasites.

The DNA was isolated by using a Qiagen Dneasy blood and tissue kit. The isolated DNA's quality was evaluated on 1.0 % agarose. The extracted DNA samples were utilized for conducting PCR amplification. The genetic markers employed in this procedure consist of the rDNA 18S, 28S, and ITS2 regions. Electrophoresis was performed by the protocol described by Sambrook (Sambrook, Fritsch, et al, 1989). To determine DNA size, gels of different agarose strengths were utilized, each matched to specific ranges of DNA base pair (bp). Electrophoresis was conducted using a

horizontal agarose slab gel apparatus. For the separation of genomic DNA or RNA, a 0.8% agarose gel was prepared in 0.5X TBE buffer (45 mM Tris-borate, 1mM EDTA, pH 8.0). To create the gel, 0.8 g of agarose was dissolved in 100 ml autoclaved water containing 0.5X TBE buffer and heated. After cooling to approximately 60°C, 1 µL of ethidium bromide (10 mg/ml in TE buffer, pH 8.0) was added, and the mixture was poured into the electrophoresis tray with fixed combs. Once solidified, the agarose gel was transferred to an electrophoresis tank containing 0.5X TBE buffer. DNA samples, along with DNA molecular weight markers/ladder, were loaded into the gel using 6X DNA loading dye. Electrophoresis was conducted at a rate of 5 V/cm. Following electrophoresis, the gel was examined under transmitted UV light (302 – 312 nm) using a transilluminator (Fotodyne Inc. USA) and then photographed. After the amplification process, the PCR product was purified using a Qiagen PCR product purification kit. Further, the purified PCR product was sent to Biosciences Bengaluru. The quality of the resulting sequences was evaluated using sequence scanner software v2, and submitted to NCBI-GenBank, where accession numbers were obtained.

The sequences obtained, in addition to sequences from other relevant helminth species, were obtained from GenBank for analysis. Various characterization tools and software programs, listed below, were employed for the analysis of these sequences. The Sequence Scanner software version 2.0 is applicable for visualizing, printing, and exporting sequence data produced by the BDT v3.1 Cycle sequencing kit on an AB1 3730x1 Genetic Analyzer. The Basic Local Alignment Search tool (BLAST) is a software designed for comparing fundamental biological sequence data, including amino acid sequences of various proteins (blastp/blastx) or nucleotides of DNA sequences (blastn). The NCBI BLAST homepage serves as a gateway for conducting sequence alignment online. A BLAST search identifies areas of similarity between biological sequences. The phylogenetic analysis involved the creation of neighbor joining trees for all the produced sequences by employing the specified bioinformatics tools MEGA 11 (Molecular Evolutionary Genetics Analysis 11). The genetic relationships among each taxon were deduced from phylogenetic analysis. The 18S and ITS2 sequence analyses of the two helminth parasites of class nematoda and trematoda from the host fish *Rita rita* revealed that they belong to *Truttaedacnitis sandeepi* n.sp. and *Opisthorchis* sp. The 28S analyses of the one cestode parasite of *Wallago attu* revealed that they belong to *Gangesia agragensis*. The obtained accession no. for the sequences of *Truttaedacnitis sandeepi* n.sp., *Opisthorchis* sp. and *Gangesia agragensis* is PP713069, PP724412 and PP725266 respectively. The phylogenetic analysis revealed that the respective homogenates of 18S, ITS2, and 28S amplicons belonged to the family of helminth nematode (*Truttaedacnitis sandeepi* n.sp.), trematode (*Opisthorchis pedicellate*), and cestode (*Gangesia agragensis*). To study the effect of different physiochemical water parameters on the parasitic infection prevalence of host fishes, *Rita rita* and *Wallago attu* at the site Kudiya ghat and Pipra ghat for the duration of July 2021 to June 2023, the physiochemical water parameters which were taken into account were pH, Temperature (°c), Total hardness (mg/l), Alkalinity (mg/l), Dissolved oxygen (mg/l) and Biochemical Oxygen demand (mg/l) were taken from the Pollution Control Board, Lucknow.

pH:

Possessing the ability to measure the concentration of hydrogen ions in water, pH serves as a tool to evaluate the level of acidity or alkalinity. It does not directly cause any harmful effects on an individual's health. pH is a fundamental parameter crucial in regulating the majority of chemical reactions in aquatic environments. The measurement of pH was conducted on-site during sample collection using a YK-2005b device from Lutron, manufactured in Taiwan.

