

**Studies on Gastrointestinal Helminths in the Snow Trouts
(Schizothoracids) as Indicators of Aquatic Pollution in
Kashmir**

Asifa Wali
(2012-441-D)



Faculty of Fisheries

**Sher-e-Kashmir University of Agricultural Sciences &
Technology of Kashmir**

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**Studies on Gastrointestinal Helminths in the Snow Trouts
(Schizothoracids) as Indicators of Aquatic Pollution in
Kashmir**

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Thesis

Submitted to

Faculty of Fisheries

**Sher-e-Kashmir University of Agricultural Sciences &
Technology of Kashmir**

in partial fulfilment of requirement for the award of the degree of

**Doctor of Philosophy in
Fisheries Resource Management**

2015



Dedicated

To my

Parents

Sher-e-Kashmir
University of Agricultural Sciences & Technology of Kashmir
Faculty of Fisheries, Rangil, Ganderbal

Certificate – I

This is to certify that the thesis entitled, “**Studies on Gastrointestinal Helminths in the Snow Trouts (Schizothoracids) as Indicators of Aquatic Pollution in Kashmir**” submitted in partial fulfilment of the requirements for the award of the degree of **Doctor of Philosophy in Fisheries Resource Management**, to the **Faculty of Fisheries, Sher-e-Kashmir University of Agricultural Sciences & Technology of Kashmir** is a record of bonafide research work carried out by **Ms. Asifa Wali (Regd. No. 2012-441-D)** under my supervision and guidance. No part of the thesis has been submitted for any other degree or diploma.

It is further certified that information received during the course of investigation has duly been acknowledged.

(Dr. M.H. Balkhi)
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This is to certify that the thesis entitled, “**Studies on Gastrointestinal Helminths in the Snow Trouts (Schizothoracids) as Indicators of Aquatic Pollution in Kashmir**” submitted by Ms. **Asifa Wali (Regd. No. 2012-441-D)** to the **Faculty of Fisheries, Sher-e-Kashmir University of Agricultural Sciences & Technology of Kashmir** in partial fulfilment of the requirements for the award of the degree of **Doctor of Philosophy in Fisheries Resource Management** was examined and approved by the Advisory Committee and External Examiner on

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Title of the Thesis : **Studies on Gastrointestinal Helminths in the Snow Trouts (Schizothoracids) as Indicators of Aquatic Pollution in Kashmir**

ABSTRACT

The present study was undertaken to determine the incidence of helminth parasites in fishes with special reference to water quality parameters, histopathological changes and the concentration of various metals in water, fish tissue and helminth parasites in Dal Lake and River Jhelum and correlate the observations. Water, fish and parasite samples were collected during different seasons from various sites and processed. Three fish species (*Schizothorax niger*, *Schizothorax esocinus*, *Schizothorax curvifrons*) were recovered from these water bodies. The physicochemical parameters Temperature, Dissolved oxygen, pH and free carbon dioxide showed variation vis-a-vis the season and location of the stations in water bodies. Highest temperature in Dal Lake was seen at Dalgate during summer (27.22 ± 1.71 °C) while the lowest temperature was also recorded at the same site during winter (6.55 ± 1.48 °C). Similarly, the highest temperature (19.44 ± 1.50) in Jhelum River was recorded at Chattabal weir while lowest (4.38 ± 1.32) was recorded at Khannabal. Besides, the temperature of water in Jhelum River was at lower degree than that of Dal Lake. pH of the waters of Dal Lake and River Jhelum was within the alkaline range. The pH of Jhelum river water was comparatively less alkaline than that of Dal Lake. In Dal Lake DO was highest at Telbal (6.78 ± 1.25 mg/l) during spring while lowest was at Hazratbal (3.55 ± 1.19 mg/l) during summer. In River Jhelum DO was highest at Khannabal

(8.16±1.43 mg/l) during spring while lowest was at Zero bridge (4.97±1.50 mg/l) during autumn. DO of River Jhelum was noted at higher level in all the seasons compared to that in Dal Lake. Free carbon dioxide in Dal Lake was highest (12.88 ± 2.21 mg/l) at Telbal during spring while the lowest was found at Saidakadal (3.16±0.93 mg/l) during autumn. In River Jhelum highest free CO₂ (8.11±2.26 mg/l) was found at Chattabal Weir during summer and lowest was also recorded (3.08±1.83 mg/l) at the same site during autumn. Free carbon dioxide was found higher in Dal Lake than in River Jhelum in all the seasons of the year. Acanthocephalan parasite *Pomphorhynchus kashmirensis* (27.47%) and two intestinal cestodes *Bothriocephalus acheilognathi* (30.63%) and *Adenoscolex oreini* (32.43%) were recovered from all the three species of *Schizothorax*. All the three parasites showed higher prevalence during summer and least during winter. Overall the mean intensity of *Pomphorhynchus kashmirensis* was highest in Dal Lake whereas its abundance was higher in River Jhelum. For both the cestodes mean intensity and abundance were higher in River Jhelum. All the three parasitic infections were prevalent more in male fishes compared to females. The mean intensity and abundance of both the cestodes was highest in summer and lowest in winter. The presence of the parasites had reduced the condition coefficient of the infected fishes in both the water bodies. Histopathologically, all the three parasites induced various intensities of enteritis coupled with hyperplastic goblet cells with increased acid mucopolysaccharide concentrations. The overall concentrations of different heavy metals in the two water bodies showed significant variations both season and location wise. In Dal Lake the concentrations of Co, Ni, Mn, Cr, Fe and Cd were higher than the permissible limits. Similarly, in River Jhelum the concentrations of Ni, Mn, Cr, Fe, Cd in water were higher than the permissible limits. Pb and Hg were below detection levels in both the water bodies while Cu and Zn were recorded only in Dal Lake and River Jhelum, respectively. In Dal Lake the ranking of mean concentration of 12 metals at Dalgate, Saidakadal, Hazratbal and Telbal were Fe > Ca > Mn > Al > Zn > Cd > Cu > Ni > Co > Cr; Fe > Mn > Ca > Al > Zn > Cd > Ni > Co > Cr > Cu; Al > Fe > Ca > Mn > Zn > Cd > Ni > Co > Cr > Cu and Fe > Al > Ca > Mn > Cu > Ni > Co > Zn > Cd > Cr respectively. In Jhelum river the ranking of the metals at Khanabal, Zero bridge, and Chattabal weir were Fe > Ca > Al > Mn > Zn > Co > Cd > Cu > Cr > Ni; Ca > Fe > Al > Mn > Zn > Ni > Cu > Co > Cd > Cr; and Fe > Mn > Ca > Al > Co > Zn > Cd > Ni > Cu > Cr respectively. In Dal lake the *Pomphorhynchus kashmirensis*; *S. niger* liver and muscle and the ranking of the 12 metals were Ca > Fe > Mn > Cr > Cu > Ni > Zn > Al > Cd > Co, Ca > Fe > Zn > Al > Cu > Mn > Cr > Ni > Co > Cd and Cd > Ca > Cr > Zn > Cu > Mn > Al > Ni > Co > Cd respectively. In river Jhelum it were Ca > Fe > Mn > Cr > Al > Cu > Ni > Zn > Co > Cd; Ca > Fe > Zn > Al > Mn > Ni > Cu > Co > Cr > Cd and Zn > Fe > Ca > Cu > Cr > Al > Mn > Co > Ni > Cd respectively. In Dal lake the *Bothriocephalus acheilognathi*; *S. niger* liver and muscle and the ranking of the 12 metals were Fe > Ca > Al > Zn > Mn > Cr > Cu > Ni > Cd > Co; Fe > Ca > Cu > Zn > Al > Mn > Ni > Cd > Cr > Co and Fe > Ca > Zn > Al > Cr > Ni > Cu > Mn > Cd > Co respectively. In river Jhelum it was Fe > Ca > Zn > Al > Cu >

Mn>Cr> Ni> Co> Cd; Fe> Ca> Zn> Cu> Al> Mn> Ni> Cr> Cd> Co and Ca> Fe> Zn> Al> Cr> Mn> Ni> Cu> Co> Cd respectively. In Dal lake the *Adenoscolex oreini*; *S. niger* liver and muscle and the ranking of the 12 metals were Ca> Zn> Fe> Mn> Al> Cu> Co> Ni> Cd> Cr; Fe> Ca> Mn> Al> Cu> Zn> Cd> Ni> Co> Cr and Fe> Ca> Al> Mn> Zn> Cu> Ni> Cd> Cr> Co respectively. In river Jhelum it were Ca> Fe> Cr> Mn> Al> Zn>Cu> Cd> Ni> Co; Ca> Fe> Al> Zn> Cu> Mn> Ni> Co> Cd> Cr and Ca> Fe> Al> Zn> Cu> Cr> Ni> Mn> Co> Cd respectively. In Dal Lake the concentrations of copper (1.25 µg/g), zinc (5.69 µg/g) and iron (29.3 µg/g) were maximum in the muscles of infected fishes which were still lower than the Maximum Limit recommended by WHO/FEPA/IAEA-407 whereas on the other the concentration of nickel (0.80 µg/g), manganese (1.69 µg/g), cadmium (0.78 µg/g), chromium (2.86 µg/g) and aluminium (2.99 µg/g) were higher than the permissible limits. In River Jhelum the maximum concentration of nickel (0.80 µg/g), manganese (0.41µg/g), aluminium (2.99 µg/g), cadmium (0.19 µg/g) and chromium (0.82µg/g) in the muscles of infected fishes were higher than the permissible limits. Whereas on the other the concentration of copper (0.86 µg/g), zinc (3.93 µg/g), iron (24.4 µg/g) in the muscles of infected fishes were still lower than the Maximum Limit recommended by WHO. Cr, Mn, Cu, Zn, Al, Ca concentrations in water showed positive correlation with water temperature. Cu concentration in *Pomphorhynchus kashmirensis* and *Adenoscolex oreini* showed positive correlation with water temperature. Cr concentration in *Pomphorhynchus kashmirensis* showed positive correlation with water temperature. Fe, Zn, Cd Cu and Co concentrations in water showed positive correlation with Free CO₂. Cu and Cr in *Pomphorhynchus kashmirensis* showed positive correlation with pH of water. Zn and Cr in *Bothriocephalus acheilognathi* showed positive correlation with pH of water. Cd concentration in *Adenoscolex* showed positive correlation with pH of water. Zn concentration of water and Fe concentration in *Bothriocephalus acheilognathi* was positively correlated with DO. However, the two parasites which have relation directly with water are *Adenoscolex oreini* and *Pomphorhynchus kashmirensis*. *Adenoscolex oreini* was positively correlated with Cd concentration in water during autumn and winter whereas *Pomphorhynchus kashmirensis* was positively correlated with Al and Cr concentration in water. Therefore, the two parasites seem to act as bio-indicators for the corresponding metals.

Key words: Dal Lake; River Jhelum; *Schizothorax* spp; Helminth; parasites; physicochemical parameters; Histopathology; Histochemistry; Heavy metals

Signature of Student

Signature of Major Advisor

Dated _____

Dated _____

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Asifa Wali

Place:

Dated: _____

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LIST OF ABBREVIATIONS

µg/l	:	Microgram per liter
AAS	:	Atomic absorption spectrophotometer
ANOVA	:	Analysis of variance
APHA	:	American Public Health Organisation
BDL	:	Below detection limit
C ₂ H ₂	:	Air- acetylene
CF	:	Condition factor
H ₂ O ₂	:	Hydrogen peroxide
HCL	:	Hydrochloric acid
HNO ₃	:	Nitric acid
mg/l	:	Milligram per litre
°C	:	Degree Celsius
pH	:	Hydrogen ion concentration
PPE	:	Personal protective equipment
Ppm	:	Parts per million
WHO	:	World health organization
Ww	:	Wet weight
Cu	:	Copper
Zn	:	Zinc
Co	:	Cobalt
Ni	:	Nickel
Mn	:	Manganese
Cr	:	Chromium
Al	:	Aluminium
Fe	:	Iron
Ca	:	Calcium
Cd	:	Cadmium

Chapter –1

INTRODUCTION

Parasites are an essential part of the aquatic environment and represent a significant proportion of aquatic biomass. Parasites occur in nearly every population. They often interact in complex ways with other stressors. In some cases, the interaction may lead to a disproportionately negative effect on the host population. In other cases, the stressor may ameliorate the effect of parasitism. Parasites are attracting increasing interest from parasite ecologists as potential indicators of environmental quality due to the variety of ways in which they respond to anthropogenic pollution. In environmental impact studies certain organisms provide valuable information about the chemical state of their environment not through their presence or absence but instead through their ability to concentrate environmental toxins within their tissues. Certain parasites, particularly intestinal acanthocephalans of fish can accumulate heavy metals to concentration order of magnitude higher than those in the host tissues or the environment (Sures, 2001).

Since parasites are sensitive to environmental change, others are more resistant than their hosts and tend to increase in numbers in polluted conditions (Sures, 2004); they are now regarded as a useful indicator of aquatic health. The relationship between environmental pollution and parasitism in aquatic organisms and the potential role of endoparasites as water quality indicators have received increasing attention during the past two decades (Sures, 2003, 2004). The ability of endoparasites to detect even lowest metal concentration due to their enormous accumulation capacity has made them suitable water pollution biomonitors than their fish hosts (Elijah *et al.*, 2010). Recently Sures (2007) indicated that parasites can also act as metal sinks for its fish host. It is apparent that any changes to metal concentration in the fish tissues are likely to alter metal bioavailability and toxicity.

Fishes are good source of quality protein, but various diseases including parasitic infections pose a threat to fish culture (Yooyen *et al.*, 2006). Helminth parasites are hazardous to a number of fish species in different water bodies. As much as 30,000 helminth species are parasites of fish (Williams and Jones, 1994). Each helminth parasite species prefer to live in a definite zone of the microhabitats, though some can migrate to the other organs, which are normally not their usual site of infection. Parasites interfere with nutrition, metabolism and secretory function of alimentary canal, damage nervous system (Markov, 1961) and even upset the normal reproduction of the fish (Faust, 1949). Extensive damage caused by helminth parasites on fish organs indirectly affects its growth, development and reproduction and thus may lead to further decline in the population of the host fish and reduction in production of various culture systems. Heavy infestation of endoparasites interrupts the normal growth of fish. Injured fishes carry heavy parasitic infection which deteriorates their food and that may be the ultimate cause of their mortality (Gupta, 1983). Intestinal parasites inhibit digestive activity inducing inhibition of vitamin and blood sugar metabolism and finally growth as well as glycogen metabolism by infecting the liver of the host fish (Rohde, 1993). Frequency of infestation and distribution of parasites within different organs of the fishes is influenced by age and diet of the fish and abundance of parasites within the host fish (Snieszko, 1983).

Intestinal helminths of fish are of increasing interest as potential bioindicators for heavy metal contamination in aquatic habitats. Among these parasites cestodes and acanthocephalans in particular have an enormous heavy metal accumulation capacity exceeding that of established free living sentinels. Metal concentrations several thousand times higher in acanthocephalans than in host tissues has been described from field and laboratory studies. Whereas larval stages inside their intermediate hosts are not able to take up high quantities of metals, young worms begin to take up metals immediately after infection of the final host. After 4-5 weeks of exposure, the parasites reach a steady-state

concentration and order of magnitude higher than the ambient water level. Thus, acanthocephalans are not only very effective in taking up metals, but they can also respond very rapidly to changes in environmental exposure. The mechanism which enables acanthocephalans to take up metals from the intestinal lumen of the host appears to be based on the presence of bile acids, which form organo-metallic complexes that are easily absorbed by the worms due to their lipophilicity (Sures, 2001). Investigations of the environmental conditions affecting metal uptake have shown that the parasites are more consistent and reliable indicators for metal pollution than the host tissues as metal levels of the latter are much more dependent on the water chemistry.

Fish harbor a variety of parasites viz., protozoans, cestodes, trematodes and acanthocephalans (Ali, 1990) and the degree of damage by infection is influenced to a large extent by the type and numbers of parasites present (Bauer, 1941). The distribution of parasites varies not only in different species of fish but also seasonally and from one water body to other. The pathogenicity of parasitism has been reported to cause extensive damage to the host leading to the lower production of the fish (Rai, 1986). In certain studies the parasite has been found to be responsible for the death of the host (Bookmer *et al.*, 1981). The relationship of parasitism and pollution is not simple and involves a double edged phenomenon in which parasitism may increase host susceptibility to toxic pollutants or pollutants may result in an increase or decrease in the prevalence of certain parasites. Pollutants may affect an intermediate or alternate hosts in parasite life cycle and on free-living life cycle stages of parasite invasion (Sindermann, 1990). Pollution stress can influence the prevalence of parasites directly or indirectly, or the parasite infestation may decrease the host resistance to toxic pollutants (Khan and Thulin 1991). Recently, various sentinel organisms such as bivalves (Gunkel, 1994) have been implemented for assessing levels of pollution in aquatic habitats. However, in the last two decades fish parasites attained increasing attention, as they appear to be a more precise tool for detecting metal loads in aquatic habitats.

Due to their enormous accumulation capacity, especially for some elements with severe toxic effects on biota (e.g. cadmium, lead), fish acanthocephalans are good candidates as bioaccumulation indicators. In a number of studies concentrations of metals were reported to be 10^2 to 10^5 times higher in the parasite than in the water column and the sediment (Sures *et al.*, 1994; Schludermann *et al.*, 2003; Thielen *et al.*, 2004).

The permanent contamination of aquatic habitats caused by human activities has become one of the major problems in the era of global industrialization and urbanization. Mostly, chemical pollution and in particular contamination with heavy metals is considered to have an anthropogenic source. However, natural geogenic deposition might be an important factor for heavy metal pollution as well. Thus, the detection and management of heavy metal loads in the aquatic environment is very important from the ecological point of view. Moreover, various aquatic organisms are used for human consumption and for this reason knowledge about metal contamination is extremely important from the public health view point.

A wide range of contaminants are continuously introduced into the aquatic environment mainly due to increased industrialization, technological development, growing human population, oil exploration and exploitation, agricultural and domestic waste run-off (Lima *et al.*, 2008). Among these contaminants, heavy metals constitute one of the most dangerous groups because of their persistent nature, toxicity, tendency to accumulate in organisms and undergoing food chain amplifications and being non-degradable (Fufeyin and Egborge, 1998). When released into the environment heavy metals find their way into the aquatic systems and are deposited in aquatic organisms like fishes through the effects of bioconcentration, bioaccumulation and the food chain process and eventually threaten the health of humans that consume them. Heavy metals concentrations in aquatic ecosystems are usually monitored by measuring their concentrations in water, sediments and biota (Oguzie, 2003). The present

study was conducted on River Jhelum and Dal Lake, Srinagar. Dal Lake is one of the most beautiful urban lakes of Kashmir. During the past few years, concern is being voiced by people over the deteriorating conditions of Dal Lake which has fallen victim to human greed as a result of which the entire ecosystem is rapidly changing (Pandit, 1993). The water quality of Lake has deteriorated considerably in the last three decades due to eutrophication, illegal encroachments, dumping of organic matter, sewage and other pollutants. Another water source River Jhelum originates from a famous spring called Verinag in Anantnag district. The water bodies serve as an important source of indigenous as well as exotic fishes. Increasing agro-chemical pollution of River Jhelum has become a matter of great concern. Pesticides are used in agricultural fields and enter the river through land drainage or with surface run-off during floods and excessive rains. These include herbicides, rodenticides, fungicides, wormicides, insecticides (like chlorinated hydrocarbons, organophosphates, carbamates, phenols) etc. These are lethal to fish if they exceed the tolerable limits.

In order to use endoparasites as bioindicators some requirements have to be met, as suggested by Kennedy (1997): most importantly, the fish host should be abundant and easy to be sampled and secondly, the parasites must be highly abundant and prevalent among the host population. The water bodies of Kashmir valley support a wide variety of indigenous and exotic fish species. The major ichthyofauna of Kashmir is represented mainly by the Central Asiatic fauna in which *Schizothorax* group is predominant (Sunder *et al.*, 1979). The Cyprinidae are mainly represented by the Schizothoracines (Snow trouts). A comprehensive survey to assess the potential of Schizothoracines from various lakes and streams of Kashmir valley was made by Raina *et al.* (1985). These are indigenous fish species and are commercially important with wide market demand. Most species of the fishes present are exploited in one way or the other and are facing immense pressure due to pollution and human interferences. Schizothoracines inhabit both lentic as well as lotic water bodies of Kashmir.

Lot of work has been done on various aspects of host-parasite interaction in fishes but very little has been studied regarding helminth parasites as bio-indicators of pollution. Hence a comprehensive work was undertaken to study the gastrointestinal helminths in snow trouts as indicators of aquatic pollution with special reference to metal pollutants in Dal Lake and River Jhelum.

The objectives of the present study were as under:

- To study the incidence of helminth parasites in fishes with special reference to water quality parameters.
- To assess the concentration of various metals in water, fish tissue and helminth parasites of the fishes in Dal Lake and River Jhelum.
- To determine the correlation among metal levels of water, fish tissues, parasites and physicochemical parameters.
- To determine the histopathological changes caused by the residues of these metals in the liver and intestines effected by different parasites.

Chapter -2

REVIEW OF LITERATURE

2.1 Physico-chemical features

Global fresh water is the most precious human resource. Frequently Earth is called the blue planet because water covers about 75% of the globe, but most of the water is saline, less than 5% of the water is fresh (Herdendorf, 1990), and much of this water is in the ice caps, glaciers and ground water. Most of the remainder is in lakes, streams and soil moisture, and an estimated 68% of the fresh liquid surface water is in 189 large lakes with surface areas greater than 500 km² (Reid and Beeton, 1992). The maintenance of water quality standards in lakes and reservoirs is necessary in order to avoid excessive growth of aquatic flora which is a problem to aquatic biota and humans. In many tropical lakes, temporal and spatial changes in the physico-chemical parameters are common in response to surface water runoff, direct precipitation, ground water recharge, rate of evaporation and human interference. These changes have impacts on the flora and fauna by imposing physiological and behavioral adaptations (Kemdirim, 2005).

George *et al.* (1966) in a limnological survey of the river Kali opined that with pH range 7.0 to 9.0 the bicarbonate concentration of water remained high and that these changes were directly related to the photosynthetic activities in water bodies. However, Zutshi and Vass (1978), while studying the chemical features of Dal Lake, reported that the water, though not rich in nutrients, was alkaline and slightly buffered (pH, 7.4-9.6) and oxygen of surface waters ranged from 7.2-17.6 mg.

Zutshi and Khan (1988) in an eutrophication level of Dal Lake studied presence of high chloride in the water was attributed to the urination and bathing activities of people on the banks of the Dal Lake.

Crucil *et al.* (1991) while investigating the chloride concentration in Lake Erie reported an overall rise from approximately 7 mg/l to 35-45 mg/l in the late

1960's. However, due to cessation of the production of the soda ash by five of the six major industrial point sources along the Detroit river in the late 1960's (which accounted for 94% of the inflow to lake Erie), a considerable decrease in the concentration of chloride from 35-45 to 15-20 mg/l was observed.

Dilshada (1995) reported the pH range of water between 7.6 to 9.3, dissolved oxygen between 10.1 to 16.8 mg/l, free carbon dioxide between 10-109 mg/l and the chlorides between 30.8 and 61 mg/l while working on the water quality of Dal Lake. The highest concentration of dissolved oxygen and free carbon dioxide was found in winter season.

Kolo (1999) assessed some physico-chemical parameters of the surface water of Shiroo Lake over a period of eighteen months reported that the wet season mean values for water and air temperatures were significantly ($P < 0.05$) higher than dry season mean values at all the stations. However, mean values for pH, Dissolved oxygen and Phosphate-phosphorus was higher in the dry season than wet season mean values.

Devi and Sharma (2004) studied the seasonal variations in some important physico-chemical characteristics of the freshwater ponds of Manipur, reported low concentration of dissolved oxygen which was due to indiscriminate use of the ponds by the local people.

Kundangar and Abubakr (2004) made a comparison of the water quality changes in Dal Lake over a period of three decades in its different basins and found out that the pH and dissolved oxygen content of Hazratbal basin was 7.7-9.5 (1977) Vs 7.1-8.6 (in 2000) and 2.2-12 mg/l (in 1977) Vs 3.6-9.4 mg/l (in 2000), respectively. Similarly, for Nishat basin it was 7.4-9.5 (in 1977) Vs 7.3-8.8 (in 2000) and 5.5-11.5 mg/l (in 1977) Vs 5.2-9.8 mg/l (in 2000) respectively. For Nehru park basin, 7.5-9.5 (in 1977) Vs 7.6-9.2 (in 2000) and 4.5-10.5 mg/l (in 1977) Vs 2.0-9.2 mg/l (in 2000) and 0.8-10 mg/l (in 1977) Vs 3.2-10 mg/l (in 2000), respectively.