Temperature:

Biological processes hinge on complex arrays of biochemical reactions, shaped by physical elements like temperature. The temperature at the sampling site was measured using a YK-2005b instrument from Lutron, produced in Taiwan, and documented in degrees Celsius at the time of sample collection (°C).

The morphological analysis of isolated parasites

To perform the epidemiological study the prevalence and population dynamics of helminth parasites of piscine host fish *Rita rita* and *Wallago attu* were recorded for the period of July 2021 to June 2023. A total of 3360 fishes belonging to the genus *Rita rita* and *Wallago attu* were subjected to parasitological investigation, out of which 1388 host fishes were found to be infected with different helminth parasites. The overall prevalence of helminth parasites in selected host fishes was observed to be 41.30 %. In the case of 1680 examined *Rita rita* host fishes, 857 fishes were found to be infected with helminth parasites with an overall prevalence % of 51.01%. Whereas in the case of *Wallago attu*, 1680 sampled fishes were examined for parasitological presence out of which 531 fishes were found to be infected with the cestode parasite with an infection prevalence of 31.96%. The highest infection prevalence was observed for trematode parasite *Opisthorchis pedicellata* (51.01%) in host *Rita rita*, followed by *Truttaedacnitis heterodonti* a nematode parasite (Prevalence %: 38.45%) in *Rita rita* and then by *Gangesia agragensis*, a cestode parasite with prevalence % of 31.96% in *Wallago attu*.

1. *Truttaedacnitis sandeepi* n.sp.

Seasonal patterns at both the sites of Gomti River, for the parasitic population of *Truttaedacnitis sandeepi* n.sp. (nematode) for two consecutive years (July 2021 to June 2023) was found that maximum infection prevalence was seen for the Rainy season followed by Summer and then winter season. The reason behind the maximum prevalence in the rainy season could be attributed to the breeding period of *Rita rita* i.e. from, May to September with the peak between July and September Saxena (1972) Rahman and Mollah (2013) and Alam (2016). The association between reproduction and the increase in prevalence and abundance of parasites has been attributed the fact to the physiological stress of the host during the breeding period, as higher investment in reproduction may decrease the energy allocated to the immune system and thereby facilitate parasitic infestation (White *et al.*, 1996; Lizama *et al.*, 2006).

1. *Opisthorchis sp.*

During the whole study, the infection of *Opisthorchis sp.* (trematode parasites) was observed throughout the year from the gallbladder of host fish *Rita rita* which was also observed by Khanum et al. (2008) in his study. Seasonal patterns at both the sites of Gomti River, for the parasitic population of *Opisthorchis sp.* (trematode) for two consecutive years (July 2021 to June 2023) was found that maximum infection prevalence was seen for the Rainy season followed by Summer and then winter season. The seasonal cycle of *Opisthorchis sp.* depends on the presence of the infectious stage in intermediate hosts, which is reliant on physicochemical environmental factors. The peak invasion of *Opisthorchis pedicellata* from August to October may be linked to the peak snail invasion during these months (Madhavi 1979). When the snail population was highest, the fluke was transmitted to the fish at the highest rate.

1. *Gangesia agraensis*

Seasonal patterns at both the sites of Gomti River, for the parasitic population of *Gangesia agraensis* (cestode) for two consecutive years (July 2021 to June 2023) was found that maximum infection prevalence was seen for the Rainy season followed by winter and then summer season. The observations give useful insight into climate change and other environmental stressors on the dynamics and distribution of parasites across specific hosts and their habitats.