Kumar *et al.* (2004) reported the bad effect of the process of pollution on lentic and lotic ecosystem in Jharkhand depicted by the physico-chemical parameters viz, temperature, turbidity, total dissolved solids, pH, dissolved oxygen, free carbon dioxide, carbonates alkalinity, total hardness, chloride, phosphate, nitrate, nitrogen and BOD.

Qadri and Yousuf (2004) studied the physico-chemical parameters of Nigeen Lake and found that the pH of the lake ranged from 7.05-7.67, suggesting the alkaline nature of the water body. The dissolved oxygen, free carbon dioxide and chloride values were recorded as 6.0-7.8, 18.6-36.5 and 11.9-13.0 mg/l respectively.

Zaaware *et al.*(2004) screened Pashan Lake (Pune) for different physicochemical parameters of water during the year 2001-2002. The result revealed that the water temperature varied from 16 to 36^oc and pH ranged between 6.5 and 7.4. A marked variation in total alkalinity was observed during summer season. The dissolved oxygen content was high during summer season. Nitrate level varied from 1.02 to 2.532 mg/l. The lowest concentration of nitrate, phosphate, chloride, carbonate, sulphate, calcium and magnesium were recorded during the pre-monsoon seasons. Their results further revealed that the physicochemical parameters of lake water significantly influenced the algae.

Njenga (2005) studied the water chemistry of four tropical lakes, three rift valley lakes (Nakuru, Elementaita and Naivasha) in Kenya and one lake in southern India (Kolleru), and reported that waters of Lakes Nakuru and Elementaita were highly alkaline in nature compared to waters of lake Naivsha and Kolleru.

Mohan *et al.* (2005) while carrying out studies on the microbial and phsico-chemical parameters of a Talab in Jodhpur, Rajasthan recorded pH that ranged from 7.2 to 9.2. The higher pH values of the Talab were attributed to the sewage discharged from surrounding human populations. The free carbon dioxide

in the Talab waters varied between 0-57 ppm, while the dissolved oxygen content varied from 1.2-3.7 mg/l. The waters of the Talab had a high prevalence of *Escherichia coli* especially in post monsoon months.

Mustapha and Omotosho (2005) studied the impact of human activities on the quality and productivity of the lake waters and attributed the lower concentrations of the dissolved oxygen to the human activities such as sewage disposal in the lakes while the higher carbon dioxide values were attributed to the decomposition of these wastes by micro-organisms

Sharma *et al.*(2005) studied the eutrophication process in urban lake system of Udaipur (Rajasthan) and found that the pH of the lake water ranged from 8.35 to 9.30 while the dissolved oxygen level was found to vary between 0.5 and 7.8 mg/l.

Zafar *et al.* (2005) while conducting study on the water quality of Dal Lake reported that the dissolved oxygen concentration of the Dal Lake basin was 7.2 to 11.6 mg/l.

Jeelani and Shah (2006) while analyzing 240 samples (in four seasons) and 17 sediment samples to monitor the natural and anthropogenic influences on the water and sediment chemistry of the Dal Lake reported predominance of carbonate and silicate weathering. Lower pH and high total dissolved solids, electrical conductivity and NO₃ values in the Gagribal basin and in some patches of other basins reflected the anthropogenic inputs in the form of sewage from surrounding population, houseboats, hotels etc. They further reported that the Dal Lake was characterized by chemical index of alteration (CIA: 87-95) reflecting extreme weathering of the catchment area.

Siraj *et al.* (2006) while studying the physico-chemical parameters of Dal Lake, Kashmir revealed that the water near the floating gardens was more enriched with nutrients due to increased level of chloride, phosphorus, nitrogen and conductivity.

Akhter (2007) reported the annual mean pH in Dal Lake water to be 7.78 and an annual chloride content to be 14.25 mg/l.

Adeyemo *et al.*(2008) carried a field study (October 2003 and March 2004) to develop a data-base on seasonal changes of physico-chemical parameters and nutrient load of the river sediment in Ibadan metropolis. All physico-chemical parameters (except dissolved oxygen) were higher than the recommended standard. The levels of these parameters in downstream were significantly elevated than the corresponding levels upstream. Their result suggested that the water quality of Ibadan river system was adversely affected and impaired by the discharge of domestic, agricultural and industrial wastes which was the usual practice in Ibadan.

Mathur *et al.* (2008) while studying the physico-chemical characteristics of Pushkar Lake in Ajmer, attributed eutrophication, anthropogenic pressures, holy rituals and tourism as the major Contributing factors for the damage, deterioration and degradation of the lake water quality. Their study further revealed higher concentration of chloride and conductivity with greater pollution levels as indicated by lower concentrations of Dissolved Oxygen.

Hussain and pandit (2008) studied some physico-chemical features of a lake Nilnag situated in the foot hills of Pir Panchal range of Himalayan mountains and observed highly significant differences in parameters like pH, conductivity, acid neutralizing capacity, calcium, sodium, NH₄-N, NO₃-N, soluble reactive phosphorus, turbidity, dissolved organic carbon, sulphate, dissolved silica and total iron of littoral water of lake Nilnag toward forest side and agriculture side. They further reported that the littoral water chemistry was a good indicator of the catchment land use.

Ibrahim *et al.* (2008) investigated the effect of environmental conditions of Abu-Zabal Lake on fish caught during March, 2005 till February, 2006. The study included biological, histological and quality aspects coupled with physic-

chemical, microbial and sensory evaluation. Results showed that the concentrations of metals (Cu, Zn, Cd and Pb) in water were higher during both summer and winter. Muscles of fish catches of spring and summer were more affected by heavy metals than during autumn and winter. Although the detected concentration of heavy metals was below the permissible levels, however, detectable histological changes in the gills, testis and muscles were observed.

Koria *et al.* (2008) studied the physico-chemical properties and biota of Keenjhar Lake (Karachi, Pakistan) from January until December 2005. A total of 142 species of phytoplankton, 37 species of zooplankton, 39 species of aquatic plants, 51 species of fishes and 8 species of Prawns were recorded. The physicochemical properties such as temperature, alkalinity, dissolved oxygen, salinity, conductivity, total dissolved solids, chlorides, turbidity, pH and hardness were determined on monthly basis and it was concluded that water of Keenjhar Lake was suitable for growth of aquatic biota.

Singh *et al.* (2008) assessed the water quality and eutrophication status of various lakes situated in the western Himalayas part of India. These workers found very distinct characteristics in Tsomoriri and Tsokhar lakes prevailing in the Ladakh region due to cold desert type climate and having a very low rainfall. Unusually higher values of certain water quality parameters, viz, pH, total dissolved solids, total hardness, chloride, calcium and magnesium were recorded in these two lakes. The eutrophication status in all the lakes was assessed by using phosphate data and it was inferred by the workers that Mansar, Surinsar and Tsomoriri were under eutrophic and Dal, Tsokar and Renuka lakes were hyper-eutrophic condition.

Bushati *et al.* (2010) studied physico-chemical parameters of Shkodra Lake water (situated on the border between Montenegro and Albania). The temperature of the water ranged from 14.1-28.2°C and pH from 8.1 to 8.5. The higher pH was attributed to the presence of carbonates and bicarbonates in high concentration in the lake. The results showed that the Shkodra Lake was a favored

environment for the living flora and fauna. The lower dissolved oxygen concentrations were observed in summer when the temperature of the water was the highest. The average values of $\text{NH}_4\text{-N}$ ranged from 0.025-0.05 mg/l, and the levels of $\text{NO}_3\text{-N}$ ranged from 0.025-0.05 mg/l and the levels of $\text{NO}_3\text{-N}$ ranged from 0.18-0.27 mg/l.

Murtaza *et al.* (2010) reported that Physico-chemical characteristics of Dal Lake undergo rapid eutrophication under increasing anthropogenic impacts of drainage of the basins. The chief cause of pollution in these basins is attributed to the addition of major plant nutrients particularly Nitrogen and Phosphorus. The sources of these nutrients were traced to human wastes, detergents, agricultural practices and were responsible for reducing water quality and BOD of the lake.

Parray *et al.* (2010) reported unusual physico-chemical parameters of Dal and some other lakes. The group also reported gradual increase in these parameters due to expanding urbanization, intensive agricultural practices and unsustainable exploitation of wetland resources.

Patra *et al.* (2010) investigated some physico-chemical parameters of water samples from three different sectors of Chilka Lake from March 2008 to Feb 2009 after the opening of new mouth near Gabakunda. Overall analysis revealed a lot of fluctuation in the physico-chemical parameters of the water samples. The pH of water was alkaline and along with it salinity varied widely throughout the lake COD was very less due to absence of chemical pollution.

Sinha and Biswa (2011) in a study monitored fifteen physico-chemical parameters of a lake at Kalyani, West Bengal, for a period of one year, to assess its pollution status. The water quality index (WQI) was calculated using twelve parameters only. The lake was considered to be trophic as evidenced by its shallow depth (2-4m), low transparency (14-34 NTU), low dissolved oxygen (2.5-6.0 mg/l) and higher concentrations of other nutrients such as phosphates, nitrates, sulphates and chloride. High value of WQI indicated high degree of pollution

making the water of the lake unsuitable for human use as well as for fish culture unless treated and disinfected properly.

Islam *et al.*(2012) assessed the hydrological properties and water quality characteristics of Chini Lake in Pahang, Malaysia. A total of seven sampling stations were established at the main Feeder Rivers of Chini Lake for measurement of stream flow. A total of 10 monitoring stations covering the study area were selected for water sampling. Fourteen water quality parameters were analyzed based on *in-situ* and *ex-situ* analysis for two seasons and laboratory analyses were carried out according to the HACH and APHA methods. Water quality in Chini Lake varied temporally and spatially and the most affected parameters were pH, TSS, turbidity, DO, ammoniacal nitrogen, phosphate and conductivity. Based on the Malaysian Water Quality Index (WQI), the water in the Chini Lake was classified under class II, meaning it is suitable for recreational activities and safe for body contact.

Nwachukwu *et al.* (2013) carried out study on the physico-chemical analysis of three fresh water springs in district Pulwama of Kashmir Valley viz: Sandyasar naag (Ladhu), Batenaag (Khrew) and Sonaraaz naag (Shar). The samples were collected on monthly basis from January to June 2011. The various parameters were analyzed after APHA (1992) and Gupta (2004). The determined values of the three springs depicted slight variations for depth, pH, temperature, alkalinity, ammonical nitrogen, calcium hardness, chlorides, DO, free CO₂ and nitrite Nitrogen. However marked differences were observed for conductivity, orthophosphorous and magnesium hardness. Moreover the variations for total hardness and TDS depicted ascending and descending trend.

Pratiksha Tambekar *et al.* (2013) collected water samples and analyzed as per standard methods. Parameters such as pH, turbidity were measured in situ during the sampling. Higher values of several physico-chemical parameters indicate the pollution of riverine ecosystem in the study area. Domestic wastes, municipal sewage, industrial effluent from paper and pulp industries as well as

agricultural runoffs are directly or indirectly responsible for deterioration of water quality. Statistical analysis including correlation method average values (AV), Standard Deviation (SD), Standard Variance (SV), Standard Error (SE) and 95% confidence limit (CL) were carried out to assess the pollution load. The results revealed that most of the water samples do not meet WHO and BIS water quality standards, while many samples showing severe water quality deterioration.

Mehboob and Balkhi (2014) studied seasonal variation in the physico-chemical parameters and bacterial load in the Dal Lake. The mean temperature seasonally varied significantly with values of 20.56 ± 0.57 , 7.78 ± 0.39 and 11.50 ± 0.7 °C, respectively during autumn, winter and spring seasons. The pH of sampled Dal water was slightly alkaline. Dissolved O₂ and chlorine content in water in different seasons showed slight variation.

2.2 Occurrence and seasonality of fish parasites

Pomphorhynchus kashmirensis is very much prevalent in the fishes of the Kashmir Valley especially in *Schizothorax* species that is evident from the literature cited below.

Datta (1936) described Helminth parasites with special reference to Acanthocephalans of fishes. Kaw (1941) provided a detailed account of Helminth fauna of fishes in Kashmir.

Cushing (1942) described the various abiotic factors responsible for the parasitic infection in fishes. The role of temperature has been greatly emphasized by the author for the incidence of infection and hence the antibody production.

Bisset (1948) worked on the immune system of fishes and concluded that infection increases with a steep rise in temperature.

Fotedar and Qadri (1974) studied the impact of introduced carp and the deteriorating ecological conditions of lakes of Kashmir especially Wular and Dal on *Schizothorax* and *Orienus*.

Amin (1975) studied the host and seasonal associations of *Acanthocephalus parjsidei* (Acanthocephala: Echinorhynchidae) in Wisconsin fishes and observed that highest abundance and maturation occur during summer and recruitment during summer and autumn.

Andryuk (1979) investigated the life cycle patterns of acanthocephalans with special reference to *Acanthocephalus lucii*.

Radujkovic *et al.* (1983) investigated the ill effects of some parasites especially Acanthocephala and Nematoda on the fish host, *Chelon labrosus*. Acanthocephalan alone was found to be responsible for macrocytic anaemia while as nematodes were neutral in this regard. They further explained that acanthocephalans may also responsible for Vitamin B and folic acid deficiency in the fish host.

Gleason (1984) studied the seasonal prevalence, intensity of infection, infrapopulation and dispersal pattern of *Pomphorhynchus bulbicollis* in *Hyperntelium nigricans*. Significant differences were found in the seasonal prevalence and mean intensity of infection; both were low in winter and high in summer.

Jr. Williams and Rogers (1984) identified new species of *Pomphorhynchus* which was collected from 14 host species representing 7 families and six orders of fishes from northern Florida and southern Alabama. It differed from all known species of *Pomphorhynchus* by possessing 20-23 proboscis hooks per row. It differed from *P. rocci* by having a longer neck in relation to trunk length, larger hooks, a longer proboscis, and a smaller trunk.

Amin (1987) worked on *Pomphorhynchus bulbocoli* from fishes of Wisconsin lake and concluded that its shows seasonal prevalence and that host specificity were not so specific.

Brown (1989) while working on seasonal dynamics of the *Pomphorhynchus laevis* in its intermediate and preferred definitive hosts observed

that the rate of parasitic growth increased with water temperature.

Dhar and Peerzada (1989) studied the seasonal variation in helminth parasites infecting *Schizothorax niger*. Three parasites viz. *Diplozoon kashmirensis*, *Adenoscolex orieni* and *Pomphorhynchus Kashmirensis* showed different seasonal peaks in summer, winter and spring respectively.

Khidr (1990) studied the seasonality of a monogenean *Enterogyrus cichlidarium* inhabiting the stomach of *Tilapia* spp. In *Tilapia nilotica* and *T. zillii* the peak of the infection was found in winter.

Nie and Kennedy (1991) found that *Paraquimperia tenerrima* (Listion) a nematode has higher prevalence during winter to late spring or early summer.

Dhar and Peerzada (1992) observed seasonal occurrence and maturation of the cestode *Adenoscolex orieni* in fishes of Wular Lake. The maturation of the parasite was seasonal and recruitment occurred in autumn.

Jha *et al.*(1992) reported seasonal occurrence of helminth parasites in fishes, and showed moderate to high occurrence during different months, while digenetic trematodes were having quite low prevalence and even the infestation was absent during certain months of investigation period.

Chishti and Peerzada (1995) studied the seasonal occurrence of Diplozoan species in fishes of Wular Lake. All hosts examined showed maximum prevalence during summer months.

Molloy *et al.*(1995) studied the population biology of *Pomphorhynchus laevis* in brown trout (*Salmo trutta*) caught from two lakes in the Burrishoole River system in Irish Republic.

Majidah and Khan (1996) studied the population dynamics of nine species of helminth fauna including *Diplozoan kashmirensis*, *Clinostomum schizothoraxi*, *Camallanus fotedari*, *R. himalayai*, *R. kashmirensis*, *Adenoscolex oreini*, *Gangesia fotedar*, *Pomphorhynchus kashmirensis*, *N. manasbalensis* from

seven species of fish (*Schizothorax hugeli*, *S. esocinus*, *S. curvifrons*, *S. niger*, *Oreinus plagiostomus*, *Nemachilus kashmirensis*, *C. carpio specularis* of Wular lake, Kashmir, India.

Morand (1996) investigated that parasite body size is positively correlated with the host body size. He further proved that parasite body size is related to host longevity. Long lived hosts would provide more energy and would harbor more long lived parasites and hence the larger ones.

Yousuf and Pandit (1996) studied the developmental process of the *Schizothorax niger* heckle inhabiting different lakes of Kashmir Valley. They observed a steep rise in temperature hastens the hatching which usually takes place after 13 days of fertilization in a temperature range of 9-12°C. Its breeding grounds are usually located in the shallow parts of lakes on spring beds.

Bakker *et al.* (1997) for the first time showed that both the parasite colour and changed intermediate host behavior promote the transmission of *Pomphorhynchus laevis* to its next host.

Chishti and Peerzada (1998) carried out a helminthological survey on the fishes of Wular. Out of 1662 specimens 155 were infected with acanthocephalans and out of which maximum infection was seen with *Pomphorhynchus kashmirensis* and least with *N. manasbalensis*. Maximum infection was recorded in spring season in all hosts and was usually dominated by males. An increase in the mean number of parasites per host with an increase in the host length was also evident.

Grutter (1998) observed the prevalence and number of *Benedenia* species on *Hemigymnus melapterus* and was significantly greater on fish from the reef flat than from the reef slope at Heron Island. This difference in parasite abundance between the habitats suggests that *H. melapterus* does not move between the reef flat and reef slope separated by only a few hundreds of meters.

Khan and Majidah (1999) studied the impact of physiochemical

parameters on the diversity of fish parasites and concluded that parasite infection showed a regular seasonal trend. Infection was highest in the late summer and early autumn months. Infection was also influenced by other biotic and abiotic factors also.

Kennedy (1999) investigated the possibility of post-cyclic transmission in *Pomphorhynchus laevis*. Rainbow trout were exposed to *P. laevis* in naturally infected *Cottus gobia*, *Nemacheilus barbatulus*, *P. phoximus* and *L. cephalus*. Post cyclic transmission of gravid parasites could occur from *C. gobia* but not from *L. cephalus* which indicates that this failure to transmit larger parasites of either sex reflects the age and so development of the proboscis bulb of *P. laevis* and the extent of the host encapsulation response.

Ahmad and Chishti (2000) carried out a helminthological survey of freshwater fishes of Kashmir valley and revealed that the fishes of Anchar and Manasbal lakes only were infected with a digenetic trematode *Clionostomum Leidy*, 1856.

Cribb *et al.*(2000) reported a heterogeneous distribution of *Pomphorhynchus heronsis* in the coral reef fish over small distances. Individual fish from the reef slope had 0-9 worms while as individual from reef flat had 1-122 worms. Other variables (year, season, size) of fish made little contribution to the variation. These results imply both that the fish have very limited local movement and that transmission of the parasite is concentrated locally.

Jahan *et al.*(2000) studied the parasites of *Schizothorax* species and *Cyprinus carpio* from River Jhelum and recorded the presence of a new cestode *Bothriocephalus* (Rudolphi, 1808).

Machado *et al.*(2000) examined the fish for Helminth parasites. Prevalence and total host length were positively correlated in fish parasitised by cestodes. Infection intensity and host length were positively correlated only for the cestode *P. microscopicus*. There were significant differences in the prevalence

of parasites in males and females of *C. monoculus*.

Cave *et al.*(2001) compared the Helminth parasite communities in eel from lagoons of Adriatic coast and Tyrrhenian coast. It was proved that there is similarity in composition and structure of Helminth communities in eels from coastal lagoons throughout Europe.

Dezfuli *et al.*(2001)studied species co occurrences and interspecific associations between intensity of infection in Helminth communities of three populations of brown trout from northern Italy. Variations in fish size and its effect on infection levels, and whether or not two Helminth species used the same or different intermediate host could not be revealed as reported by other authors. Therefore, they suggested that interspecific associations may be condition dependent: even in apparently similar localities, the same combinations of helminthes species show different associations.

Evans *et al.*(2001) gave first record of the *Pomphorhynchus laevis* (Acanthocephala) in fishes from Northern Ireland.

Guillen-Hernandez and Whitfield (2001) revealed the sympatric occurrence of the freshwater and marine/estuarine strains of the *Pomphorhynchus laevis* to compare their infection levels in *Platichthys flesus*. They observed that freshwater worms were larger and had more eggs than marine/estuarine worms.

Lyndon and Kennedy (2001) worked on acanthocephalan parasites in freshwater fish from the British Isles. They proposed that all the known species have been able to successfully colonize by a variety of means. Foremost among these is the utilization of a migratory fish host in their life cycle, allowing colonization of new areas and rescue effects in established areas. In addition all six species appear to exhibit resource partitioning by host at either or both the larval stages, thus reducing the potential for competition and further facilitating colonization and survival.

Tingbao and Xianghua (2001) studied the seasonal population dynamics

of *Neoechinorhynchus quinghaiensis* in *Gymnocypris species*. Prevalence values were above 44% in all seasons sampled without a distinct seasonal trend. Mean intensity reached a peak in the autumn, and then decreased throughout the winter and spring to reach its level in summer. The sex ratio of female to male was both high in winter (1.51:1) and spring (1.48:1). These authors believed that the higher proportion of females and the change in the worm sex ratio in winter can be attributed to the reduced longevity of male worms.

Aloo (2002) conducted a helminthological survey on two fish species and recovered five larval Helminth parasites in them: a nematode, an acanthocephalan, *Polyacanthorhynchus kenyensis*, a digenetic trematode and two cestodes. Both prevalence and intensity of the infection of these helminthes increased in large sized fish, whereas male fish were more heavily infected than females. No seasonality in infection was observed by the researcher.

Akifumi *et al.* (2002) investigated that fish hosts were heavily infected in lakes with dense population of the isopod intermediate host. In rainbow trout, male worms were abundant from winter to spring and female worms were immature during these seasons. Gravid females were abundant during summer and autumn. They concluded that *Acanthocephalus* sp. is an annual species and its recruitment for the intermediate host to the fish occurs mainly in winter and spring.

Poulin (2002) has examined the relationship between the species diversity of taxa, the mean number of article published per year on each taxon, the mean impact factor of the journals in which they appear. Six taxa of Helminths: Nematophora, Acanthocephala, Monogena, trematode, cestoda and Nematoda were considered. Out of these six taxa the mean journal impact factor correlated positively and significantly with the mean annual number of papers published. More number of papers was published on Nematodes, Trematodes, Cestodes than Nematomorphs or Aacathocephalans.

Amin *et al.* (2003) gave a description of *Pomphorhynchus spindletruncatus* from freshwater fishes in Northern Iraq and keys to genera of the Pomphorhynchidae and the specie of *P. monticelli*, 1905.

Blanco *et al.*(2003) described the best management practices, in aquaculture which control infectious diseases and improves safety of the fish products. These authors stress on the implementation of integrated measures at the production level.

Nedeva *et al.*(2003)studied morphology particularly the morphometry of *Pomphorhynchus laevis* from river Danube. Extensity and intensity of the invasion in different fish hosts species were also investigated.

Ziolkowska and Rokicki (2003) searched the real intermediate host of *Pomphorhynchus laevis* in brackish waters of the Baltic Sea. They concluded that *Gammarus zadolachi* is probably an intermediate host for *P. laevis* in the Baltic Sea.

Rauque *et al.*(2006) investigated the seasonality of recruitment and reproduction of *Acanthocephalus tumescens* at the component population level. Overall prevalence, mean intensity, and coefficient of dispersion showed the same pattern of seasonal changes. The seasonal feeding pattern of fishes affects the occurrence of *A. tumescens* producing 1 peak in spring and the other peak in autumn. The low temperature in winter delay reproductive process after the autumn periods of recruitment.

Simkova *et al.*(2006) investigated the patterns and likely processes connected with evolution of host specificity in congeneric monogeneans parasitizing fish species of the Cyprinidae. They confirmed the hypothesis of specialization i.e. specialist parasites with larger anchors tend to live on fish species with larger body size and greater longevity. The mapping of morphological characters of the attachment organ onto the parasite phylogenetic tree reveals that morphological evolution of the characters of attachment organ is

connected with host specificity in the context of fish relatedness, especially at the level of host sub families.

Beneshet *et al.*(2007) examined the life cycle pattern of acanthocephalans and concluded the various approximate factors that play an important role for its completion.

Mustafa and Altunel (2007) examined three fish species from Enne Dam Lake of Turkey for parasitic infections. There was a significant positive correlation between fish length, fish weight, infection rate in Crucian carp but there was no clear correlation existing between length, weight, and parasite infections in bleak. In addition, a significant negative correlation was found between water temperature and infections in golden carp.