It is observed that the fishes belonging to intermediate length groups are highly infected with the helminth parasites showing the highest infection prevalence as compared to the smaller and larger length groups of fishes. Nahar (1988) also reported a similar finding that the hosts with intermediate size were more infected by the parasites than the smaller and larger individuals. Similar reports by Polyanski (1961) also supported the view that intermediate-length group hosts had a higher prevalence than those of smaller and larger-length groups. Oniye and Aken'Ova (1999) stated that the high prevalence in lengthy fishes could be attributed to the longer time of exposure to the environment by body size. The high prevalence in lengthy and bigger fishes may be due to the bigger internal organs of the hosts leading to the increase in the surface areas of infection (El-Naggar and Khidr, 1986; Khidr, 1990; Hagraas et al., 1995; Muzzall et al., 1990). Bashirullah (1973) and Dogiel (1961) reported that the degree of parasitism was related to the food habits and age of the fish.

The fishes belonging to intermediate weight groups are highly infected with the helminth parasites showing the highest infection prevalence as compared to the smaller and larger weight groups of fishes. Kumari (2018) in her study found that the mid-weighted snakeheads were more prone to infection as compared to lower and higher-weight groups. Polyanski (1961) supported the view that intermediate-weight group hosts had a higher prevalence than those of smaller and larger weight groups.

Several scientific studies reflect that the sex of host fishes has a detrimental effect on the parasitological presence. (Smith, 1969; Sanwal and Agarwal, 1974; Sinha, 1984; Rajaiah, 1977). During our study, we also found that the infestation prevalence based on sex is higher in female fishes as compared to male fishes. Thomas (1964), Chandra (1985), Khanum and Parveen (1997), Khanum et al. (2008), Rahman and Saidin (2011), and Holden and Reed (1972) also reported that the infection rates of helminth parasites were observed higher in female host fishes and suggested that this may be due to lower physiological resistance of the female fishes. Omeji et al. (2013), Emere and Egbe (2006), and Ayanda (2009) reported higher parasitic prevalence in female *C. gariepinus* and suggested that during breeding season increased rate of food intake by female fish to meet their requirements for the development of their egg might have exposed them to more contact with the parasites, which subsequently increased their chance of being infected.

The parasites collected from different body parts of fishes belonged to three major classes of helminth parasites i.e. Nematoda, Trematoda, and Cestoda. The *Truttaedacnitis sandeepi n.sp.* a nematode parasites was collected from the intestine of *Rita rita* fish while the *Opisthorchis sp.* a trematode parasite was collected from the gallbladder of *Rita rita*. *Gangesia agragensis* a cestode parasites were found in the intestine of *Wallago attu*. The alimentary canal of the host was found to be highly infected with helminth parasites, followed by the gallbladder.

The parasite prevalence was higher in catfishes obtained from the Pipra Ghat sampling site than at Kudiya Ghat of Gomti River Lucknow. The reason behind this may be attributed to the current pollution status of Pipra Ghat is found to be very high owing to its higher Biological Oxygen Demands (BOD) and lower Dissolved Oxygen (DO) levels as revealed in the present study, which stresses fish and increases parasitological infection rates, also as corroborated by Ali et al. (2014). Environmental stimuli can affect parasites in various ways, and parasites respond to them in sophisticated ways (Lafferty and Kuris, 1999). Fish are typically more vulnerable to

parasite infection in unstable environmental circumstances and contaminated water (Svobodova and Kolarova, 2004).