Ahmad *et al.*(2008) undertook a study to find out the host specificity among the parasites of freshwater fishes of Kashmir and concluded that host specificity of *Rhabdochona guptai* and *Allocreadium nemachilus* was highly specific while as *Pomphorhynchus kashmirensis* showed least host specificity.

Benesh *et al.*(2008) recorded five traits from isopods infected with an acanthocephalan (*A. lucii*)and suggested that the host behavior tremendously changes over time with the acanthocephalan infection.

Custodio *et al.*(2008) conducted research work for the first time on metazoan parasites of common carp from the river Limpopo and the lagoon Chuali. Nine metazoan parasites were detected including one acanthocephalan (*Acanthogyrus tilapiae*). The parasites communities from river Limpopo and Lagoon Chuali were very similar, exhibiting low diversity and were dominated by a single species, *Pomphorhynchus samfya*.

Ehab and Faisal (2008) studied Largemouth bass *Micropterus salmoides*(L.) which is a popular freshwater sport fish in Michigan. These authors noticed that this fish is severely plagued with endoparasites especially the bass tapeworm, *Proteocephalus ambloplitis* Leidy and different species of

acanthocephalans. They observed an inverse correlation: when the number of *Proteocephalus ambloplitis* significantly increased in the ovary and the number of *Neoechinorhynchus* sp. decreased in the intestines. Acanthocephalan adults were found in the intestines or in the pyloric caecae and were usually associated with damage of intestinal mucosa at sites of attachment.

Hermida, *et al.*(2008) investigated the gills, digestive tract and swim bladder of eels from Ria de Aveiro for the presence of the parasites. Fifteen metazoan parasite species were found including the acanthocephalans parasitizing the fish host.

Rubio *et al.* (2008) worked on the farmed rainbow trout *Oncorhynchus mykiss* and brown trout *Salmo trutta*. These hosts were monitored for infection with the blood feeding gill fluke *Discocotyle sagittata*. They observed new infections were dominated during summer/autumn and was negligible during winter/spring season. Thus, they again showed temperature is having a vital role in parasitic infection.

Selda *et al.*(2008) investigated monthly variations and the effects of host size on parasite prevalence and mean intensity in common carp from Beysehir Lake in Turkey.

Ahangar *et al.*(2012) investigated parasites of schizothorax (Native fish) collected from River Jhelum, there was mixed infections of cestode, Adenoscolex and Pomphorhynchus. However, there was maximum infection of *Pomphorhynchus* species (Acanthocephala) in the *Schizothorax* species of River Jhelum. Tremendous reduction in weight was observed in the infected fishes. The intestine of the infected fish with *Pomphorhynchus* species presented a nodulated appearance due to the protruded bulbs of the parasites. These parasites were seen to have penetrated through the intestinal wall with their ensheated ends floating freely in the coelom and the part of the trunk in the lumen of the intestine. The intestinal wall was completely disrupted at the point of penetration. During the

investigation period, a single Acanthocephla was found penetrated in the liver of *schizothorax* sp.

Yakhchali *et al.* (2012) this investigation was undertaken to verify the prevalence of helminths parasites in the gastrointestinal tract of catfish. A total number of 116 catfish (*Silurus glanis*) were collected from Zarrine-roud River and examined for helminths. Fish were examined after washing contents of gastrointestinal tract and observed for the presence of helminths using a stereo microscope and a light microscope. Results indicated that 18.96% of the examined catfish were infected with digenean trematodes including *Orientocreadium siluri* (27%), *Crowcrocoecum skrjabini* (39%), and cestode *Bothriocephalus gowkongensis* (34%).

Khurshid *et al.* (2013) conducted an investigation of Helminth parasites of *Schizothorax* (Native fish) and *Cyprinus carpio* (Exotic fish) collected from Shallabugh Wetland was undertaken for a period of one year from august 2010 to July 2011. Out of 486 fishes collected equally throughout the year, a marked helminth infestation was observed in *Schizothorax* in comparison to *Cyprinus carpio* which showed a little trematode infection during the entire period of study. Species of *Schizothorax* were found to be abundantly infested with trematodes followed by cestodes and acanthocephala. However, less infestation of trematodes, cestodes and acanthocephalans was observed in *Cyprinus carpio*, indicating the susceptible nature of the *Schizothorax* species to helminth infestation. From the present study, it may be inferred that the susceptibility of *Schizothorax* species to Helminth infestation may be considered as one of the factors responsible for the decline of this native fish from the water bodies of Kashmir valley.

Shah *et al.* (2013) correlated prevalence of helminth parasites with water temperature, pH, dissolved oxygen (DO of water body. *Adenoscolex* sp. (Cestode parasite) showed negative correlation with temperature which was statistically significant and showed positive correlation with pH. The *Bothriocephalus* sp. showed positive correlation with water temperature and DO. The correlation

between *Diplozoon* sp. and *Clinostomum* sp. with the physico-chemical parameters was non-significant.

Das and Goswami (2014) conducted the study to identify and determine the distribution along with prevalence and intensity of infestation brought about by the Helminth parasites in *Anabas testudineus* (Bloch), *Colisa fasciata* (Bloch and Schneider) and *Trichogaster lalius* (Hamilton) from three wetlands of Goalpara district, Assam during a period of Feb 2012 to March 2013. A total of 14 Helminth parasites (7 trematodes, 6 nematodes and 1 acanthocephala) were detected from different digestive organs with the highest rate of infestation from intestine (74.4, 59.5 and 45.6% viz. in *A.testudineus*, *C.fasciata* and *T.lalius*). The study reveals that helminthes show seasonality in prevalence, mean intensity and abundance of infestation.

Ibraq and Fayaz (2014) compared the impact of different levels of pollution in the two water boodies n the same species of fish to determine prevalence of parasitic infection.

Qayoom *et al.* (2015) studied helminth parasitic distribution in five species of native schizothoraecines of river Jhelum of Jammu and Kashmir in summer and autumn season.

Zargar *et al.* (2015) examined monogenean gill parasite, *Diplozoon kashmirensis* of the *Carassins carassius* to evaluate the infection level and the factors influencing the infection. Results showed the highest prevalence (34.22%) of *D. kashmirensis* was in the Lake having high trophic status and least prevalence (10.90%) in the lake having least trophic status.

Zargar *et al.* (2015) evaluated the relationship between fish parasites and water quality in Kashmir Himalayas and assessed helminth parasite densities in *Schizothorax niger* Heckel, 1838 (an endemic cyprind fish of Kashmir) from three Lakes, namely Anchar, Mansbal and Dal which reflenced the varied stages of Eutrophication.

2.3 Heavy metal concentrations and their bioaccumulation

Fishes are continuously exposed to waterborne and particulate heavy metals due to continuous flow of water through gills and through food sources. Metals bioaccumulated in different tissues follow different patterns of bioaccumulation factors (Fatima and Usmani, 2013). The mechanism of bioaccumulation of heavy metal in fish includes different processes in dynamic manner. Both physiological/biochemical responses and metal geochemistry are responsible for the differences in metal concentrations observed in different populations of aquatic species. It was confirmed that the internalization of metals into the cells of gills and internal epithelia follows similar mechanisms from different bioaccumulation studies (Noegrohati, 2006). A number of factors such as sex, age, season, spawning period, variability of food habitats and pollutant exposure and phylogenetic differences in regulatory mechanisms may influence the uptake, retention and bioaccumulation of trace contaminants in fish tissues (Nesto *et al.*, 2007).

Javed and Hayat (1996) determined that different genera of algae can act as indicator in appraising and estimating aquatic pollution with heavy metals. Significant correlation between the metallic toxicity of planktonic biota and water demonstrated the capability of plankton to bio-concentrate heavy metals from aquatic environment.

Sues *et al.*(1997) conducted study in a freshwater subalpine lake in Austria with localized contamination from motorway runoff. It compares the accumulation of lead and cadmium in the mussel *Dreissenapolyomorpha* with that occurring in a common fish species and its intestinal acanthocephalan parasite (*Acanthocephalus lucii*)in close proximity to the motorway and at a distant reference site.

Canli *et al.* (1998) studied the heavy metals, chromium, copper, nickel, cadmium and lead in the fish species, *Chondrostoma regium*, *Cyprinus carpio* and

Barbus capito at five sampling sites established on the Seyhan River, Turkey. They reported higher metal levels in fish liver and gill as compared to the muscle. Heavy metals were also found beyond the safe limits for human consumption.

Yilmazer and Yaman (1999) studied the annual variations in the concentrations of major dissolved ions and metals in the water and suspended matter of the aquatic ecosystem of Ceyan River. The concentration of heavy metals in suspended matter followed the order: aluminum> manganese> nickel> chromium> lead> zinc> cobalt> copper> cadmium. The high concentration of heavy metals in suspended particles was attributed to anthropogenic inputs and pollution due to vehicle fuel remains and phosphate fertilizers.

Javed and Hayat (1999) studied the role of plankton as an indicator of heavy metal pollution by zinc, cadmium, iron, lead, nickel, and manganese and reported significant discrepancies in the metallic ion concentrations of water due to discharges of domestic sewage, untreated industrial effluent and wastewater into the river due to various effluent discharging tributaries.

Kotze *et al.* (1999) determined zinc and copper bioaccumulation in fish species, *Clarias gariepinus* and *Oreochromis mossambicus* obtained from Olifants river, South Africa and reported significantly high concentrations of metals correlated with metallic toxicity of the ecosystem. The degree and extent of metals accumulation varied between the two species attributed to differences in behavior and feeding of these fish. Copper concentration in the fish tissues followed the order: liver> gills> skin> muscle while zinc did not show any particular pattern. Zinc accumulation was significantly higher in liver than that of muscle tissues.

Sweet and Zelikoff (2001) studied that majority of obnoxious and toxic chemicals are dumped into the aquatic water bodies and water that can act as a source of these chemicals and several toxic agents. Therefore, the occurrence of toxic chemicals and environmental pollutants contaminate the aquatic ecosystems

that would affect the physical condition and endurance rate of fish.

Gbem *et al.* (2001) investigated the consequences of tannery effluents on the concentration and distribution of copper, cadmium, zinc and lead in *Clarias gariepinus* and the order of the distribution of metals in fish was lead>chromium>copper>zinc. The heavy metal concentrations in fish liver were significantly higher ($p<0.05$) as compared to other body organs. Metals bio-accumulation in fish liver was followed by the gills and the gut while metals were found lower in muscle tissue.

Fatoki *et al.* (2002) determined the toxicity of manganese, copper, iron, aluminium, cadmium, lead and zinc in the Umtata River, Nigeria. The mean levels of aluminium in the river water varied between 0.22 - 0.36 mgL⁻¹ which were beyond the permissible limits.

Smolders *et al.* (2003) studied the effects of industrial effluents and domestic sewage on the aquatic ecosystems and revealed great ecological disturbances due to continuous contribution of pollutants and contaminants in the aquatic ecosystems.

Canli and Atli (2003) and Mendil and Uluozlu (2007) reported that accumulation of heavy metals in various tissues of different fish species living in the same habitat varied significantly.

Ozmen *et al.* (2004) investigated the concentration of metals, zinc, iron, manganese, nickel, lead, cobalt, copper, chromium and major elements, calcium and magnesium contamination in the surface water and deep sediment samples at various sampling sites in the Hazar Lake, Turkey. Lake water manifested higher macro-nutrient levels and lower heavy metal concentration as compared to World Health Organization standard due to intensive domestic activities and sewage water infiltration resulting in the eutrophication of the lake. The variability of heavy metals and major elements in the sediments was in the order: iron>magnesium> calcium> manganese> zinc> nickel> chromium> copper>

cobalt > lead.

Olaifa *et al.* (2004) analysed samples of gill, bone, intestine, muscle and water for five metals viz. manganese, copper, zinc, iron, and chromium by atomic absorption spectroscopy (AAS) in two separate experiments. In each case, two tissues were compared with the levels of the metals in water viz. gill, bone, and water; intestine, muscle and water. Generally, lower concentrations of the metals were recorded in water than fish tissues. Higher concentrations of zinc than recommended by the Federal Environmental Protection Agency were recorded in the fish during the dry season. Iron was the dominant metal in the muscle while chromium was the least. Significant differences ($p < 0.05$) were recorded in copper and zinc concentrations in the muscle, intestine and water during the dry and rainy seasons. In gill, bones and water, significant differences ($p < 0.05$) were only recorded for the two stations for copper during the rainy season and only zinc was significantly different ($p < 0.05$) in the dry season. It was concluded that though the heavy metals of interest were present in measurable quantities there were still within safe limits for consumption.

Milenkovic *et al.* (2005) determined the concentrations of arsenic, iron, chromium, manganese, copper, nickel, cadmium, lead, mercury and zinc in the sediments of river Danube and found that the range of mean concentrations of heavy metals increased by 46.60 to 156.20% due to enhanced industrial effluent discharges in the river. The metallic toxicity of copper, chromium, nickel, zinc and particularly cadmium in the sediments was higher than the target values indicating potential risk to the ecosystem. The behavior of metals in aquatic habitats depends upon the bed and suspended sediment composition and the physico-chemistry of water (Osmond *et al.*, 1995).

Rieumont *et al.* (2005) studied the spatial distribution of heavy metals, cadmium, copper, chromium, cobalt and lead in the sediments of Almendares River, Cuba. They reported higher metal levels than the probable effect concentrations indicating local eco-toxicological impacts and 62% of heavy

metals bound to the sediments could be released back into the river water.

Gobiel *et al.* (2005) studied the contribution of metropolitan and domestic wastes to the total metal fluxes in the St. Lawrence River and reported seasonal variations in the metallic ion toxicity. The ratios of nickel, zinc, cadmium, chromium and copper to aluminum in suspended particles were high when compared with the pre-industrial sediments that were suggestive of higher trace element fluxes.

Storelli *et al.* (2005) found that in eco-toxicology, both essential and non-essential elements have their specific significance because of their ability to persist in water for a longer period of time to affect the living organisms due to their tendency to bio-concentrate in the aquatic food chain/ecosystems.

Dalman *et al.* (2005) in this study, heavy metals (Pb, Cd) and trace elements (Cu, Zn) were analyzed in fish (*D. labrax*) and sediments in the Bay of Gu' llu'k by atomic absorption spectrometry. The average metal concentrations in the fish varied in the following ranges: Pb; <0.02-0.4, Cd; <0.01-0.04, Cu; <0.1, Zn; <0.5-7.2 mg kg⁻¹. In addition, seven sediment samples were analyzed and average concentrations of them were found as Zn; 80.8 } 0.45, Cu; 25.2 } 0.14, Pb; 20.0 } 2, Cd; 0.560.08 mg kg⁻¹. The accuracy and precision of our results were checked by using International Certified Reference samples (fish: DORM-2, sediment: HISS-1).

Ali and Fishar (2005) Concentrations of major metals (Na, K, Ca, Mg) and some trace metals (Fe, Zn, Mn, Ni, Cu, Co, Pb, Cr, Cd) were determined in water, sediment, benthos and some common fish species from Lake Qarun. Water and sediment samples were collected from seven stations where, the benthos and fish species were collected from three sites representing east, middle and west of the lake. Distribution of studied metals showed that, east part generally had higher contamination than west one which may be attributed to the impact of pollution sources in this area which coming from El-Batts Drain and pumping station in the

east part.

Obasohan *et al.* (2006) investigated the fish species, *alapterurus electricus* and *Chrysichthysnigrodigitatus* from the Ogba River for the determination of heavy metals toxicity in order to evaluate the pollution status of river ecosystem. The levels of copper, manganese, chromium and nickel in both species of fish were higher than the WHO and FEPA recommended maximum allowable standards for fish.

Ghosh *et al.* (2006) studied that arsenic exerts toxic and deleterious effects to fish even at non-lethal concentrations. Arsenic can also provoke time-dependent and tissue-specific changes in immune system of fish by impairing the functions of B-and T-cells, resulting in infectious diseases.

Dural *et al.* (2007) investigated the concentrations of zinc, copper, iron cadmium and lead in the liver, muscle, gill and gonad of three carnivorous fish species, *Sparus aurata*, *Dicentrarchus labrax* and *Mugil cephalus*. *Dicentrarchus labrax* and *Mugil cephalus* accumulated lowest concentration of heavy metals in their muscle while the highest metal levels were observed in the liver of *Mugil cephalus*.

Dural *et al.* (2007) reported liver, kidney and gills as metabolically active organs to accumulate more amounts of heavy metals than the other tissue like muscle. Fish liver and gills were reported as target organs for assessing metal accumulation. The amounts of metals in gills reflect the concentration of metallic ions in waters in which the fish live.

Kumar and Achyuthan (2007) Heavy metals disposed through anthropogenic activities find their way into the oceans and seas through the rivers or through direct fallout from factory effluents. These heavy metals resuspend back into the water column along with the sediments and are known to affect the marine animals. Marine animals like fish, prawn, crab and mussel were collected along the East Coast (off Pulicat lake to Chennai Harbour) to evaluate trace metal

concentrations in various tissues. The above specimens accumulated heavy metals such as Zn, Pb, Cu, Co, Cr, Ni and Cd. Fish, prawn, crab and mussel revealed higher concentration of heavy metals such as Zn, Pb, Cr, Co, Cu and Ni and Cd in low levels. The results revealed that the heavy metal concentrations in the marine animals are below the threshold levels associated with the toxicological effects and the regulatory limits. The bioconcentration factors revealed that the animals have accumulated heavy metals along the food chain rather than from the water column and sediment.

Rafiu *et al.* (2007) determined the concentrations of trace metals, cadmium, lead, manganese, zinc, copper and nickel in surface water and sediments along the Blaauwbankspruit stream, a tributary of the Crocodile River, South Africa. This investigation revealed higher metallic load in sediments than that of water due to wastewater discharge from sewage treatment plant and effluents from a gold mine.

Rauf *et al.* (2008) studied the heavy metal concentrations in the gills, liver, scales, muscles and skin of *Cirrhinamrigala*, *Labeo rohita* and *Catla catla* from the river Ravi and reported significantly variable concentrations of metals in fish organs. Fish liver showed significantly higher tendency for the accumulation of chromium and cadmium as 6.23 ± 1.14 and $4.26 \pm 1.57 \mu\text{g g}^{-1}$ as compared to gills that showed minimum levels of 1.46 ± 0.52 and $1.10 \pm 0.53 \mu\text{g g}^{-1}$, respectively.

Obasohan (2008) investigated the bioaccumulation of nickel in the fish, *Hemichromis fasciatus* from the Ogba river and nickel accumulations were significantly variable $p < 0.05$ in fish depending upon the season.

Papafilippaki *et al.* (2008) investigated the seasonal variations of copper, lead, chromium, zinc and cadmium in the surface water of the Keritis river, Greece. The toxicity of these metals varied significantly between the warm and the wet periods. Seasonal variations were attributed to agricultural activities, wastewater discharges and the physico-chemistry of water, temperature, and flow

rate, pH and redox conditions. The contamination of water with copper, cadmium, lead, chromium and zinc was positively related to the pH.

Ozan and Kir (2008) Concentrations of Al, B, Ba, Cd, Cr, Ni, Pb and Sr were analyzed by inductively coupled plasma opticaemission spectroscopy (ICP-OES) in the *Ligula intestinalis* plerocercoid (L., 1758), its host tissues (*Tincatinca* L., 1758), sediment and water from Beyşehir Lake. Al, Ba, Cd, Cr, Ni, Pb and Sr were highest in sediment, while B was the highest in liver. Al in *Ligula intestinalis* plerocercoid was 6.91 times higher than in fish muscle. The Al, Ba and Sr levels in *Ligula intestinalis* plerocercoid, were 2.99x, 1.23x and 2.26x respectively, higher than those in fish liver. Compared with water, all heavy metal concentrations in *Ligula intestinalis* plerocercoid were higher. This study supports the idea that cestodes aren't useful to determine the heavy metal pollution in aquatic systems when they are located in their intermediate host's body cavity.

Culioli *et al.* (2009) studied the impact of arsenic in the brown trout, *Salmo trutta*, living in a contaminated river of Corsica, France. Fish organs, liver, operculum and gills showed the highest concentrations of arsenic whereas concentrations in muscle were lower. Among the fish tissues, liver was the main storage compartment of arsenic (6.52 µg/g), followed by gills (4.83 µg/g), axial skeleton (2.03 µg/g) and muscle (1.45 µg/g).

Palaniappan and Karthikeyan (2009) investigated the bioaccumulation of chromium in the selected organs of *Cirrhina mrigala*, individually and in mixed solutions of chromium and nickel and found that the kidney was the target organ for chromium accumulation. In addition, the metal accumulation of the binary mixture of chromium and nickel was substantially higher than that of the individual metals in gills and intestine of the fish.

Begum *et al.* (2009) studied the metallic toxicity of water, plankton, sediments and fish in the Cauvery River. Heavy metal concentrations in water, fish muscle, phytoplankton and sediments varied significantly and increased in the

downstream of river indicating pollution due to anthropogenic activities. Fish can absorb metals directly from water via gills and through ingestion of contaminated food. Upon ingestion, metals are accumulated in fish liver, gills and other organs including lipids, muscle and membranes. Heavy metals concentrated in fish gills and liver tissues.

Ozturket *al.* (2009) In the present study, some heavy metals (Cd, Cr, Cu, Fe, Ni and Pb) were seasonally determined in water, sediment and some tissues of *Cyprinus carpio* from Avsar Dam Lake, which is an important water source for irrigation and drinking in Turkey. Heavy metal levels in water, sediment and fish samples were analyzed by inductively coupled plasma spectroscopy (ICP/AES). The obtained results showed that the average values of Fe in water samples were higher than the respective reference values for fresh water. Results for levels in water were compared with national and international water quality guidelines, as well as literature data reported for the lakes. The analysis of heavy metals in sediments indicated that among the six heavy metals tested, Fe was maximally accumulated, followed by Ni, Cu, Cr, Pb and Cd. Heavy metal concentrations were found to decrease in sequence of the *Cyprinus carpio* samples, in the muscle and stomach-intestine as Fe > Cu > Pb > Ni > Cr > Cd; in the gill, heart and liver as Fe > Cu > Ni > Pb > Cr > Cd and in the air sac as Fe > Cu > Ni > Pb > Cd > Cr. In the fish samples, cadmium, chromium, nickel and lead concentrations exceeded the tolerable values provided by international institutions.

Retief *et al.* (2009) eighty largemouth yellowfish, *Labeobarbus kimberleyensis*, were collected between April 2005 and February 2006 with gill nets close to the island (26°52, 249' S, 28°10, 249' E) in the Vaal Dam. The fish were killed, weighed and their length determined. Muscle, liver and spinal cord tissues were collected from each fish and the intestines removed and opened to expose *Bothriocephalus acheilognathi*. The tapeworms were collected in glass bottles and frozen. Water and sediment, as well as liver, muscle and tapeworm samples were digested and thereafter metal concentrations of 23 elements

(lithium, beryllium, titanium, vanadium, chromium, manganese, iron, cobalt, nickel, copper, zinc, arsenic, selenium, molybdenum, cadmium, tin, antimony, tellurium, barium, mercury, thallium, lead and uranium) were determined with an ICP-MS. Bioconcentration of metals (selenium, mercury, and lead during autumn; copper, zinc, selenium, cadmium, antimony, thallium and mercury during winter; lithium, zinc, selenium, cadmium and antimony during spring; and zinc during summer) occurred in tapeworms. The highest mean value was recorded in sediment, followed by water, tapeworms and host tissue. A seasonal trend showed that a higher concentration of the metals had accumulated in tapeworms during winter when water levels were at their lowest.

Sekabira *et al.* (2010) studied that rivers running through urban areas has faced water quality issues due to discharge of untreated domestic sewage, municipal wastes and industrial effluents into them leading to increase the metallic toxicity in the river waters.

Sharmin *et al.* (2010) investigated the heavy metals mobility pattern in sediments of the aquatic ecosystem of Nomi River, Japan. The heavy metal mobility in the sediments was in the order: cadmium > copper > chromium > nickel > iron > manganese. The presence of various clay minerals was found to be the major depository of heavy metals in sediments.

Elijah *et al.* (2010) evaluated the effects of a cestode parasite, *Ligula intestinalis*, on the accumulation of lead (Pb), cadmium (Cd), chromium (Cr) and copper (Cu) in the cyprinid fish, *Rastreneobola argentea*, in Lake Victoria, Kenya. This *L. intestinalis/R. argentea* model also was assessed as a bioindicator system for heavy metal contamination in the lake. The Pb, Cr and Cd concentrations in the *L. intestinalis* were higher than in the fish samples by a factor 11, 18 and 14 respectively, whereas the Cu concentration in *L. intestinalis* was increased by a factor of 2.5, relative to the Cu concentration in fish. The Pb, Cd and Cr concentrations in the parasite body increased, relative to their concentrations in fish samples, suggesting in the bioaccumulation of these metals

by the parasite. The Cu concentration in the fish parasite decreased, relative to increased Cu levels in the fish.

Akan *et al.* (2010) determined the extent of cobalt, zinc, copper, magnesium, cadmium, arsenic, nickel, manganese, iron, lead and chromium contamination and the degree of river sediment quality deterioration of Ngada river. The study revealed that heavy metals toxicity increased significantly with increasing sediment depth, indicating age-long accumulation of heavy metals due to anthropogenic sources and were higher than the WHO's standard sediment guideline limits exposing the aquatic food chain at high risk of induced heavy metal contamination.

Moustafa *et al.* (2011) evaluated some heavy metals pollution on *Oreochromis niloticus* in River Nile and Ismailia Canal. Results compared with the recommended standard levels of these metals in water it revealed that the levels of these metals in River Nile and Ismailia canal were found at critical limits that constitute a great potential health hazards.

Eneji *et al.* (2011) Heavy metals concentrations were determined in fish organs of *Tilapia zilli* and *Clarias gariepinus* from River Benue along Makurdi metropolis using atomic absorption spectrophotometer. The results indicated that *Tilapia zilli* gills contained the highest concentration (52.2%) of all the detected heavy metals, followed by the intestine (26.3%), while the muscle tissues appeared to be the least preferred site for the bioaccumulation of metals as the lowest metal concentration (21.5%) were detected in this tissue. Similarly, the *Clarias gariepinus* gills contained the highest concentration (40.3%) of all the detected heavy metals, followed by the intestine (31.6%), while the muscle tissue (28.1%) was the lowest.

Deenet *et al.* (2011) explained the relationship between Cadmium pollution in water and Crustacean gill parasites in freshwater cultured *Tilapia zilli* fish. A total of 375 adults cultured *Tilapia zilli* were studied the effect of water cadmium

pollution on clinical examination and the prevalent seasonal crustacean gill parasitic infestations in the period 2009-2010. This investigation revealed the appearance of the parasites during spring, summer and autumn and their disappearance during winter.

Javed and Usmani (2011) Investigations on the accumulation of heavy metals (Cu, Ni, Fe, Co, Mn, Cr and Zn) were carried out three commercially important fishes namely *Channa punctatus* (murrel), *Clarias gariepinus* (cat fish) and *Labeo rohita* (carp). The accumulation was observed in tissues of muscles, liver, kidney, gills, and Integument. The results revealed that the Fe and Zn concentrations were the highest in all tissues analyzed, followed by Ni, Cu, Co, Mn and Cr in almost all the three species. In the muscles of *Channa punctatus* the order of accumulation is Fe > Zn > Ni > Cu > Co > Mn, whereas in *Clarias gariepinus* it was Fe > Zn > Ni > Cu=Mn > Co > Cr. In *Labeo rohita* the pattern of accumulation was Zn > Fe > Ni > Cu > Co > Mn. The order of accumulation of heavy metals was similar in murrel and cat fish.

Alina *et al.* (2012) studied heavy metals (mercury, arsenic, cadmium, plumbum) in selected marine fish and shellfish along the Straits of Malacca. In this study, the marine fish and shellfish from the Straits of Malacca were analyzed using Inductively Coupled Plasma Optical Emission Spectrophotometer (ICP-OES) and Flow Injection Mercury System (FIMS) for Cd, As and Pb and Hg, respectively. The range of heavy metals in samples were 1.0-3-6.5-3 µg/g wet sample for Hg, 0.5-2-47-2 µg/g wet sample for Cd, 0.01-0.39 µg/g wet sample for Pb and 0.14-6.57 µg/g wet sample for As.

Zhao *et al.* (2012) shown correlation of heavy metals in the tissue of fish to their living environments both qualitatively and quantitatively and there was diverse metal bioaccumulation characteristics which was significantly affected by environment factors and living habits.

Okoth *et al.* (2012) determined how a cyprinid fish (*Rastrineobola*

argentea) partitioned four metals (Cd, Cr, Zn and Cu) in the subcellular fractions of the gut in presence of an endoparasite (*Ligula intestinalis*). The fish were sampled along four sites in Lake Victoria, Kenya differing in metal contamination. Accumulation of Cd, Cr and Zn was higher in the whole body and in the gut of parasitized fish compared to non-parasitized fish, while Cu was depleted in parasitized fish.

Chalpathi (2012) analyzed heavy metal (Cu, Zn and Pb) concentration in muscle tissues (gill, liver, eye, muscle, kidney, heart and intestine) of *Catla catla* and *Lebeo rohita* fish species from Mallemadugu area, near Renigunta, Tirupati. A simple preconcentration method was developed for the determination of copper, zinc and lead in fish samples. The detection limits of this method for Cu(II), Zn(II) and Pb(II) ions, were 0.65, 1.9 and 1.2 $\mu\text{g. L}^{-1}$ respectively. The maximum concentration of heavy metals was found in liver and gill, the order of heavy metals level in various organs liver >gill> intestine> muscle> heart> kidney >eye.

Barus *et al.*(2012) The tissue of two tapeworm species (*Ligula intestinalis* and *Bathybothrium rectangulum*) and body muscles of their fish host species were analyzed for heavy metal concentrations by standard methods using atomic absorption spectrometry. Regarding the values of accumulation ratio, the *L. intestinalis* accumulated 12.5-18.9 \times more lead, 2.3-3 \times more cadmium, and 4.4-14.1 \times more chrome, compared to respective metal concentrations in muscles of cyprinid intermediate fishhosts.

Khan *et al.* (2013) the present study was undertaken to study the concentration of some metals in the Lake. The mean value of Lead observed was 0.087 mg/l, where as cadmium was 0.053 mg/l and cobalt was 0.194 mg/l. Due to various anthropogenic activities and increase in pollution the water of the Lake has deteriorated.

Ibrahim and Omar (2013) worked out heavy metals concentrations;

iron (Fe), copper (Cu), cadmium (Cd), lead (Pb), zinc (Zn), chromium (Cr), manganese (Mn), mercury (Hg) and nickel (Ni) in water, sediment, and fish muscles of *Clarias gariepinus* collected from six areas at Assiut Governorate on river Nile using inductively coupled plasma mass spectrometry (ICP-MS). The results revealed that Zn, Cu and Fe concentrations were the highest in water and muscles, followed by Mn, Cr, Pb, Cd, Ni and Hg in areas under investigation. Also, summer was the highest accumulation season and winter was the lowest one.

Enuneku *et al.* (2013) determined heavy metals (Iron, Zinc, Nickel and Manganese) concentrations in surface water and tissues of fish (*Clarias gariepinus*) from River Owan. Water and fish samples were collected from three stations (Upstream, Midstream and Downstream). The physiochemical parameters were analysed using APHA standards. Heavy metal concentrations in water and fish were analysed using an atomic absorption spectrophotometer. Heavy metal concentration in surface water and fish were in the order: Fe>Ni>Mn>Zn and Fe>Zn>Mn>Ni. In fish, Fe recorded the highest level of bioaccumulation (58.96mg/kg) and Nickel (Ni) had the least bioaccumulation (2.38mg/kg).

Edward *et al.* (2013) assessed the concentration of heavy metals namely, Zn, Mn, Cu, Fe, Pb and Cd in three matrices including sediment, water and Fish organs (gills, flesh, kidney and liver). The fish sample, *Clarias gariepinus* was collected from Odo-Ayo River in Ado-Ekiti, Ekiti State, Nigeria. The results obtained showed that the concentration of heavy metal in water (Zn-4.65, Mn-0.79, Cu-0.84, Fe-5.87, Pb-0.16, Cd-0.13) was lower than that of the concentration of heavy metal in sediment (Zn-5.04, Mn-0.98, Cu-1.37, Fe-6.94, Pb-0.30, Cd-0.20), While the concentration of heavy metals in water was higher than that of fish parts (Zn-0.95, Mn-0.82, Cu-0.66, Fe-1.09, Pb-0.09, Cd-0.04).

Aladetohum *et al.* (2013) Monitoring the concentration of heavy metals in water, bio-accumulated in aquatic organism alongside parasite infestation has been of great importance for scientific studies. To evaluate the level of

contamination, the metal concentrations such as zinc, Iron, chromium, copper, lead and cadmium were analyzed in the liver and gills of *Liza falcipinnis* infected with a parasitic polychaetes (worms) collected from a period of six months (Feb. - July, 2013). These metal concentrations were measured from the digested fish and then aspirated into the flame of the Atomic Absorption Spectrophotometer (AAS) Perkin Elmer Analyst 200 using air-acetylene flame for the metal analysis against standard metal solutions.

Alkallak (2013) studied estimation of the accumulated concentration of cadmium and lead by using atomic absorption spectrometer, in the tissues of the cestode *Postgangesia inarmata* De Chambrier, Alkallak and Mariaux, 2003 and some organs of its final host, the catfish *Silurus glanis*, such as liver, kidneys, intestine, gills and muscles. The results showed a significant difference (0.05) in the accumulation concentration of cadmium and lead in the liver, kidneys, intestine, gills and muscles of the infected and uninfected fishes. The accumulation concentration of cadmium and lead in the cestode was 24.24 and 787.87 µg/g, respectively.

Shaikh(2013) The concentrations of heavy metals (Cd, Cr, Pb, Ni and Zn) in sample water and organs (muscle, gills and liver) of fish *Cirrhina mrigala* of river Godavari, at Nathsagar Dam in Maharashtra were analyzed. There was an appreciable decrease in metal concentrations in sample water from site I to site II. The heavy metal concentrations in sample water was in the order Ni>Pb>Cd>Zn>Cr. The concentration of Nickel was 7.53 53µg/L and that of Lead was 6.2653µg/L. Accumulation of heavy metals in the organs of fish *Cirrhina mrigala* was found in the order gills>muscle>liver.

Bashir *et al.*(2013) estimated concentrations of heavy metals, namely, Cd, Cu, Mn, and Zn, in the muscle, liver, and gills of two commercially important marine fishes, namely, *Arius thalassinus* and *Johnius belangeri*. The fish samples were collected from Kapar and Mersing, which are the west and east coastal waters of Peninsular Malaysia, respectively. The results showed that the muscle

had the lowest metal concentrations compared with the liver and gills. Among the estimated heavy metal concentrations, those of Zn and Cd were the highest and the lowest, respectively, for both species in muscle, liver and gills. Moreover, our results indicate that *A. thalassinus* has higher metal concentrations than *J. belangeri* in both areas. None of the values in the muscles exceeded the standard guideline values and hence would not pose any health hazard to consumers.

Imtiyaz *et al.* (2013) the present study was carried in the famous Dal Lake Kashmir. The metal contamination has been assessed in the drinking water of Dal dwellers. The study showed that there is an increasing trend in the metal contamination in the lake, no doubt still out of danger.

Yousafzai *et al.* (2014) analyzed copper (Cu) bioaccumulation in water, sediments and indifferent tissues of common carp (*Cyprinus carpio*), like muscles, Intestine, skin, liver and gills collected from Kalpani stream Mardan. The results recorded for bioaccumulation of copper was like that, in water samples the mean value for copper concentration was 0.014 ppm; in sediment samples mean value for copper concentration was 0.47 ppm. Similarly the mean value for copper concentration in Muscle tissue was 0.003 ppm; in Intestine tissue was 0.10 ppm; in Skin tissue was 0.06 ppm; in Liver tissue was 0.03 ppm and in Gills tissue was 0.07 ppm.

2.4 Histopathology

Histopathological alterations can be used as indicators for the effects of various anthropogenic pollutants on organisms and a reflection of the overall health of the entire population in the ecosystem. These histopathological biomarkers are closely related to other biomarkers of stress since many pollutants have to undergo metabolic activation in order to be able to provoke cellular change in the affected organism. For example, the mechanism of action of several xenobiotics could initiate the formation of a specific enzyme that causes changes in metabolism, further leading to cellular intoxication and death at a cellular level.

This manifests as necrosis at the tissue level (Bailey *et al.*, 1996).

Reichenbach (1976) has done pioneering work in the field of fish pathology as he presented collection of eight papers on the histopathology of fish caused by the metal pollution and parasitic infection and concluded that the parasitic infection is more alarming as compared to the environmental pollution.

Korting (1977) presented a brief review on hyperplasia and mechanical irritation caused due to cestode parasites, and concluded that this association does not cause any serious tissue inflammation.

Scott and Grizzle (1979) made an observation on pathology of Cyprinid fishes caused by *Bothriocephalus idella* and reported the disorders like focal pressure necrosis and excessive mucous production.

Jaraz and Szerow (1981) studied the histopathological change caused by a trematode in carp, and concluded that the parasite cause enteritis, which is frequently associated with congestion (extravasacularization) and oedema of the intestinal wall.

Sekretaryuk (1982) observed the interrelationship among the *Bothriocephalus* helminths parasites in carp, and suggested that in heavy infections parasites fail to find room for attachment on the host intestine, and derive their nourishment through the intermediary of another individual.

Bose and Sinha (1984) made an extensive study on *Heteropneustes fossilis* infected with nematode parasites, and concluded that the stomach tissue which was attacked by the parasites showed ulceration and desquamation and were undergoing lysis. The surface epithelium, lamina propria, muscle tissue and submucosa were disintegrated at the site of entry of the nematodes into the wall. Adjacent cells become flattened with displaced nuclei. Worm portions present in the sub mucosa were surrounded by fibroblasts, eosinophils and lymphocytes but no capsule was present. Blood vessels were congested and dilated, and in heavy infections there was wide spread haemorrhage and complete loss of muscle layers.

Otto and Heckman (1984) studied the host tissue response for trout infected with *Diphyllobothrium cordiceps* and concluded that the pleurocercoids were encapsulated with connective tissue that was infiltrated with lymphocytes and macrophages.

Ventura and Paperna (1985) described complete picture of histopathological disorders arising out of *Icthyophthirius multifiliis*, and claimed that the secondary infection by this parasite causes integumentary epithelium inflammation and repeated infection leads to massive cellular necrosis.

Wannstall *et al.* (1986) evaluated the consequences caused due to *P.levis* infection in rainbow trout and concluded that the mucosal epithelium of the gut adjacent to the metasoma of the worm suffered compression and abrasion. The prosoma of the *P.levis* penetrated the mucosal epithelium, lamina propria, stratum compactum, stratum granulosum, muscularis and serosa of the gut wall, and was invested by fibrous capsule of inflammation tissue.

Muralidhar and Shinde (1987) studied the histopathological of cestode *Acanthobothrium uncinatum*. A series of longitudinal and transverse sections revealed that the parasites approach the crypts of the lieberkuhn, attach by hooks and destroy the crypts, and finally reaching the intestinal submucosa which forms a plaque around the parasite.

Grishchenko and Kuznetsova (1988) found the effect of molybdenum on young rainbow trout, this was an histopathological study and concluded that there was an histological evidence of irritation of gills and skin.

Noga *et al.* (1990) studied the pathology caused by *Aeromonas salmonicida* in *Anguilla rostrata*.

Bell *et al.* (1990) reported the pathological effect of *Renibacterium salmoninarum* in *Anoplasma fimbria* (Sable fish).

Willer *et al.* (1991) worked on the histopathology of the swim bladder of cisco *Coregoniia artedii*, due to the presence of nematode *Cystidicola farionics*.

Chein *et al.*(1993) studied the pathological effect of *Aeromonas hydrophila* on *Anguilla japonica*.

Estene (1995) studied the pathogenicity of live bacteria and extracellular products of motile *Aeromonas* isolated from eel.

Flano *et al.* (1996) studied the histopathology of renal and splenic haemopoietic tissues of coho salmon, *Oncorhynchus Kisutch* infected with *Renibacterium salmoninarium*.

Wakabayashi *et al.* (1996) studied the haemorrhagic ascites of *plecoglossus altevelis* caused by *Pseudomonas* *sps.*

Aydin *et al.*(1997) worked on the hematological and pathological changes in rainbow trout *Oncorhynchus mykiss* due to the *Echerichia vulneris*.

Stehr *et al.* (2003) analysed in Vancouver Harbour, Canada, the occurrence of toxicopathic lesions in liver, English sole was statistically associated with concentrations of aromatic hydrocarbon (AH) metabolites in sediment and AH metabolite levels measured in bile. In general, liver histopathological lesions are not specific to pollutants. Furthermore, not all hepatic lesions identified in feral fish can be used as biomarkers since certain liver lesions appear to be species specific.

Antonio *et al.*(2007) analysed reared Nile tilapia, *Oreochromis niloticus*, of both sexes in freshwater and exposed to 0.5, 1.0 and 2.5mg L⁻¹ of waterborne copper for a period of 21 days. Liver and gill samples were collected after 21 days of exposure to copper and lesions were analyzed by light microscopy. The main histopathological changes observed in gills exposed to the highest concentration were edema, lifting of lamellar epithelia and an intense vasodilatation of the lamellar vascular axis. Although less frequent, lamellar fusion caused by the filamentar epithelium proliferation and some lamellar aneurisms were also found. The liver of control group exhibited a quite normal architecture, while the fish exposed to copper showed vacuolation and necrosis.

Marina and Claudia (2007) evaluated histological changes in gills, kidney and liver of the Neotropical fish species *Prochilodus lineatus*, subjected to *in situ* tests for 7 days in a disturbed urban stream and in a reference site, during winter and summer. Histopathology showed to be a very suitable biomarker for use in conjunction with the *in situ* test, because the seasonal variation did not interfere in the results and it was possible to differentiate the sites in the urban stream from the reference site.

Mohamed (2008) described the histopathological alterations in the liver of fish *Oreochromis niloticus* and *Lates niloticus* obtained from four khors (El-Ramla, Kalabsha, Korosko and Tushka) of Lake Nasser, Egypt, they included vacuolar degeneration in the hepatocytes, focal areas of necrosis, haemorrhage and haemolysis between the hepatocytes and dilation and intravascular haemolysis in hepatoportal blood vessels. Moreover, haemosiderin was seen around central veins and hepatoportal blood vessels.

Fatma (2008) investigated the concentration of some heavy metals (Fe, Zn, Cu, Pb, Cd and Co) in water and liver, gills, intestine, testis, heart and muscle of *O. niloticus* and *L. niloticus* obtained from four khors (El-Ramla, Kalabsha, Korosko and Tushka) of Lake Nasser, Egypt during 2006 (atomic absorption spectrophotometry) with emphasis on the histological alterations in these organs. Metal concentrations in the water of khors (mg/l) followed an abundance of: Fe > Zn > Pb > Cu > Cd > Co. Several histopathological alterations, including vacuolar degeneration with focal areas of necrosis in liver, proliferation in the epithelium of gill filaments and fusion of secondary lamellae, severe degenerative and necrotic changes in the intestinal mucosa and seminiferous tubules, degeneration and atrophy in cardiac muscle fibers and degeneration in muscle bundles were observed in the studied tissues of both fish as a result of the accumulated metals.

El-Naggar *et al.* (2009) determined the accumulation of iron, manganese, copper, zinc, cadmium and lead in liver of collected *O. niloticus* fish. In addition, the same liver samples were examined histopathologically. Results showed that

trace metals accumulations in fish liver at area under investigation were detected in following descending order: Fe > Cu > Zn > Mn > Pb > Cd. Histological study indicated that the liver of *O. niloticus* living in the studied stations showed several pathological alterations including: degeneration, fatty degeneration, necrosis and edema. Also congestion, branching (anastomosis), hemorrhage, hemolysis, hemosiderin and parasitic forms were seen in blood vessels.

Dezfuli *et al.* (2009) studied damage caused by digeneans of the mucosal epithelium of the villi. Necrosis and degeneration of epithelial cells were also evident. At the site of digenean infection, a high number of rodlet cells (RCs) and mucous cells were observed in the epithelium, with both types of cells exhibiting discharge activity.

Khoshnood *et al.* (2010) evaluated heavy metals bioaccumulation and explore their histopathological effects on hepatocytes of oriental sole (*Euryglossa orientalis*) and deep flounder (*Psettoodes erumei*), fishes were caught from two areas of north coast of the Persian Gulf, Bandar Abbas and Bandar Lengeh. Concentrations of nickel (Ni) and vanadium (V) in liver of both species in two sampling regions were in the following order: Bandar Abbas > Bandar Lengeh. Between the two species, these quantities were higher in *P. erumei* than *E. orientalis* in both sampling regions. Histopathology of the liver shows some cellular alterations including degeneration, necrosis and tissue disruption, and histopathological effects were severe in *P. erumei* than *E. orientalis*.

Yousuf *et al.* (2013) evaluated the presence and subsequent effect of trace metals (copper, zinc, iron and manganese) on the histomorphology of liver of *Schizothorax niger* (endemic fish) from Dal lake of Kashmir Valley Using Atomic Absorption Spectrophotometry. The highest concentration of metals was observed in the summer seasons and the lowest concentrations in the winter seasons during the entire study period.

Selvanathan *et al.* (2013) investigated heavy metals like mercury

and cadmium on fresh water fish *Clarias batrachus* carried out in the lab. The total autopsy was completed in less than 4 mins. The results showed that the degree of distortion of the gill, liver was proportional to the exposure period and concentration of the metals was found to be dose and time dependent.

Khan *et al.* (2014) evaluated the acute toxicity effects of Pb^{2+} on essential trace metals behavior and histopathology of Crucian carp (*Carassius auratus gibelio*). The fish were exposed to sub lethal concentration of $5mgL^{-1}$ as environmentally relevant Pb^{2+} for a period of 96 h. Trace metals (Pb, Cu, Zn, Fe and Ca) levels were determined in the gill, liver, kidney and muscles tissue by ICP-OES. Histopathological changes in the gills of exposed fish were characterized by lamellar shrinkage, disruption of cartilaginous core, epithelial lifting, lamellar shortening with desquamation and curling of the secondary lamella. The trunk kidney had severe shrinkage of glomeruli, hypoplasia of hemopoietic tissue as well as mild glomerular and tubular necrosis. The liver showed cellular edema, necrosis of hepatocytes with nuclear degeneration and pyknotic nuclei. The brain exhibited severe proliferation of glial cells, cellular necrosis, severe perivascular edema and satellitosis.

Chavan and Muley (2014) explored the impact of heavy metal mercuric chloride ($HgCl_2$), *Cirrhinus mrigala* was chosen as a model for the study. After determination of LC for mercuric chloride, 50 fishes were treated with $1/10^{th}$ and $1/20^{th}$ concentration of LC of mercuric chloride for a period of 30 days. The histopathological studies revealed that, the heavy metal under study is capable of inducing changes in different fish organ like kidney and intestine. The prominent histopathological changes in intestine include degenerative and necrotic changes in the mucosa, loss of structural integrity of villi, while kidney showed expanded renal tubules with a dissociated epithelial lining. Destruction of epithelial and mesenchymal cells, loss of tubular integrity due to necrosis was notable in kidney. The histopathological changes were positively correlated with concentration and time of exposure to heavy metal.

Kaoud *et al.* (2014) studied the effect of mercury (Hg) toxicity, its impact on liver histopathology, hematological and biochemical changes in Nile tilapia (*Oreochromis niloticus*). Significant changes in plasma aspartate aminotransferase (AST), alanine aminotransferase (ALT), alkaline phosphatase (ALP) and acid phosphatase (ACP) were observed in fingerlings. Results also, indicated that the addition of *Spirulina platensis* or chitosan to the Hg polluted media reduced significantly ($P < 0.05$). Hg level in aquarium's water as compared to that of Hg alone. They improve the hematological parameters (RBCs, Hb, Hct) and ameliorate the toxic effect of Hg in the aquatic environment.

Paul *et al.* (2014) studied histology of liver exposed to 36 hour LC50 concentration of copper sulfate ($\text{CuSO}_4 \cdot 5\text{H}_2\text{O}$) and cadmium chloride ($\text{CdCl}_2 \cdot \text{H}_2\text{O}$) with varying water pH (4 ± 0.5 , 7 ± 0.5 and 8 ± 0.5) in an air-breathing catfish (*Heteropneustes fossilis*). The histopathological changes observed in the liver tissue post exposure included necrosis, degradation of hepatocytes, degeneration of blood vessels, distended sinusoids with pyknotic nuclei and vacuolation of cells.

Bhuyan *et al.* (2014) analyzed qualitative and quantitative histological alteration in fish *Oreochromis mossambicus* collected from Kedilam River at three stations, which receive mostly industrial effluent and municipal runoff. Results showed that number of histological alteration observed in gill like structural alteration of epithelium, epithelial lifting, fusion of secondary lamellae and hyperplasia. In liver blood congestion, regressive changes like degeneration of hepatocytes, vacuolation, and necrosis observed where as in kidney it shows glomerular congestion, tubular degeneration, progressive changes like hypertrophied epithelial cells, haemorrhage in bowman's space.