Many ecological factors can have an impact on the parasite population dynamics in fish living in freshwater or riverine environments. It is essential to comprehend these dynamics to evaluate the condition of the riverine ecosystem as a whole as well as the fish populations. With their varied water quality, temperatures, and nutrient levels, rivers offer a special kind of habitat. The abundance and distribution of parasites can be impacted by modifications to these environmental factors. The effect of different physiochemical water parameters {(pH, Temperature (°c), Total hardness (mg/l), Alkalinity (mg/l), Dissolved oxygen (mg/l) and Biochemical Oxygen demand (mg/l)} on the parasitic infection prevalence of host fishes, *Rita rita* and *Wallago attu* was evaluated by Backward elimination method of Multiple Linear Regression (MLR) model for the period of July 2021 to June 2023

For the period of July 2021 to June 2022, Total hardness (TH), Alkalinity (ALK) and Dissolved oxygen (DO) emerges as the most effective hydrobiological parameter in predicting the prevalence of *Truttaedacnitis sandeepi* n.sp in male *Rita rita* fish at the site Pipra ghat whereas alkalinity (ALK) serve as important water physiochemical water parameter in predicting the prevalence of *Truttaedacnitis sandeepi* n.sp in female *Rita rita* fish at the site Kudiya ghat.

At the site Kudiya ghat the water temperature and Biological Oxygen Demand (BOD) show a significant impact on the infection prevalence of *Opisthorchis* sp. in male and female *Rita rita* fish respectively whereas total hardness (TH) serves as an only water physiochemical parameter that helps in dictating the infection prevalence of *Opisthorchis* sp. of female *Rita rita* fish at Pipra ghat for the investigation period of July 2021 to June 2022.

For the period of July 2021 to June 2022, Total hardness (TH) and alkalinity (ALK) serve as an important water physiochemical parameter in predicting the infection prevalence of *Gangesia agransis* from male *Wallago attu* at site Pipra ghat. Moreover at the same site only total hardness (TH) shows a significant effect on infection prevalence of *Gangesia agransis* from female *Wallago attu*.

For the period of July 2022 to June 2023, dissolved oxygen (DO) and biological oxygen demand (BOD) emerge as the most effective hydrobiological parameter in predicting the infection

prevalence of *Truttaedacnitis sandeepi* n.sp in male *Rita rita* fish whereas the water temperature, total hardness (TH), alkalinity (ALK) serve as an specific hydrobiological parameter to significantly impact the infection prevalence of *Truttaedacnitis sandeepi* n.sp in female *Rita rita* fish at site Kudiya ghat. Moreover at the site Pipra ghat water temperature effect the infection prevalence of *Truttaedacnitis sandeepi* n.sp in male *Rita rita* fish but alkalinity (ALK) and dissolved oxygen (DO) were found to affect the infection prevalence of *Truttaedacnitis sandeepi* n.sp in female *Rita rita* fish.

The infection prevalence of *Opisthorchis* sp. in female *Rita rita* fish was found to be significantly affected by water temperature at both the sites for the period of July 2022 to June 2023.

For the duration of July 2022 to June 2023 infection prevalence of *Gangesia agragensis* in male *Wallago attu* the water temperature and dissolved oxygen (DO) was found to be significantly affecting the infection prevalence at the site Kudiya ghat whereas water temperature, alkalinity (ALK), dissolved oxygen (DO), and biological oxygen demand (BOD) are important hydrobiological parameters in predicting the infection prevalence of *Gangesia agragensis* from female *Wallago attu* at the site Kudiya ghat. Moreover, total hardness (TH) emerges as the most effective water hydrobiological parameter in predicting the infection prevalence of *Gangesia agragensis* from female *Wallago attu* at the site Pipra ghat.

The current study was conducted just after Covid 19 Lockdown. During the course of study for two consecutive years i.e. from July 2021 to June 2023 the infection prevalence was observed lower for the duration of July 2021 to June 2022 as compared to the duration of July 2022 to June 2023. The reason behind the lower infection indices of parasites in host fish *Rita rita* and *Wallago attu* for the duration of July 2021 to June 2022 could be attributed to the duration of Covid 19 lockdown during which all the economic as well as all the anthropogenic activities were completely prohibited which creates a positive impact on the health of aquatic ecosystem so the ichthyfaunal health was improvised and prevalence of helminth parasites was comparatively less. But after removing lockdown from government of India all the economic and anthropogenic activities were resumed which created negative impact on the health of aquatic ecosystem resulting in the deterioration of fish health and immune system which led to the higher parasitological infection indices for the period of July 2022 to June 2023.