Chapter3

MATERIALS AND METHODS

3.1 Study site

The valley of Kashmir is situated in the mid-Himalayas in the North West and South East direction within the coordinates 33°01-35°00N latitude and 73°48-75°30E longitude at an altitude of ≥ 1500 m above sea level. The study was carried out in two water bodies viz., the Dal Lake (34°07N-74°52E) and the River Jhelum (32°56' N and 73°44' East). Four sites that were selected from Dal Lake viz, Hazratbal Basin, Telbal Nallah, Dalgate ghat and Saidakadal. Three sites were selected from River Jhelum viz, Khannabal (Anantnag), Chattabal Weir and Zerobridge (Figs. 1 to 7).

The field collection was conducted on seasonal basis. The four seasons include spring (March-May, summer (June-August), autumn (September-November), spring (March-May) and winter (December-February).

3.2 Sample collection

Water and fish samples were collected seasonally. Water samples from the selected sites were collected ten centimetres below the surface. Three replicate of surface water samples were collected from each point of the same location using polyethylene sampling bottle. Prior to use the bottles were washed and soaked in 5% nitric acid and rinsed with deionised water. Further before sampling, the bottles were rinsed at least three times with water from the sampling site. The water samples were acidified immediately after collection by adding 5 ml nitric acid to minimize adsorption of heavy metals onto the walls of the bottles (APHA, 1998; Golterman and Clymo, 1978). Bottled samples taken to laboratory were filtered through 0.45 μm Whatman filter paper immediately after the samples have been transported to the laboratory and stored at 4°C until metal analysis.



Fig. 1 : Chattabal weir



Fig. 2 : Dalgate Ghat



Fig. 3 : Hazratbal basin



Fig. 4 : Saidakadal



Fig. 5 : Telbal Nallah



Fig. 6 : Zerobridge



Fig. 7 : Khannabal

Fish species(*Schizothorax niger*, *Schizothorax esocinus*, *Schizothorax curvifrons*) along with their parasites were collected randomly from two water bodies with the aid of local fishermen, quickly killed and stored on ice. The fish samples were immediately transported to laboratory where morphometric measurements involving wet weight (ww), total length, and weight of each of these fishes were carried out.

3.3 Physicochemical parameters of water

Water samples were collected during each survey and analyzed for various physicochemical parameters like water temperature, dissolved oxygen, free carbon dioxide and pH. For dissolved oxygen, water samples were collected in separate B.O.D bottles of 250ml capacity each as per the standard procedure. The samples were fixed on the spot as per the Winkler's unmodified method and brought to faculty of fisheries for further detailed analysis. Collected samples were analysed according to APHA (2005) for different physico-chemical parameters.

3.3.1 Temperature

Temperature of the water was recorded with the help of Celsius thermometer at the time of sampling.

3.3.2 pH

The pH of water was measured with a portable digital pH meter (Eutech) which was standardised with freshly prepared known Buffer solutions of pH 4.0, 7.0 and 9.2.

3.3.3 Dissolved oxygen

Dissolved oxygen was determined by modified Winkler's method (APHA, 2005). The sample was collected in 300 ml glass stoppered BOD bottle. Two ml of manganous sulphate and two ml of alkaline iodide azide solutions were

added to fix the dissolved oxygen. It was shaken thoroughly and the brown precipitate which appeared was allowed to settle. Two ml of concentrated sulphuric acid was added to it through the sides of the bottle and shaken well to dissolve the precipitate. 50 ml of this solution was taken in a conical flask and titrated with 0.025 N sodium thiosulphate solution using starch as an indicator to a colourless end point and was calculated using the following formula :

$$\text{Dissolved oxygen (mg/l)} = \frac{X \times N \times 8 \times 1000}{Y}$$

Where X = ml of titrant used
 Y = ml of sample
 N = normality of titrant

3.3.4 Free carbon dioxide

The method of APHA (2005) was followed for analysis of free carbon dioxide in the samples. Free carbon dioxide was analyzed by using phenolphthalein indicator and sodium hydroxide titrant. For this purpose, the water (50ml) was taken in a conical flask and 5 drops of phenolphthalein indicator were added to it. In case the water turned pink in colour, it indicated the absence of free carbon dioxide. If the water remained colourless, sodium hydroxide (0.227 N) was added to it until a permanent pink colour appeared and was calculated using the following formula :

$$\text{FreeCO}_2(\text{mg/l}) = \frac{X \times N \times 8 \times 1000 \times 44}{Y}$$

Where, X = ml of titrant used
 Y = ml of sample
 N = normality of titrant

3.4 Examination of live helminth parasites of fishes

3.4.1 Preliminary treatment of the fish

The fishes were first given serial number and then the total length (cm) and sex of each fish were recorded. After collection, the fish samples was subjected to helminthological examination and then preserved in ice. Fishes were dissected in the laboratory. Intestine was removed and examined for gastrointestinal helminth parasites.

3.4.2 Isolation of helminth parasites

The fishes were examined for the endoparasites by killing them by the usual method of a blow on the head for any parasite. The parasites belonging to the major groups were separated, identified and counted. Identification was carried out by morphological examination, as described by (Yamaguti, 1958; Soota, 1983 and Retief *et al.*, 2009). The cestodes and acanthocephalans were removed from the host without any form of treatment prior to preservation except that acanthocephalans were relaxed in tap water so that specimens with proboscis fully everted were produced.

In case of acanthocephalans, if the anterior end was deeply bored in the mucosa of the intestine, a few crystals of the methanol were added to the normal saline, containing the parasites adhered to the intestinal wall. This led to immobilization of the parasites & loosening of the grip on the intestinal wall and facilitated the detachment of proboscis in case of acanthocephalans without causing any distortion in the arrangement of hooks.

Cestodes were killed by pouring them in a flask containing saline heated up to 60-80°C and then shaken vigorously for 20 seconds. The scolex of the cestodes were fixed separately on a clean glass slide by placing a cover glass on it while rest of the body was fixed by placing another glass slide, on the previous slide containing the cestode. Weight was put on the slide so that it got fixed and acquired the required transparency so that internal organs could be visualised.

The regular record of the collection was maintained and the prevalence of cestodes and Acanthocephalans were carried out by the following formulas and such Ecological terms are studied as per Margolis *et al.* (1982) and Gudivada *et al.*(2010).

$$\text{Prevalence} = \frac{\text{Total No. of hosts infected}}{\text{Total No. of hosts examined}} \times 100$$

$$\text{Mean intensity} = \frac{\text{Total No. of parasites}}{\text{Total No. of infected hosts examined}}$$

$$\text{Abundance or relative density} = \frac{\text{Total No. of parasites}}{\text{Total No. of hosts examined}}$$

3.5 Metal estimation

3.5.1 Reagents and Solutions

The reagents used in this study are listed below:

1. Stock solution concentration of 1000 μ g/ml each for zinc, copper, iron, nickel, cobalt, aluminium, cadmium, calcium, chromium, manganese, lead and mercury were from (E-Merck, Germany).
2. Nitric acid (HNO₃), concentrated (67-70%), reagent grade minimum, Hydrochloric acid (HCl), concentrated (34-37%), reagent grade minimum and Hydrogen peroxide (H₂O₂) 30%, reagent grade minimum.
3. De-ionized Water
4. 0.45 μ m Whatman filter paper

3.5.2 Safety

Nitric and hydrochloric acid should not be premixed rather added

individually to each sample vessel. Mixtures of nitric and hydrochloric acid must not be stored in closed containers. Slowly and carefully add the acids to water, otherwise can cause a violent exothermic reaction. Perform digestions of samples under a fume hood and wear appropriate PPE (Personal Protective Equipment) including lab coat, gloves, and safety glasses.

3.5.3 Preparation of standard solutions

Standard solutions were prepared from the stock solution (1000 μ g/ml) of each metal. Initial 10 μ g/ml concentration working solutions were prepared from the stock solutions by pipetting 1ml of the 1000 μ g/ml into 100-mL volumetric flasks and diluting to the mark with deionized water. Depending on the linear response range of the metal, the calibration standards were then prepared by appropriate accurate and precise dilution of the 10 μ g/ml working solutions. The calibration standards for all of the metals had a concentration working range of 0.1 to 2.0 ppm.

3.5.4 Instrumentation (Fig. 8)

The absorbance of the calibration standards and samples were measured by Atomic Absorption Spectrophotometer (model, A Analyst 800; Make, Perkin Elmer Ltd) by using an air acetylene flame for the determination of these metals. Kjeldahl flask, were used to gently heat fish samples to near dryness in the digestion process.

3.5.5 Atomic absorption measurement conditions and parameters

Hollow cathode lamps of metals were used to provide the line sources needed for the determination of selected metals. The absorbance of a metal in FAAS depends on the type of flame used, fuel and oxidant ratio, flow rate of fuel gas, and burner height. The measurement conditions for the Perkin Elmer Ltd AA Model 800 used are shown in Table-1. For all samples analyzed, the flame used was air-acetylene (C₂H₂), and the burner angle and support gas flow rate were zero degree and 15.0L/min, respectively.



Fig. 8: Perkin Elmer Ltd AA Model 800 Flame Atomic Absorption Spectrophotometer

Table 1 : Atomic absorption measurement parameters used in this study

Metal gas(L/min)	Wavelength (nm)	Flow rate of fuel (nm)	Slit width (nm)	Burner height(mA)
Copper(Cu)	324.8	1.8	0.7	7.0
Zinc(Zn)	213.9	2.0	0.7	7.0
Cobalt(Co)	345.4	2.0	0.1	7.0
Nickel(Ni)	232.0	1.6	0.2	7.0
Manganese (Mn)	403.1	0.5	0.1	5.0
Chromium(Cr)	425.4	0.4	0.1	7.0
Aluminum(Al)	396.1	1-2	0.1	5-8
Iron(Fe)	248.3	2.2	0.2	9.0
Calcium(Ca)	422.7	1	0.5	10
Cadmium(Cd)	228.8	1.8	0.7	4.0
Lead (Pb)	217.0	2.0	0.7	7.0
Mercury(Hg)	253.7	2.0	0.5	4.0

3.5.6 Sample preparation and treatment

3.5.6.1 Water

Processing of water samples for metal analysis was carried out as per the standard methodology of American Public Health Association (A.P.H.A., 2005). Measured volume of well mixed, acid-preserved sample appropriate for the expected metals concentrations was transferred to a flask or beaker. 5 ml concentrated HNO₃ was added to it and the sample was covered with a ribbed watch glass. The sample was boiled in Kjeldahl flask and then evaporated to 15 to 20 ml. 5 ml concentrated HNO₃ and 10 ml concentrated H₂SO₄ was added cooling flask or beaker between additions. The samples were evaporated on a Kjeldahl apparatus until dense white fumes of SO₃ appear. If solution does not appear clear, 10 ml concentrated HNO₃ was added again and the process repeated until evaporation of SO₃ fumes. The sample was heated to remove all HNO₃ before continuing the treatment. All HNO₃ removed when the solution is clear and no brownish fumes are evident. Sample was not let to be dry during digestion. The sample was cooled and diluted upto 50 ml with deionised water. The finely treated and appropriately concentrated water samples was kept in thoroughly cleaned and air tight glass flasks, before being aspirated directly into the Atomic Absorption Spectrophotometer.

3.5.6.2 Fish tissue and parasites

Fish tissues and parasite were processed for metal analysis (Chernoff, 1975; Abdallah and Moustafa, 2002). Samples were digested by wet digestion method. The tissue sub-samples were oven dried at 105°C until they reached a constant weight (Sures *et al.*, 1995). A mixture of nitric, Perchloric and sulphuric acids in a ratio of 3:1:1 volume was used in a Kjeldahl flask. 10 ml of this mixture was usually sufficed for 10g fresh tissue. Blank reagents without fish samples were also digested using the same method. Digest samples for a minimum of 1 hour at a temperature of 95±5°C. Dilute the entire sample with de-ionized water to

the 100 ml volume. The samples were filtered three more times to remove any small particles that could remain before being analyzed. The samples were then analyzed for heavy metals on Atomic Absorption Spectrophotometer. The metal concentrations in tissues were reported as $\mu\text{g/g}$ dry weights. Dry weight concentrations were converted into wet weight concentrations by following formula using on average 80 % moisture content in fish tissues:

$$\text{Wet weight concentration} = \frac{(\text{Dry weight concentration}) \times (1 - \% \text{ moisture content})}{100}$$

3.5.7 Sample analysis

Standard solutions were prepared from the stock solution of each heavy metal. The absorbance of these solutions were measured using the Perkin Elmer Ltd AA Model 800 flame atomic absorption spectrophotometer. Each sample was aspirated into the nebulizer through a capillary tube where the samples were converted into a fine mist or aerosol. Then, the aspirated sample in aerosol form enters the atomizer.

For each metal, the concentration was determined from each linear calibration curve respectively. All standard solutions for a specific metal were prepared immediately before analysis to avoid adsorption of metals on to the containers and decomposition. The same instrumental conditions were used to run the standard solutions that were used for the samples for each metal. When the absorbance of a sample exceeded that of the highest concentration standard solution of a particular metal, appropriate dilution was made to bring the sample concentration within the linear response range. Triplicate measurements of each standard solution were taken along with triplicate measurements of each water and fish sample. The muscle tissue samples were divided into triplicate aliquots and each triplicate measured three times.

3.6 Pathology

3.6.1 Gross pathology

Fishes were systematically subjected to detailed macroscopic examination with special emphasis on liver, intestine and the lesions were recorded.

3.6.2 Histopathology

Representative tissue samples from the liver, intestine affected by parasites were collected in 10% formalin. The tissue samples were processed for routine paraffin embedding technique and 5 μ thin section were stained with Harris haematoxylin and Eosin (Luna, 1968; Bernet *et al.*, 1999).

3.6.3 Histochemistry

Parallel tissue section selected on the basis of histopathological examination were stained for following histochemical observation.

3.6.3.1 Determination of Acid and Neutral Mucin by combined Alcian blue PAS stain (Bancroft and Gamble, 2002)

Tissue sections were immersed in Alcian Blue (pH=2.5), heated in Microwave at 900 watt power for 45 seconds and allowed to stand in solution for 5 minutes. After washing in running tap water for 5 minutes followed by rinsing in distilled water sections were immersed in 0.5% periodic acid for 5 minutes. The slides were dipped in Schiff's reagent heated in Microwave at 900 watt power for 45 seconds and allowed to stand for 5 minutes. Again the slides were washed in running tap water for 5 minutes followed by rinsing in distilled water. Counter staining was done with haematoxylin differentiation with acid alcohol and bluing with ammonia water. The sections were dehydrated, cleared and were mounted in DPX.

3.6.3.2 Determination of Mast cells by Toluidine Blue Staining Protocol (Gandalfo *et al.*, 2006)

Formalin fixed tissue were cut into paraffin sections of 4 μ . Deparaffinise the tissue and hydrate to distilled water. The slides were dipped in working

Toluidine blue, 1-2 minutes. Rinse the slides with 3 changes of distilled water. Dehydrate the slides quickly through the 95% and Absolute alcohols. The sections were dehydrated, cleared and were mounted in DPX.

3.7 Statistical analysis

SPSS for Windows 17.0 (SPSS Inc.) was used for statistical analysis of the data. One-way ANOVA (Analysis of Variance) was performed for statistically significant difference in the mean value of heavy metal concentrations and physicochemical parameters between the seven sampling sites. Difference in mean values was accepted as being statistically significant if $P < 0.05$. The concentrations of all metals in fish sample are expressed in $\mu\text{g/g}$ and the water concentrations are expressed in $\mu\text{g/L}$. The metal content data being expressed on wet weight basis were compared with the WHO (2004) standards. Post hoc test should be used to interpret the data. One-way analysis of variance (ANOVA) was used to see the significant differences in intensity and abundance within two water bodies.

Non-parametric analysis i.e. Mann-whitney test was used to compare the condition factor of uninfected and infected fishes from the same populations.

$$U = n_1 n_2 + \frac{n_2 (n_2 + 1)}{2} - \sum_{i=r_1+1}^{n_2} R_i$$

Pearson's correlation (r) was used to assess associations between different physico-chemical parameters and parasitic infection. Also, correlation (r) was determined among metal levels in fish tissues, water and different physico-chemical parameters. The mathematical formula for computing r is:

$$r = \frac{n \sum xy - (\sum x)(\sum y)}{\sqrt{n(\sum x^2) - (\sum x)^2} \sqrt{n(\sum y^2) - (\sum y)^2}}$$

Where n is the number of pairs of data.

Chapter -4

EXPERIMENTAL FINDINGS

4.1 Physicochemical parameters

The present study was conducted from September, 2013 to August, 2014 with the aim to record the physico-chemical nature of water at different sites of Dal Lake and River Jhelum in order to assess the changing physical properties, and chemical nature of water. Four sites in Dal Lake and three sites in River Jhelum were selected for the purpose and the samples were collected on seasonal basis. The data is presented in Tables 2 to 5. The present study showed that physico-chemical parameters did not remain stable for prolonged period at a particular place and show fluctuations from region to region, and season to season. Marked differences were observed in the two water bodies during the study period.

4.1.1 Temperature (Table2, Fig. 9)

The average water temperatures (mean±sd) recorded at Dalgate, Saidakadal, Hazratbal and Telbal of Dal Lake were $17.42\pm 8.45^{\circ}\text{C}$, $17.35\pm 7.96^{\circ}\text{C}$, $17.78\pm 8.05^{\circ}\text{C}$ and $16.70\pm 7.65^{\circ}\text{C}$, respectively while as at Chattabal weir, Zerobridge and Khannabal of River Jhelum were $13.09\pm 6.09^{\circ}\text{C}$, $13.41\pm 5.76^{\circ}\text{C}$ and $12.21\pm 5.69^{\circ}\text{C}$, respectively. The temperatures among different locations did not vary significantly ($p>0.05$). The overall mean±SD of temperatures recorded in autumn, winter, spring and summer seasons of different locations were $16.38\pm 2.08^{\circ}\text{C}$, $5.93\pm 1.081^{\circ}\text{C}$, $16.34\pm 2.60^{\circ}\text{C}$ and $23.04\pm 4.05^{\circ}\text{C}$ respectively. At Dalgate, the average water temperatures in autumn, winter, spring and summer were $18.03\pm 4.08^{\circ}\text{C}$, $6.55\pm 1.48^{\circ}\text{C}$, $17.88\pm 2.97^{\circ}\text{C}$ and $27.22\pm 1.71^{\circ}\text{C}$, respectively. The trend in temperature difference during different seasons varied significantly ($p<0.05$). At Hazratbal, the average water temperatures in autumn, winter, spring and summer were $19.24\pm 5.46^{\circ}\text{C}$, $7.04\pm 2.89^{\circ}\text{C}$, $18.27\pm 2.92^{\circ}\text{C}$ and $26.55\pm 2.45^{\circ}\text{C}$, respectively. The temperature during autumn ($p<0.05$) was significantly higher in

comparison to winter and spring ($p>0.05$). At Telbal, the temperatures in autumn, winter, spring and summer were $17.22\pm 2.88^{\circ}\text{C}$, $6.68\pm 2.20^{\circ}\text{C}$, $17.55\pm 3.12^{\circ}\text{C}$ and $25.33\pm 2.34^{\circ}\text{C}$ respectively and the trend of variability in temperatures were significant ($p<0.05$) during all the four seasons with the highest temperature being recorded during summer and the lowest during winter season. At Saidakadal, the average water temperatures in autumn, winter, spring and summer were $17.07\pm 4.18^{\circ}\text{C}$, $6.77\pm 1.85^{\circ}\text{C}$, $19.66\pm 3.60^{\circ}\text{C}$ and $25.88\pm 1.36^{\circ}\text{C}$, respectively. At Zerobridge site, the average water temperatures in autumn, winter, spring and summer were $15.44\pm 2.662^{\circ}\text{C}$, $5.333\pm 1.322^{\circ}\text{C}$, $14.00\pm 3.84^{\circ}\text{C}$ and $18.8\pm 1.26^{\circ}\text{C}$, respectively. At Chattabal weir, the average water temperatures in autumn, winter, spring and summer were $14.16\pm 2.91^{\circ}\text{C}$, $4.77\pm 1.39^{\circ}\text{C}$, $14.0\pm 3.90^{\circ}\text{C}$ and $19.44\pm 1.50^{\circ}\text{C}$, respectively showed a significant variation ($p<0.05$). At Khannabal, the average water temperatures in autumn, winter, spring and summer station were $13.50\pm 3.39^{\circ}\text{C}$, $4.37\pm 1.32^{\circ}\text{C}$, $13.00\pm 3.39^{\circ}\text{C}$ and $18.00\pm 1.32^{\circ}\text{C}$ respectively, showing a significant seasonal variation ($P<0.05$). There was an increasing trend from winter to summer in water temperature in all the sites. The minimum and maximum temperatures recorded in different stations during different season ranged from 4 to 27°C .

4.1.2 Dissolved oxygen (Table3, Fig10)

The average dissolved oxygen concentrations (Mean \pm SD) recorded at Dalgate, Saidakadal, Hazratbal and Telbal of Dal Lake were 4.89 ± 0.864 mg/l, 4.73 ± 0.63 mg/l, 4.68 ± 0.97 mg/l and 6.18 ± 0.68 mg/l, respectively while as at Chattabal weir, Zerobridge and Khannabal of River Jhelum during different seasons were 5.75 ± 0.59 mg/l, 5.71 ± 0.74 mg/l and 6.66 ± 1.02 mg/l, respectively. The difference in the dissolved oxygen concentrations in different sites did not vary significantly ($p>0.05$). At Dalgate, the dissolved oxygen concentrations recorded during autumn, winter, spring and summer were 4.18 ± 0.38 mg/l, 5.87 ± 1.27 mg/l,

Table 2 : Seasonal mean temperatures ($^{\circ}\text{C}$) of the water samples collected from different sites of Dal Lake and River Jhelum

Location	Seasons				Overall Mean	C.D
	Autumn (Mean \pm SD)	Winter (Mean \pm SD)	Spring (Mean \pm SD)	Summer (Mean \pm SD)		
Dal Lake						
Dalgate	18.03 \pm 4.08	6.55 \pm 1.48	17.88 \pm 2.97	27.22 \pm1.71	17.42 \pm 8.45	2.67
Saidakadal	17.07 \pm 4.18	6.77 \pm 1.85	19.66 \pm 3.60	25.88 \pm 1.36	17.35 \pm 7.96	2.88
Hazratbal	19.24 \pm 5.46	7.04 \pm 2.89	18.27 \pm 2.92	26.55 \pm 2.45	17.78 \pm 8.05	3.50
Telbal	17.22 \pm 2.88	6.68 \pm 2.20	17.55 \pm 3.12	25.33 \pm 2.34	16.70 \pm 7.65	2.57
River Jhelum						
ChattabalWeir	14.16 \pm 2.91	4.77 \pm 1.39	14.00 \pm 3.90	19.44 \pm 1.50	13.09 \pm 6.09	2.55
Zerobridge	15.44 \pm 2.66	5.33 \pm 1.32	14.00 \pm 3.84	18.88 \pm 1.26	13.41 \pm 5.76	2.88
Khannabal	13.50 \pm 3.39	4.37 \pm 1.32	13.00 \pm 3.39	18.00 \pm 1.32	12.21 \pm 5.69	2.88
Overall Mean	16.38\pm2.08	5.93 \pm1.08	16.34 \pm2.60	23.04\pm 4.05	15.42 \pm 2.40	

Location = 1.32

CD

Seasons = 1.00

Location x Seasons = 2.64

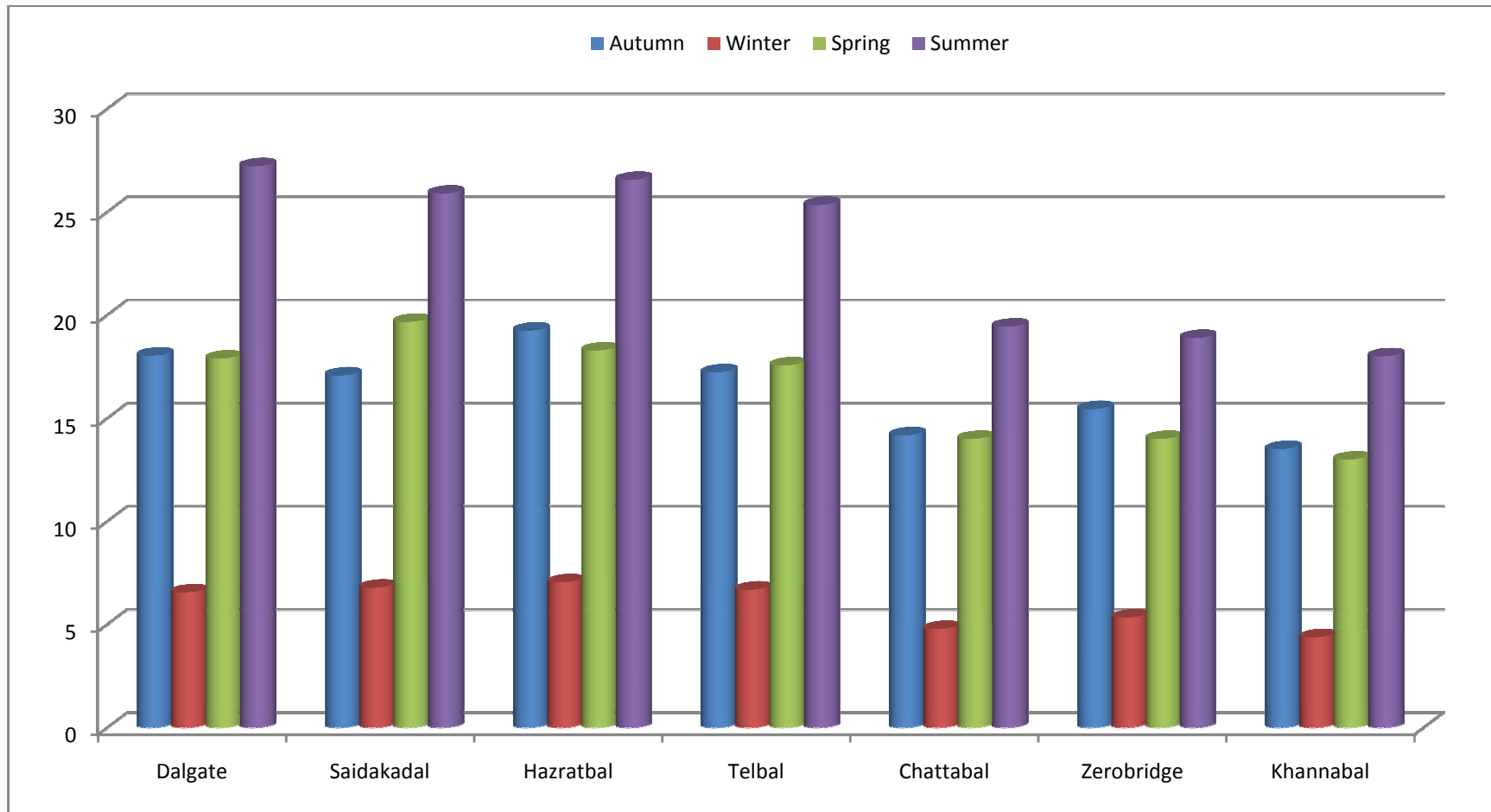


Fig. 9 : Mean seasonal temperature (°C) of water collected from Dal Lake and River Jhelum

Table 3: Seasonal mean dissolved oxygen (mg/l) of the water samples collected from different sites of Dal Lake and River Jhelum

Location	Seasons				Overall Mean	C.D
	Autumn (Mean ± SD)	Winter (Mean ± SD)	Spring (Mean ± SD)	Summer (Mean ± SD)		
Dal Lake						
Dalgate	4.18 ± 0.38	5.87 ± 1.27	5.35 ± 0.73	4.14±1.32	4.89 ±0.86	0.97
Saidakadal	4.21± 1.55	5.57 ± 1.10	4.84 ± 1.02	4.28±1.03	4.73±0.63	N.S.
Hazratbal	4.20 ± 1.22	5.67± 0.88	5.28 ±1.70	3.55±1.19	4.68 ±0.97	1.24
Telbal	5.41 ± 1.38	6.73 ± 1.13	6.78 ± 1.25	5.80±1.15	6.18 ±0.68	N.S.
River Jhelum						
ChattabalWeir	5.58± 1.50	6.16 ± 0.79	6.28 ±0.93	4.98±1.28	5.75 ±0.59	N.S.
Zerobridge	4.97± 1.50	5.76± 0.66	6.72 ±1.44	5.41±1.38	5.71 ±0.74	N.S.
Khannabal	5.86± 1.66	6.26 ± 1.13	8.16 ±1.43	6.37±1.42	6.66 ±1.02	N.S.
Overall Mean	4.92 ± 0.72	6.01 ± 0.40	6.20 ± 1.14	4.93±1.00	5.51±0.78	

Location = 0.57

CD

Seasons = 0.43

Location x Seasons = N.S

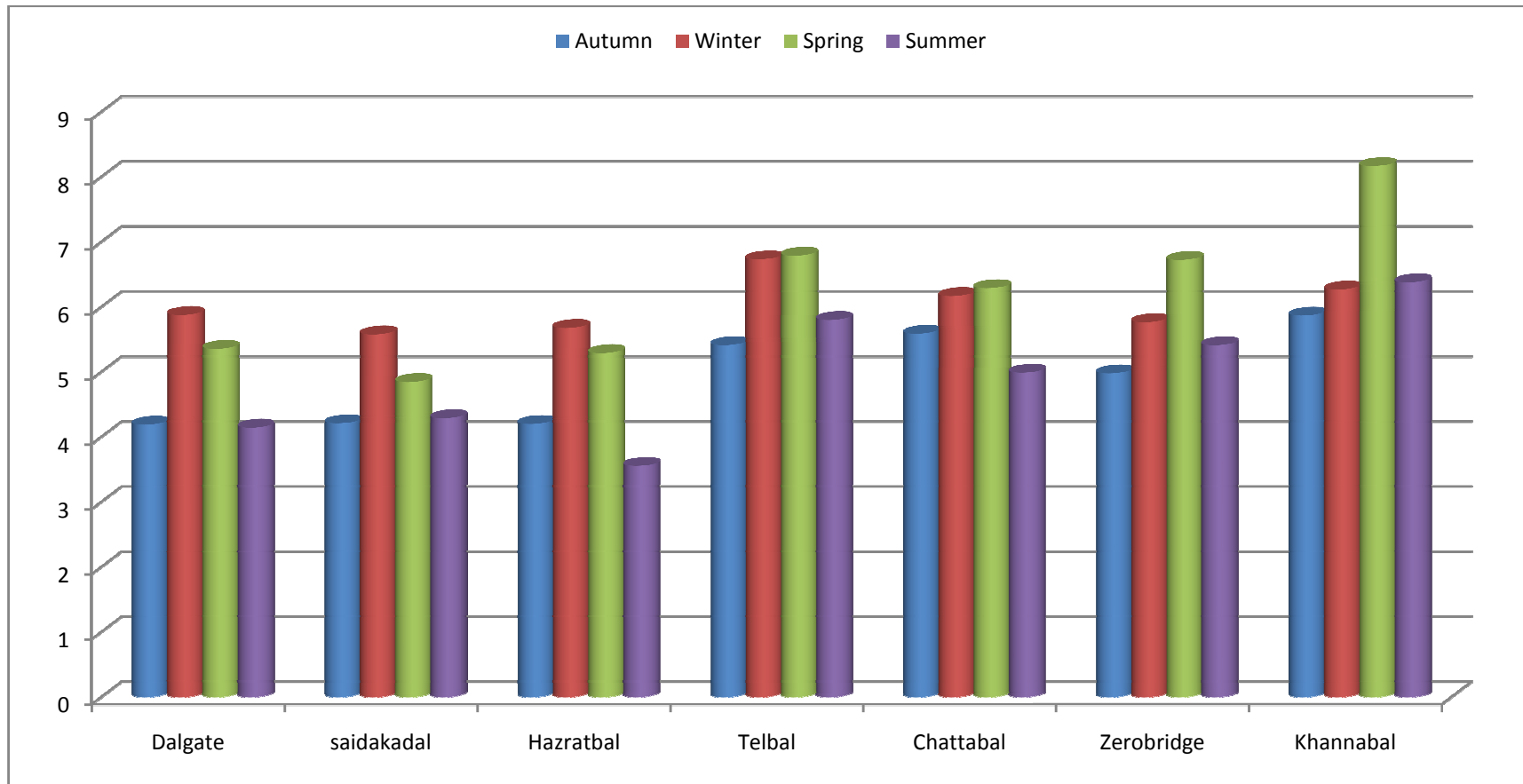


Fig. 10 : Mean seasonal dissolved oxygen (mg/l) content of water collected from Dal Lake and River Jhelum

Table 4 : Seasonal mean pH of the water samples collected from different sites of Dal Lake and River Jhelum

Location	Seasons				Overall Mean	C.D
	Autumn (Mean ± SD)	Winter (Mean ± SD)	Spring (Mean ± SD)	Summer (Mean ± SD)		
Dal Lake						
Dalgate	7.45 ± 0.35	7.87 ± 0.39	7.60±0.26	8.31±0.42	7.81± 0.37	0.35
Saidakadal	7.93 ± 0.50	8.04 ±0.48	8.16±0.36	8.14 ±0.40	8.07± 0.10	N.S.
Hazratbal	8.20 ± 0.61	8.23 ±0.34	8.42 ±0.43	8.47± 0.41	8.33 ± 0.13	N.S.
Telbal	7.63 ± 0.43	7.83 ±0.25	7.65± 0.44	7.86± 0.35	7.74 ± 0.12	N.S.
River Jhelum						
ChattabalWeir	7.03 ± 0.28	7.49 ± 0.40	7.67 ±0.32	7.87 ±0.30	7.52 ± 0.35	0.48
Zerobridge	7.20 ± 0.59	7.97 ± 0.43	8.10 ±0.48	7.85 ±0.41	7.78 ± 0.40	0.51
Khannabal	7.84 ± 0.52	7.94 ± 0.80	8.35±0.51	8.07±0.40	8.05 ± 0.22	N.S.
Overall Mean	7.61 ± 0.41	7.91 ± 0.22	7.99± 0.34	8.08 ±0.24	7.90±0.24	

Location = 0.20

CD

Seasons = 0.15

Location x Seasons = 0.41

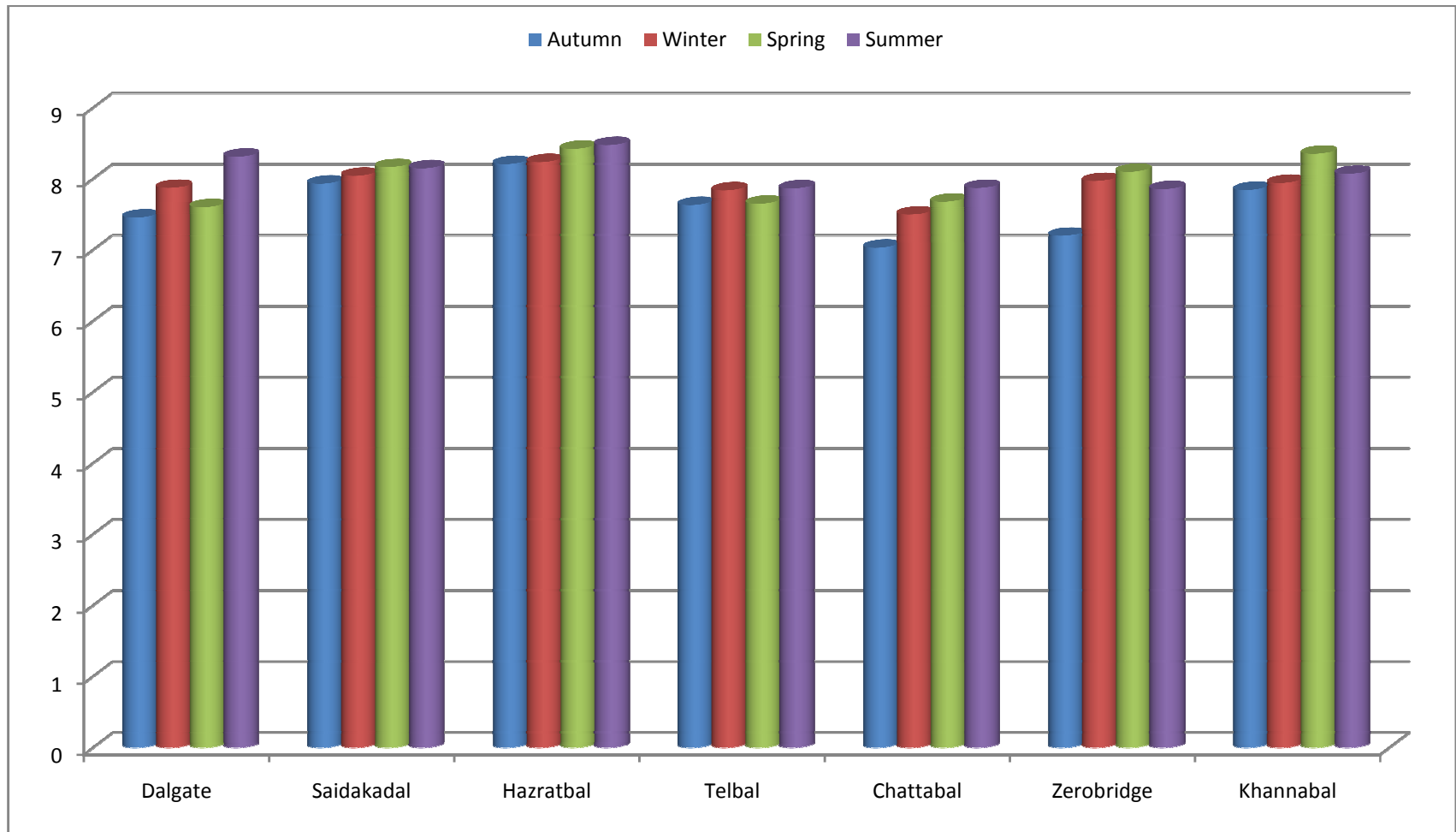


Fig. 11 : Mean seasonal pH of water collected from Dal Lake and River Jhelum

Table 5 : Seasonal mean free carbon dioxide (mg/l) of the water samples collected from different sites of Dal Lake and River Jhelum

Location	Seasons				Overall Mean	C.D
	Autumn (Mean ± SD)	Winter (Mean ± SD)	Spring (Mean ± SD)	Summer (Mean ± SD)		
Dal Lake						
Dalgate	3.94± 0.72	3.38 ±1.39	7.11±1.69	6.88±3.37	5.33± 1.94	1.97
Saidakadal	3.16± 0.93	5.55 ±1.42	9.50±3.33	7.55 ±7.46	6.44± 2.71	4.03
Hazratbal	5.77 ± 1.20	6.43 ±1.31	5.10 ±3.84	3.55 ±1.23	5.21 ± 1.23	N.S.
Telbal	3.83 ± 1.00	5.22± 1.48	12.88±7.21	5.44 ±2.69	6.84± 4.09	3.81
River Jhelum						
ChattabalWeir	3.08 ±1.89	4.61 ±2.52	7.33 ±1.41	8.11 ±2.26	5.78 ± 2.34	2.19
Zerobridge	3.27 ± 0.90	3.55 ±1.57	4.84± 1.00	8.05 ±6.28	4.93 ± 2.19	3.32
Khannabal	3.55 ± 1.33	3.92 ± 0.95	5.88± 3.68	8.44 ±5.43	5.45 ± 2.24	3.40
Overall Mean	3.80 ± 0.92	4.67 ±1.12	7.52 ± 2.84	6.86±1.77	5.71±2.39	

Location = N.S

CD

Seasons = 1.10

Location x Seasons = 2.91

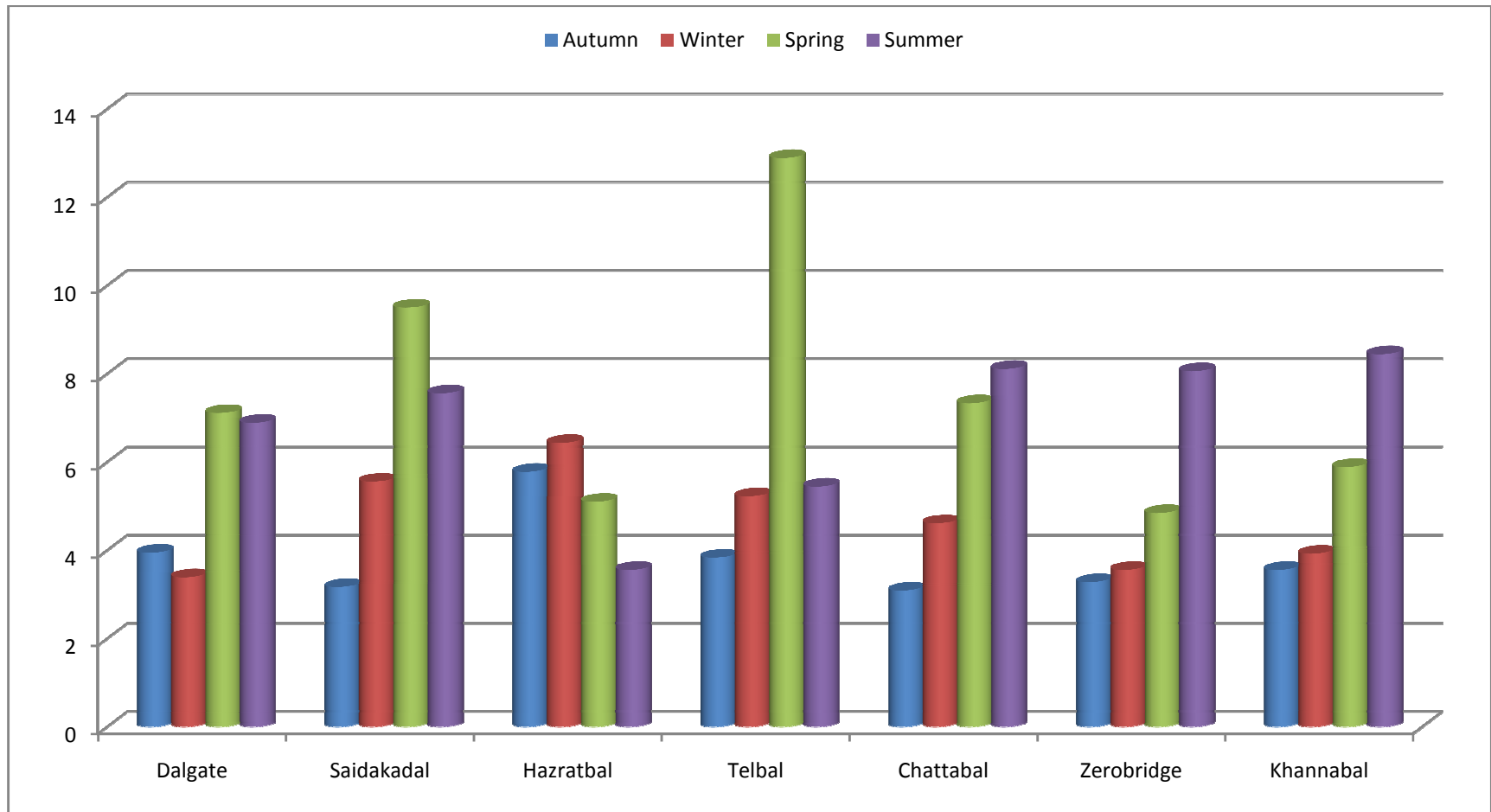


Fig. 12 : Mean seasonal free carbon dioxide (mg/l) content of water collected from Dal Lake and River Jhelum

5.35±0.73 mg/l and 4.14±1.32 mg/l, respectively. There were a significant variation ($p<0.05$) in dissolved oxygen concentrations during different seasons with the highest concentration during spring, followed by winter, autumn and summer. At Hazratbal, the dissolved oxygen concentrations during autumn, winter, spring and summer were 4.20±1.22 mg/l, 5.67±0.88 mg/l, 5.28±1.70 mg/l and 3.55±1.19 mg/l, respectively. A similar trend of variability in dissolved oxygen concentrations were observed at Hazratbal to that of Dalgate and was highly significant ($p<0.05$). At Telbal, the dissolved oxygen concentrations during autumn, winter, spring and summer were 5.41±1.38 mg/l, 6.73±1.13 mg/l, 6.78±1.25 mg/l and 5.80±1.15 mg/l. At Saidakadal, the dissolved oxygen concentration during autumn, winter, spring and summer were 4.21±1.55 mg/l, 5.57±1.10 mg/l, 4.84±1.02 mg/l and 4.28±1.03 mg/l, respectively. At Zerobridge, the dissolved oxygen concentrations during autumn, winter, spring and summer were 4.97±1.50 mg/l, 5.76±0.66 mg/l, 6.72±1.44 mg/l and 5.41±1.38 mg/l, respectively ($p>0.05$). At Chattabal weir, the dissolved oxygen concentrations during autumn, winter, spring and summer were 5.58±1.50 mg/l, 6.16±0.79 mg/l, 6.28±0.93 mg/l and 4.98±1.28 mg/l, respectively ($p>0.05$). At Khannabal, the dissolved oxygen concentrations during autumn, winter, spring and summer were 5.86±1.66 mg/l, 6.26±1.13 mg/l, 8.16±1.43 mg/l and 6.37±1.42 mg/l, respectively ($p>0.05$). The dissolved oxygen concentrations during autumn and summer were significantly ($p<0.05$) lower than those in spring and winter season.

4.1.3 pH (Table 4, Fig 11)

The average pH (mean±sd) recorded at Dalgate, Saidakadal, Hazratbal and Telbal of Dal Lake were 7.81±0.37, 8.07±0.10, 8.33±0.13 and 7.74±0.12 respectively, while as at Chattabal weir, Zerobridge and Khannabal of River Jhelum were 7.52±0.35, 7.78±0.40 and 8.05±0.22, respectively. At Dalgate, the average pH value during autumn, winter, spring and summer were 7.456 ±0.350, 7.874±0.398, 7.604±0.264 and 8.313±0.423, respectively. The pH value was highest during summer and lowest during autumn. At Hazratbal, the pH value were

with slight difference during summer (8.47 ± 0.41), spring (8.42 ± 0.43), winter (8.23 ± 0.34) and autumn season (8.20 ± 0.61). At Telbal during autumn, winter, spring and summer were 7.63 ± 0.43 , 7.83 ± 0.25 , 7.65 ± 0.44 and 7.86 ± 0.35 , respectively ($p>0.05$). At Saidakadal the average pH recorded during autumn, winter, spring and summer were 7.93 ± 0.50 , 8.04 ± 0.48 , 8.16 ± 0.36 and 8.14 ± 0.40 respectively ($p>0.05$). At Chattabal weir, the pH value was highest during summer (7.87 ± 0.30) followed by spring (7.67 ± 0.32) and winter season (7.49 ± 0.40), whereas it was significantly ($P<0.05$) lower during autumn season (7.03 ± 0.28). At Zerobridge the average pH recorded were 7.20 ± 0.59 , 7.97 ± 0.43 , 8.10 ± 0.48 and 7.85 ± 0.41 respectively ($p<0.05$). At Khannabal the average pH recorded were 7.84 ± 0.52 , 7.94 ± 0.80 , 8.35 ± 0.51 and 8.07 ± 0.40 respectively ($p>0.05$). pH was alkaline ranges from 7.5 to 8.4. The maximum pH value recorded was 8.0 during summer and minimum was 7.6 during autumn. Most of bio-chemical and chemical reactions are influenced by the pH.

4.1.4 Free carbon dioxide (Table5, Fig. 12)

The average free carbon dioxide concentrations (mean \pm sd) recorded at Dalgate, Saidakadal, Hazratbal and Telbal of Dal Lake were 5.33 ± 1.94 mg/l, 6.44 ± 2.71 mg/l, 5.21 ± 1.23 mg/l, 6.84 ± 4.09 mg/l, respectively while as at Chattabal weir, Zerobridge and Khannabal of River Jhelum were 5.78 ± 2.34 mg/l, 4.93 ± 2.19 mg/l, 5.45 ± 2.24 mg/l, respectively during different seasons were non-significant ($p>0.05$). The free carbon dioxide contents were significantly ($P<0.05$) lower during autumn followed by winter, summer and spring season in all the locations. The average free carbon dioxide concentrations of water samples collected from different stations during autumn, winter, spring and summer were 3.80 ± 0.92 mg/l, 4.67 ± 1.12 mg/l, 7.52 ± 2.84 mg/l and 6.86 ± 1.775 mg/l, respectively, were significant ($p<0.05$). During autumn, winter, spring and summer seasons the free carbon dioxide concentrations recorded at Dalgate were 3.944 ± 0.726 mg/l, 3.389 ± 1.399 mg/l, 7.11 ± 1.69 mg/l and 6.88 ± 3.37 mg/l, at Hazratbal 5.77 ± 1.20 mg/l, 6.43 ± 1.31 mg/l, 5.10 ± 3.84 mg/l and 3.55 ± 1.23 mg/l,

at Telbal 3.83±1.00 mg/l, 5.22±1.48 mg/l, 12.8±7.21 mg/l and 5.44±2.69 mg/l, at Saidakadal 3.16±0.93 mg/l, 5.55±1.42 mg/l, 9.50±3.33 mg/l and 7.55±7.46 mg/l, at Zerobridge 3.27±0.90 mg/l, 3.55 ±1.57 mg/l, 4.84±1.00 mg/l and 8.05 ±6.28 mg/l, at Chattabal weir 3.08±1.83 mg/l, 4.61±2.52 mg/l, 7.33±1.41 mg/l and 8.11±2.26 mg/l and at Khannabal were 3.55±1.33 mg/l, 3.92±0.95 mg/l, 5.88±3.68 mg/l and 8.44±5.43 mg/l, respectively. The study sites vary significantly in their free carbon dioxide concentrations seasonally except Hazratbal station ($p>0.05$). The minimum free carbon dioxide concentration was 3.80±0.92 mg/l during autumn whereas the maximum concentration was 7.52±2.84 mg/l during spring season.

4.2 Distribution of *Schizothorax* spp.

Out of 224 *Schizothorax* spp. recovered from different locations of Dal Lake 76 were of *S.niger*, 77 of *S. esocinus* and 71 of *S. curvifrons* while out of 220 species recovered from River Jhelum 76 were of *S. niger*, 73 of *S. esocinus* and 71 of *S. curvifrons*. Out of 152 *S. niger* recovered from both the locations males and females were recovered in equal numbers of 76 each. Similarly, out of 150 *S. esocinus* 77 were males and 73 were females and for *S. curvifrons* 89 were males and 53 were females.

4.2.1 Prevalence of helminth parasite in *Schizothorax* spp.

A total of three helminth parasitic species were recovered from *Schizothorax* spp. These include acanthocephalan parasite (*Pomphorhynchus kashmirensis*) (27.47%) and two intestinal cestodes (*Bothriocephalus acheilognathi*) (30.63%) and cestode (*Adenoscolex oreini*) (32.43%). The host wise, season wise, gender wise prevalences of the parasites are given in Tables 6 to 14 and Figs. 13 to 30.

4.2.2 Prevalence of *Pomphorhynchus kashmirensis* in Dal Lake and River Jhelum

4.2.2.1 Host-wise Prevalence (Table 6 and Fig. 13&14)

Overall out of 444 fish specimens examined 122 (27.47%) were found to harbour the *Pomphorhynchus kashmirensis*. Out of 224 specimens examined from the Dal Lake only 47 specimens were found infected with *Pomphorhynchus kashmirensis* (20.98%). Host-wise distribution of the parasite was significantly varied ($p < 0.01$) as *S. niger*, *S. esocinus* and *S. curvifrons* (27.63, 18.18 and 16.90%), respectively. Out of 220 *Schizothorax* spp. examined from River Jhelum only 75 (34.07%) were infected with *Pomphorhynchus kashmirensis* which include *S. niger* (30.20%), *S. esocinus* (30.13%) and *S. curvifrons* (42.25%).

The mean intensity of *Pomphorhynchus kashmirensis* in *S. niger*, *S. esocinus* and *S. curvifrons* of Dal Lake were 1.76, 2.0 and 1.75 having an abundance 0.48, 0.36 and 0.29, respectively. Whereas it was 1.30, 2.09 and 1.63 in the respective fishes of Jhelum River which had an abundance of 0.39, 0.63 and 0.69 respectively. The overall mean intensities of *Pomphorhynchus kashmirensis* in *Schizothorax* spp. of Dal Lake and River Jhelum was 2.04 and 1.66 having an abundance of 0.42 and 0.56 respectively.

4.2.2.2 Seasonal prevalence (Table 7 and Fig. 15 & 16)

Seasonal prevalence of *Pomphorhynchus kashmirensis* showed a definite trend. The infection was highest in summer and lowest in winter. There was a gradual increase in the prevalence rate from spring to summer which fell down with onset of autumn and later on was least observed during winter season. In Dal Lake during summer the overall prevalence of *Pomphorhynchus kashmirensis* infection was highest (29.6%) and the species-wise prevalences were *S. niger* (38.8%), *S. esocinus* (29.4%) and *S. curvifrons* (21.05%) while as during winter it was least (8.47%) with species-wise distribution as *S. niger* (10.5%), *S. esocinus* (9.09%) and *S. curvifrons* (5.55%). Contrary to this in River Jhelum during summer the prevalence was highest (51.02%) with species-wise pattern as *S.*

niger(50%), *S. esocinus* (44.4%) and *S. curvifrons* (60%) and the least prevalence (14.2%) was found during the winter season with species-wise distribution as *S. niger*(12%),*S. esocinus*(12.5%) and *S. curvifrons*(20%).

The mean intensities of the parasite during summer were 1.28, 2.6 and 1.50 in *S. niger*, *S. esocinus* and *S. curvifrons*, respectively with the abundance of the parasites as 0.50, 0.76 and 0.31 for the respective fishes in Dal Lake. Whereas during winter the mean intensities were 5.5, 1.50 and 2.0 with abundance of 0.57, 0.13 and 0.11 in the corresponding fishes. In River Jhelum the mean intensities of the parasite during summer were 0.87, 1.87 and 2.0 in *S. niger*, *S. esocinus* and *S. curvifrons* respectively with the abundance of the parasites as 0.43, 0.83 and 1.20. Whereas during winter the mean intensities were 1.33, 5.5 and 3.6 with abundance of 0.16, 0.68 and 0.73 in the corresponding fishes.

4.2.2.3 Gender wise prevalence of *Pomphorhynchus kashmirensis* (Table 8 and Fig. 17 &18)

Gender-wise distribution of *Pomphorhynchus kashmirensis* in *Schizothorax* spp. of Dal Lake revealed that the overall prevalence of the parasite in males was 23.1% (*S. niger*, 37.2%; *S. esocinus*, 20.5% and *S. curvifrons*,13.46%) whereas it was 17.7% in females (*S. niger*, 15.5%; *S. esocinus*, 15.7% and *S. curvifrons*, 26.3%). In River Jhelum the overall prevalence in males was 35.8% (*S. niger*, 36.6%; *S. esocinus*, 29.78% and *S. curvifrons*, 43.24%) while as in females it was 32.03% (*S. niger*, 25.58%;*S. esocinus*, 25.58% and *S. curvifrons*,41.17%).

The overall mean intensities of the parasite in males and females of *Schizothorax* spp. were 1.8 and 2.3, with an abundance of 0.43 and 0.42, respectively, in Dal Lake. In River Jhelum the overall mean intensities of the parasite in males and females of *Schizothorax* spp. were 1.9 and 1.36 with an abundance of 0.68 and 0.43, respectively.

Table 6: Overall prevalence of *Pomphorhynchus kashmirensis* in various host species

Host	Dal Lake							River Jhelum						
	No. Examined	No. infected	Prevalence (%)	No. of parasites	Mean Intensity	Abundance	P-value	No. Examined	No. infected	Prevalence (%)	No. of parasites	Mean Intensity	Abundance	P-value
<i>S. niger</i>	76	21	27.63	47	1.76	0.48	< 0.01	76	23	30.2	30	1.30	0.39	< 0.01
<i>S. esocinus</i>	77	14	18.18	28	2.0	0.36	< 0.01	73	22	30.13	46	2.09	0.63	< 0.01
<i>S. curvifrons</i>	71	12	16.9	21	1.75	0.29	< 0.01	71	30	42.25	49	1.63	0.69	> 0.05
Total	224	47	20.98	96	2.04	0.42	< 0.01	220	75	34.09	125	1.66	0.56	< 0.01

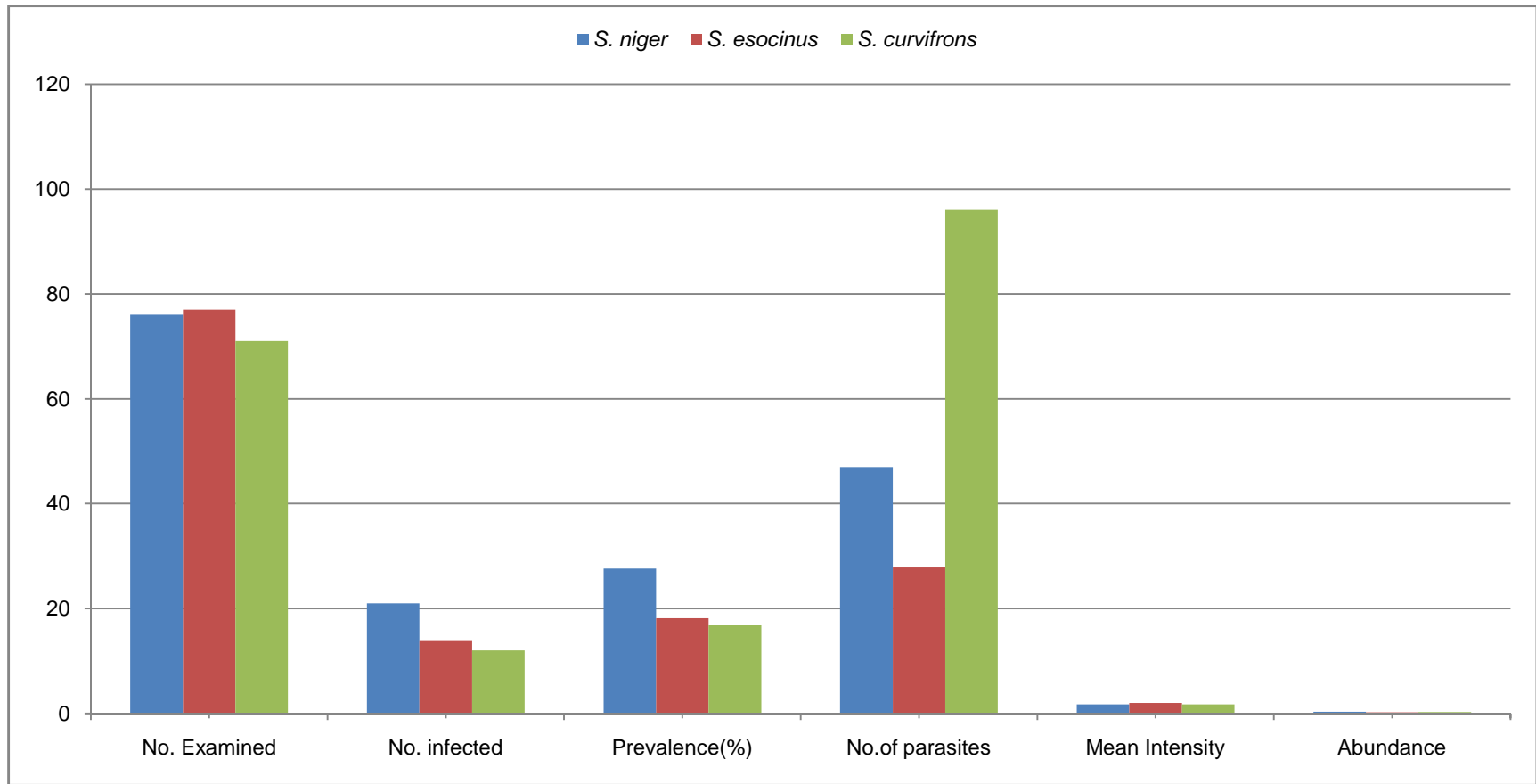


Fig. 13 : Prevalance of *Pomphorhynchus kashmirensis* in various host species of Dal Lake

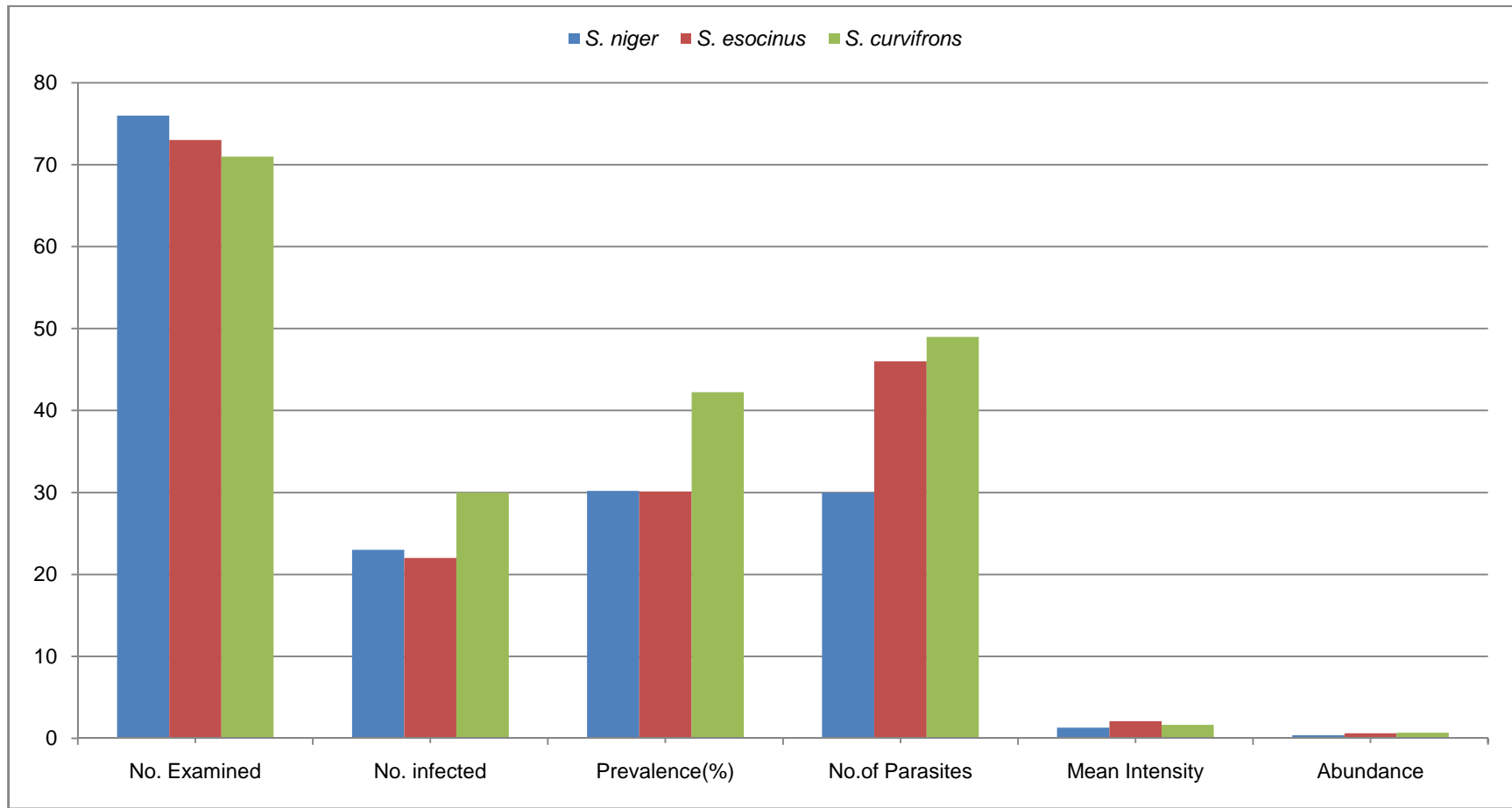


Fig. 14: Prevalence of *Pomphorhynchus kashmirensis* in various host species of River Jehlum

Table 7: Infection dynamics of *Pomphorhynchus kashmirensis* recorded of *Schizothorax* spp. from Dal Lake and River Jehlum

Season	Host	Dal Lake							River Jehlum						
		No. Examined	No. infected	Prevalence (%)	No. of parasites	Mean Intensity	Abundance	P-value	No. Examined	No. infected	Prevalence (%)	No. of parasites	Mean Intensity	Abundance	P-value
Spring	<i>S. niger</i>	14	6	42.85	14	2.33	1	>0.05	15	4	26.6	12	3	0.8	>0.05
	<i>S. esocinus</i>	19	3	15.78	3	1	0.05	<0.01	20	5	25	8	1.6	0.4	<0.05
	<i>S. curvifrons</i>	16	2	12.5	6	3	0.18	<0.01	19	9	47.3	6	0.6	0.31	>0.05
Summer	<i>S. niger</i>	18	7	38.8	9	1.28	0.5	>0.05	16	8	50	7	0.87	0.43	>0.05
	<i>S. esocinus</i>	17	5	29.4	13	2.6	0.76	>0.05	18	8	44.4	15	1.87	0.83	>0.05
	<i>S. curvifrons</i>	19	4	21.05	6	1.5	0.31	<0.05	15	9	60	18	2	1.2	>0.05
Autumn	<i>S. niger</i>	25	6	24	13	2.16	0.52	<0.05	20	8	40	7	0.87	0.35	>0.05
	<i>S. esocinus</i>	19	4	21.05	9	2.25	0.47	<0.05	19	7	36.84	12	1.71	0.63	>0.05
	<i>S. curvifrons</i>	18	5	27.7	7	1.4	0.38	>0.05	22	9	40.9	14	1.55	0.63	>0.05
Winter	<i>S. niger</i>	19	2	10.5	11	5.5	0.57	<0.01	25	3	12	4	1.33	0.16	<0.01
	<i>S. esocinus</i>	22	2	9.09	3	1.5	0.13	<0.01	16	2	12.5	11	5.5	0.68	<0.01
	<i>S. curvifrons</i>	18	1	5.55	2	2	0.11	<0.01	15	3	20	11	3.6	0.73	<0.05
	Total	224	47	20.98	96	2.04	0.42	<0.01	220	75	34.09	125	1.66	0.56	<0.01

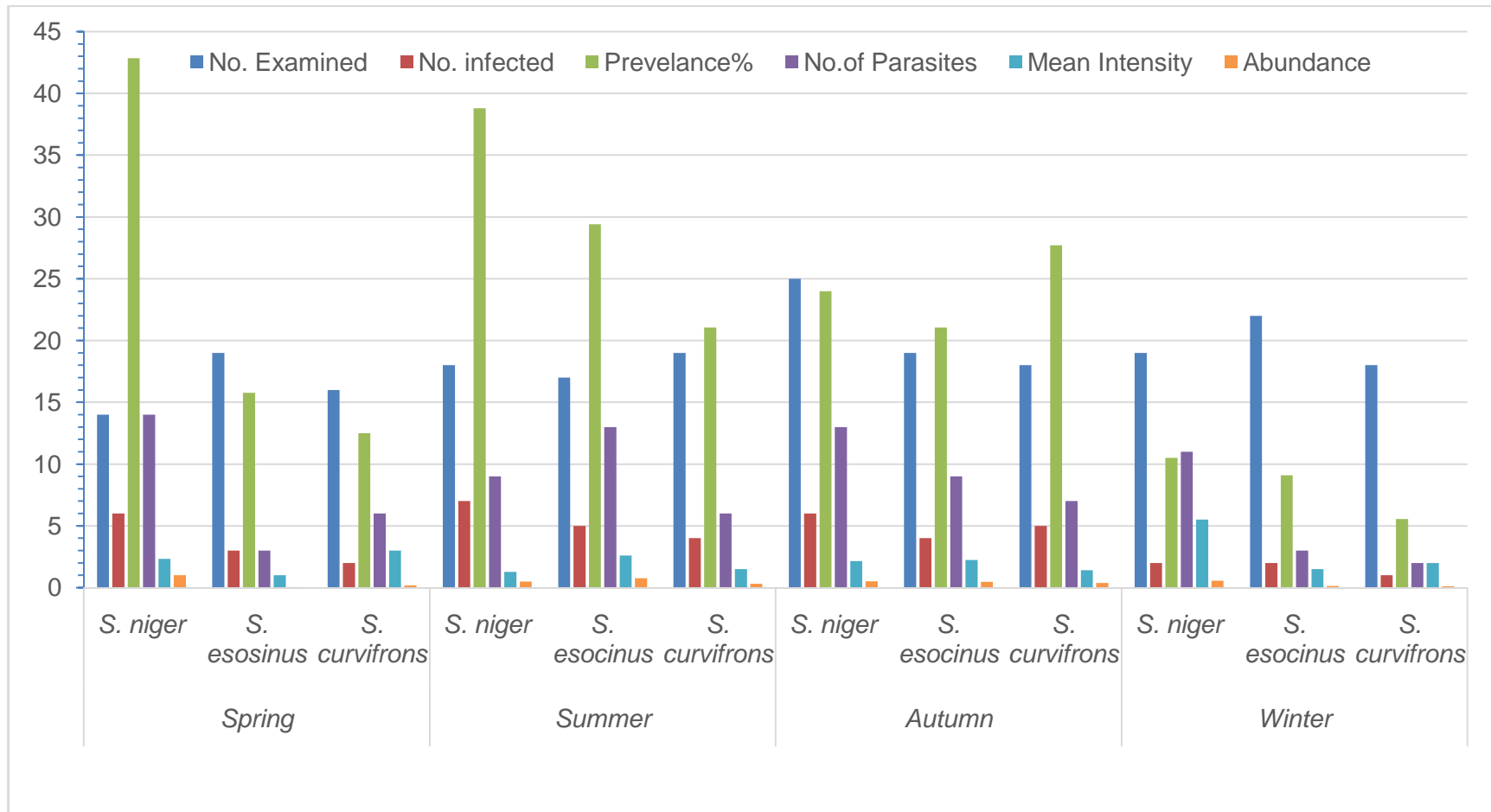


Fig. 15 : Seasonal prevalence of *Pomphorhynchus kashmirensis* of *Schizothorax* sp from Dal Lake

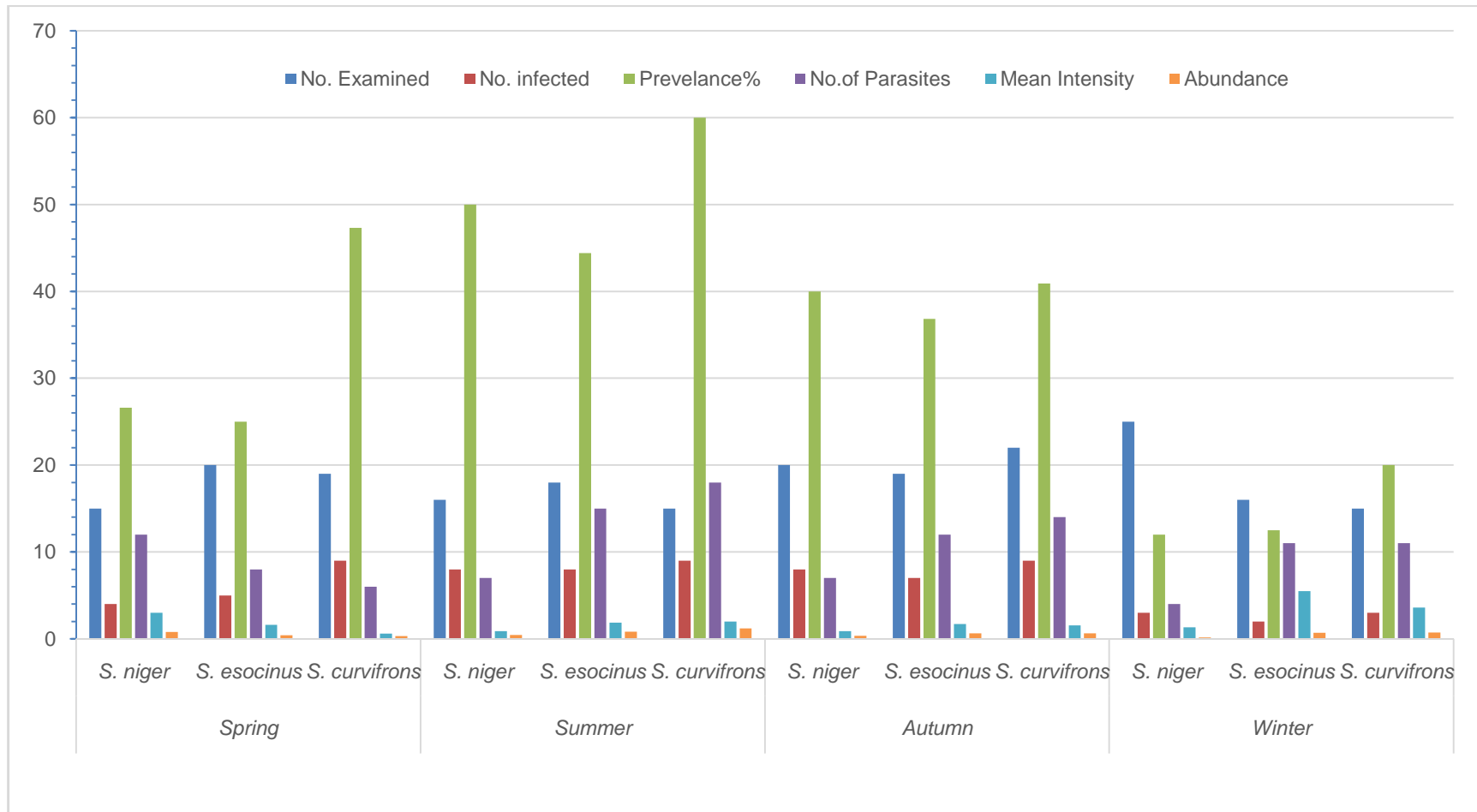


Fig. 16 : Seasonal prevalence of *Pomphorhynchus kashmirensis* of *Schizothorax* spp. from River Jhelum

Table 8: Genderwise infection dynamics of *Pomphorhynchus kashmirensis* recorded of *Schizothorax* spp. from Dal Lake and River Jehlum

Season	Host	Dal Lake							River Jehlum						
		No. Examined	No. infected	Prevalence (%)	No. of parasites	Mean Intensity	Abundance	P-value	No. Examined	No. infected	Prevalence (%)	No. of parasites	Mean Intensity	Abundance	P-value
<i>S. niger</i>	Male	43	16	37.2	28	1.75	0.65	<0.05	33	12	36.36	22	1.83	0.66	<0.05
	Female	33	5	15.5	19	3.8	0.57		43	11	25.58	8	0.72	0.18	
<i>S. esocinus</i>	Male	39	8	20.5	15	1.8	0.38	>0.05	47	14	29.78	26	1.8	0.55	>0.05
	Female	38	6	15.7	13	2.16	0.34		26	8	30.76	20	2.5	0.76	
<i>S. curvifrons</i>	Male	52	7	13.46	15	2.14	0.28	>0.05	37	16	43.24	32	2	0.86	>0.05
	Female	19	5	26.3	6	1.2	0.31		34	14	41.17	17	1.2	0.5	

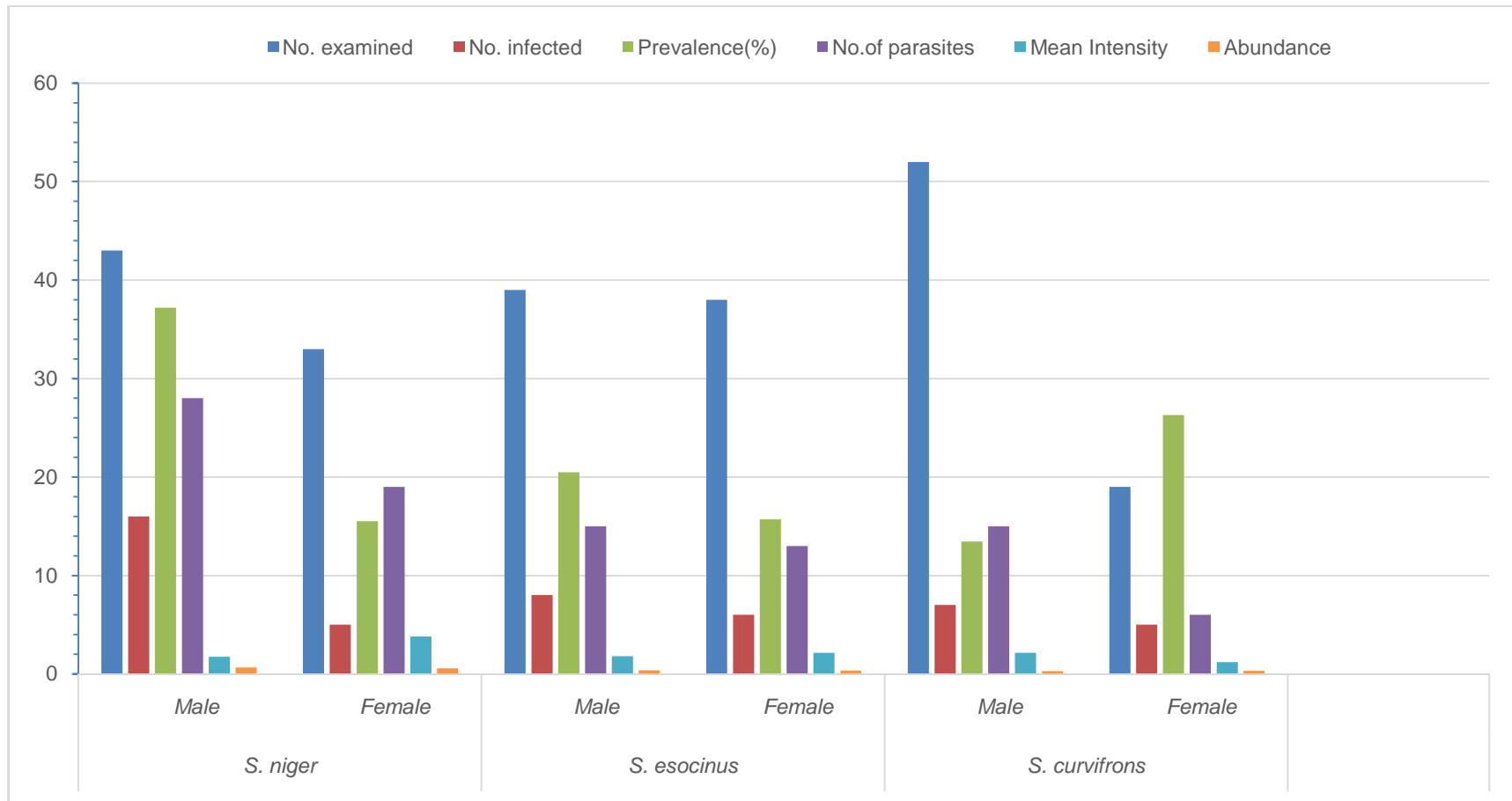


Fig. 17 : Genderwise prevalence of *Pomphorhynchus kashmirensis* in *Schizothorax* spp. from Dal Lake

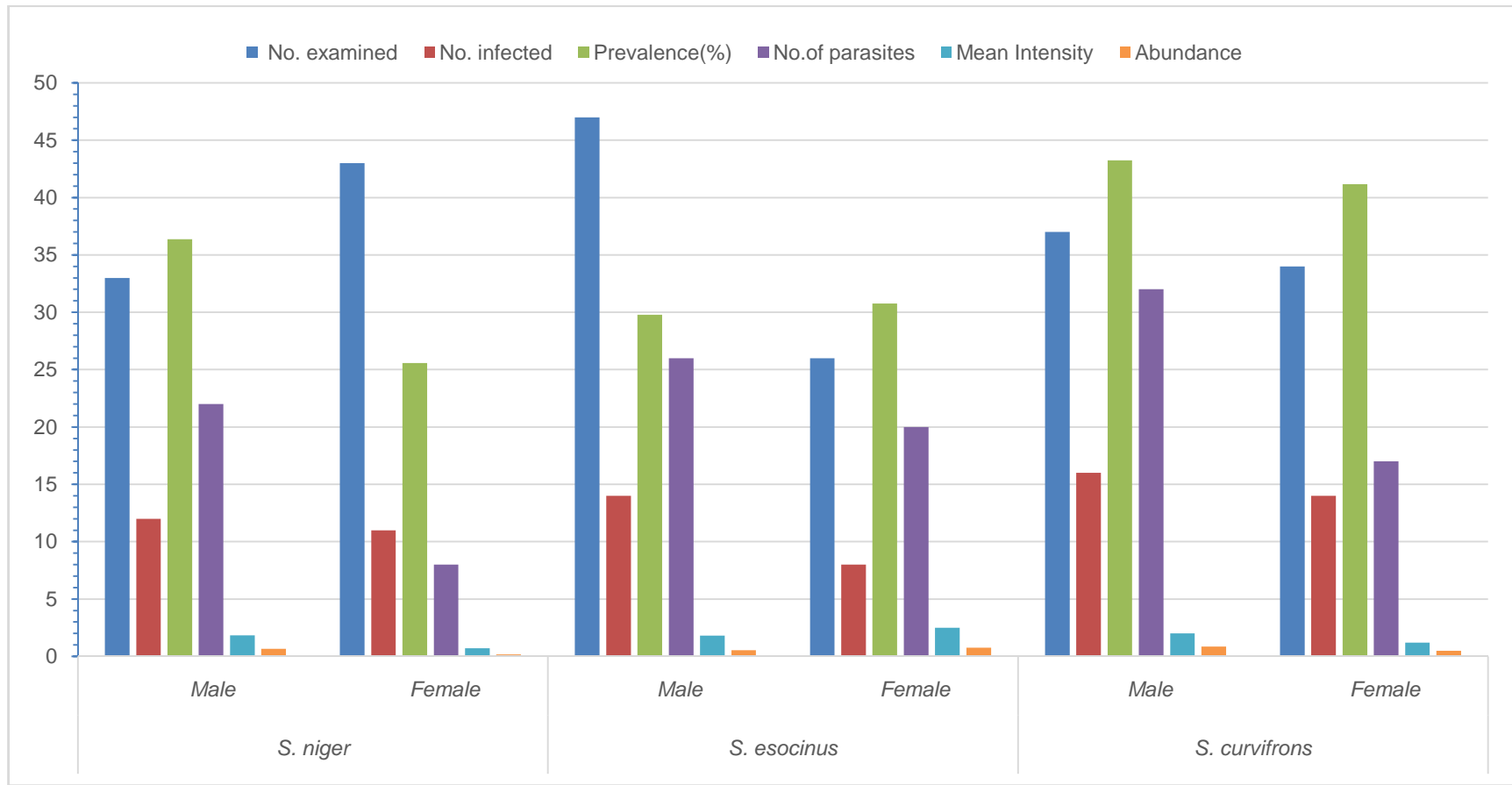


Fig. 18 : Genderwise prevalence of *Pomphorhynchus kashmirensis* of *Schizothorax* sp. from River Jehlum

4.2.3 Prevalence of *Bothriocephalus acheilognathi* in Dal Lake and River Jhelum

4.2.3.1 Host-wise prevalence (Table 9 and Fig. 19&20)

Out of 444 fish specimens of *Schizothorax* species 136 (30.63%) were found to harbor the *Bothriocephalus acheilognathi* parasite. Out of 224 fishes examined from the Dal Lake only 63 (28.14%) were found infected with *Bothriocephalus acheilognathi*. Host-wise distribution of the parasite was highly significant ($p < 0.01$) which showed *S. niger* (28.94%), *S. esocinus* (31.16%) and *S. curvifrons* (23.94%). Out of 220 *Schizothorax* spp. from River Jhelum 73 (33.18%) were infected with the *Bothriocephalus acheilognathi* which include *S. niger* (34.21%), *S. esocinus* (34.24%) and *S. curvifrons* (30.98%).

The mean intensity of *Bothriocephalus acheilognathi* in *S. niger*, *S. esocinus* and *S. curvifrons* of Dal Lake was 6.9, 5.7 and 4.7 with an abundance of 2.0, 1.8 and 1.1, respectively. Whereas in respective fishes of river Jhelum it were 7.7, 5.7 and 7 having abundance of 2.6, 1.9 and 2.1, respectively. The overall mean intensities of *Bothriocephalus acheilognathi* in *Schizothorax* spp. of Dal Lake and River Jhelum were 5.9 and 6.8 with an abundance of 1.6 and 2.2 respectively.

4.2.3.2 Seasonal Prevalence (Table 10 and Fig. 21&22)

Bothriocephalus acheilognathi showed a definite trend in the seasonal prevalence with highest infection in summer and lowest in winter. There was a gradual increase in the prevalence rate from spring to summer which fell down with onset of autumn and least observed prevalence was observed during winter season. In Dal Lake during summer the overall prevalence of *Bothriocephalus acheilognathi* infection was highest (31.4%) and the species-wise prevalences were *S. niger* (33.3%), *S. esocinus* (41.1%) and *S. curvifrons* (21.0%) while as during winter it was least (18.6%) with species-wise distribution as *S. niger* (21.05%), *S. esocinus* (22.7%) and *S. curvifrons* (11.11%). Contrary to this in River Jhelum during summer the prevalence was highest (46.9%) with species-

wise pattern as *S. niger* (56.25%), *S. esocinus* (44.44%) and *S. curvifrons* (40.0%) and the least prevalence (17.85%) was found during the winter season with species-wise distribution as *S. niger* (16.0%), *S. esocinus* (18.7%) and *S. curvifrons* (20.0%).

The mean intensities of the parasite in Dal Lake during summer were 6.3, 1.2 and 3 in *S. niger*, *S. esocinus* and *S. curvifrons*, respectively, with the abundance of the parasites as 2.1, 0.5 and 0.6 for the respective fishes. Whereas during winter the mean intensities were 7.2, 7.8 and 4.5 with abundance of 1.5, 1.7 and 0.5 in the corresponding fishes. In River Jhelum the mean intensities of the parasite during summer were 9.1, 4.5 and 4.6 in *S. niger*, *S. esocinus* and *S. curvifrons* respectively with the abundance of the parasites as 5.1, 2 and 1.8. Whereas during winter the overall mean intensities were 2.2, 4.0 and 6.0 with abundance of 0.3, 0.7 and 1.2 in the corresponding fishes.

4.2.3.3 Gender-wise prevalence of *Bothriocephalus acheilognathi* (Table 11 and Fig. 23&24)

Gender-wise distribution of *Bothriocephalus acheilognathi* in *Schizothorax* spp. of Dal Lake revealed that the overall prevalence of the parasite in males was 31.34% (*S. niger* 30.23%, *S. esocinus* 41.0% and *S. curvifrons* 25.0%) whereas it was 23.3% in females (*S. niger* 27.27%, *S. esocinus* 21.0% and *S. curvifrons* 21.05%). In River Jhelum the overall prevalence in males was 41.02% (*S. niger* 51.5%, *S. esocinus* 40.4% and *S. curvifrons* 32.4%) while as in females it was 24.29% (*S. niger* 29.9%, *S. esocinus* 23.07% and *S. curvifrons* 29.4%).

The overall mean intensities of the parasite in males and females of *Schizothorax* spp. were 5.9 and 5.7, with an abundance of 1.8 and 1.3 respectively in Dal Lake. In river Jhelum the overall mean intensities of the parasite in males and females of *Schizothorax* spp. were 6.7 and 7.12 with an abundance of 2.7 and 1.7, respectively.

Table 9: Overall prevalence of *Bothriocephalus acheilognathi* in various host species

Host	Dal Lake							River Jhelum						
	No. Examined	No. infected	Prevalence (%)	No. of parasites	Mean Intensity	Abundance	P-value	No. Examined	No. infected	Prevalence (%)	No. of parasites	Mean Intensity	Abundance	P-value
<i>S. niger</i>	76	22	28.94	153	6.9	2.01	< 0.01	76	26	34.21	202	7.7	2.65	< 0.01
<i>S. esocinus</i>	77	24	31.16	139	5.7	1.80	< 0.05	73	25	34.24	144	5.7	1.97	< 0.05
<i>S. curvifrons</i>	71	17	23.94	80	4.7	1.12	< 0.01	71	22	30.98	155	7	2.18	< 0.01
Total	224	63	28.12	372	5.9	1.66	< 0.01	220	73	33.18	501	6.8	2.27	< 0.01

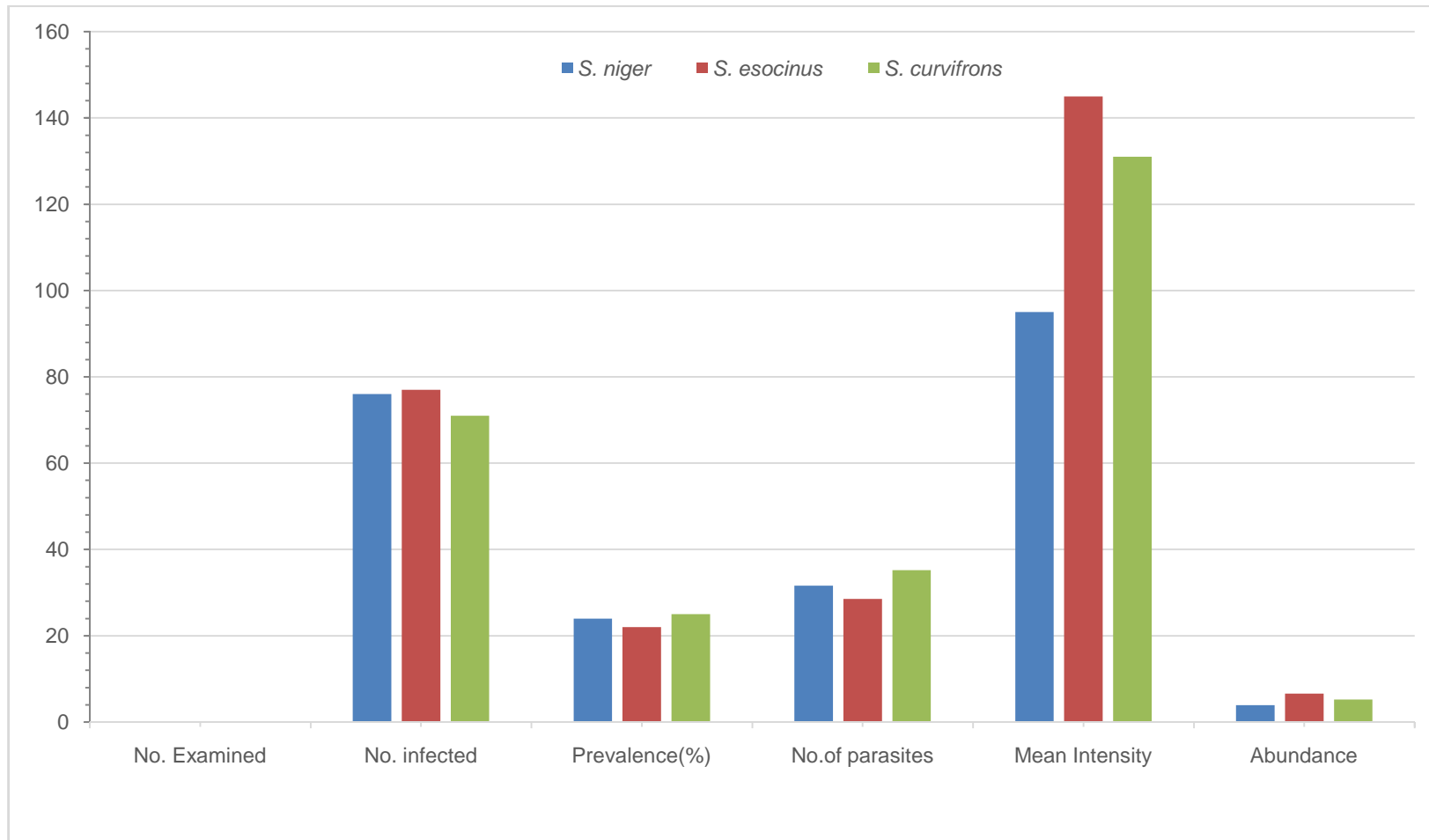


Fig. 19 : Prevalence of *Bothriocephalusacheilognathi* in various host species of Dal Lake

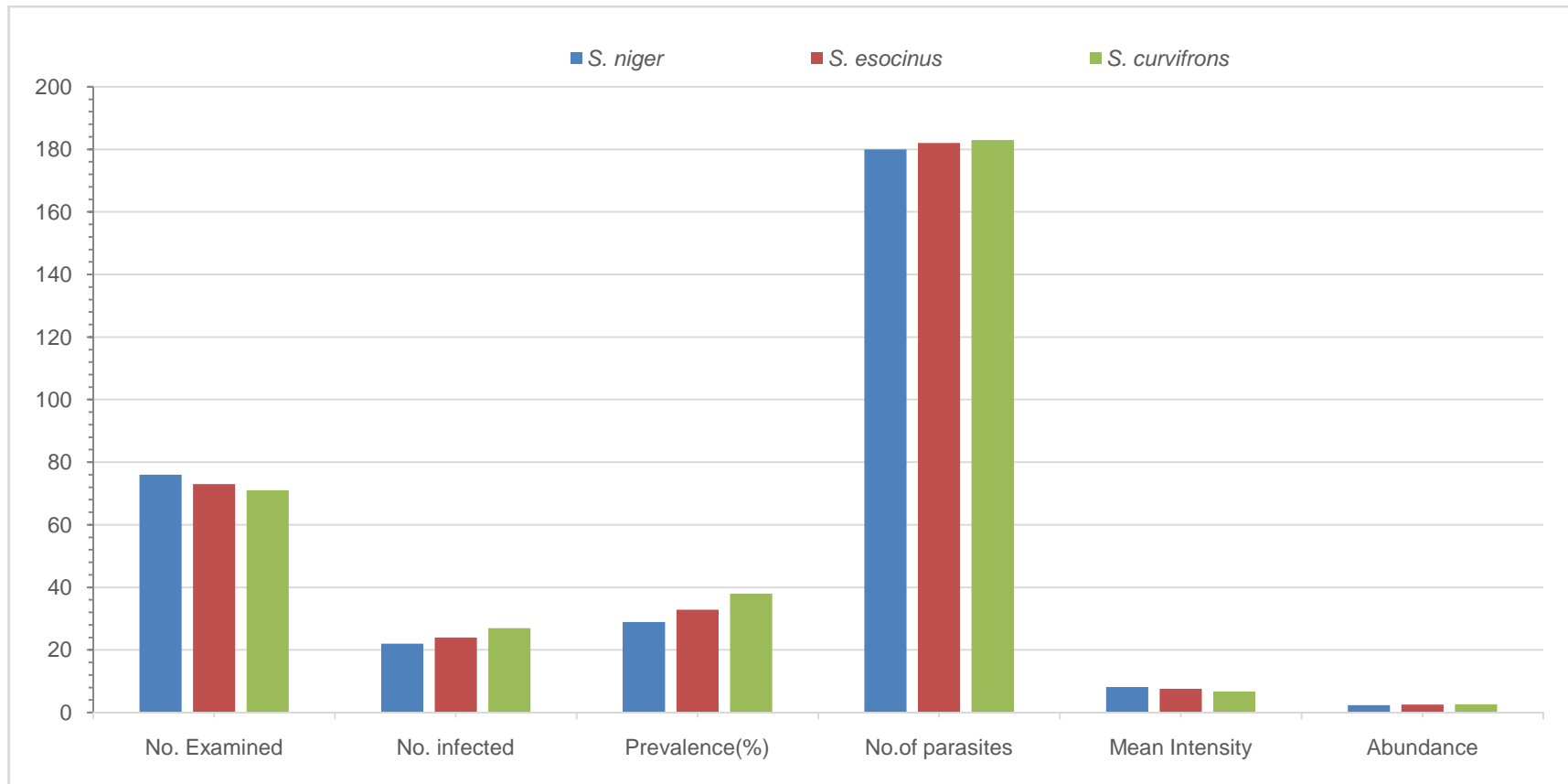


Fig. 20 : Prevalence of *Bothriocephalus acheilognathi* in various host species of River Jehlum

Table 10: Infection dynamics of *Bothriocephalus acheilognathi* recorded of *Schizothorax* spp. from Dal Lake and River Jehlum

Season	Host	Dal Lake							River Jehlum						
		No. Examined	No. infected	Prevalence (%)	No. of parasites	Mean Intensity	Abundance	P-value	No. Examined	No. infected	Prevalence (%)	No. of parasites	Mean Intensity	Abundance	P-value
Spring	<i>S. niger</i>	14	9	64.28	80	8.8	5.71	> 0.05	15	8	53.33	56	7	3.73	> 0.05
	<i>S. esocinus</i>	19	7	36.84	56	8	2.94	> 0.05	20	7	35	48	6.8	2.4	> 0.05
	<i>S. curvifrons</i>	16	7	43.75	42	6	2.62	> 0.05	19	5	26.31	35	7	1.84	> 0.05
Summer	<i>S. niger</i>	18	6	33.33	38	6.3	2.11	> 0.05	16	9	56.25	82	9.1	5.12	> 0.05
	<i>S. esocinus</i>	17	7	41.17	9	1.2	0.52	> 0.05	18	8	44.44	36	4.5	2	> 0.05
	<i>S. curvifrons</i>	19	4	21.05	12	3	0.63	< 0.05	15	6	40	28	4.6	1.86	> 0.05
Autumn	<i>S. niger</i>	25	3	12	6	2	0.24	< 0.01	20	5	25	55	11	2.75	< 0.05
	<i>S. esocinus</i>	19	5	26.31	35	7	1.84	> 0.05	19	7	36.84	48	6.8	2.52	> 0.05
	<i>S. curvifrons</i>	18	4	22.22	17	4.2	0.94	< 0.05	22	8	36.36	74	9.2	3.36	> 0.05
Winter	<i>S. niger</i>	19	4	21.05	29	7.2	1.52	< 0.05	25	4	16	9	2.2	0.36	< 0.01
	<i>S. esocinus</i>	22	5	22.72	39	7.8	1.77	< 0.05	16	3	18.75	12	4	0.75	< 0.05
	<i>S. curvifrons</i>	18	2	11.11	9	4.5	0.5	< 0.05	15	3	20	18	6	1.2	< 0.05
	Total	224	63	28.12	372	5.9	1.66	< 0.01	220	73	33.18	501	6.8	2.27	< 0.01

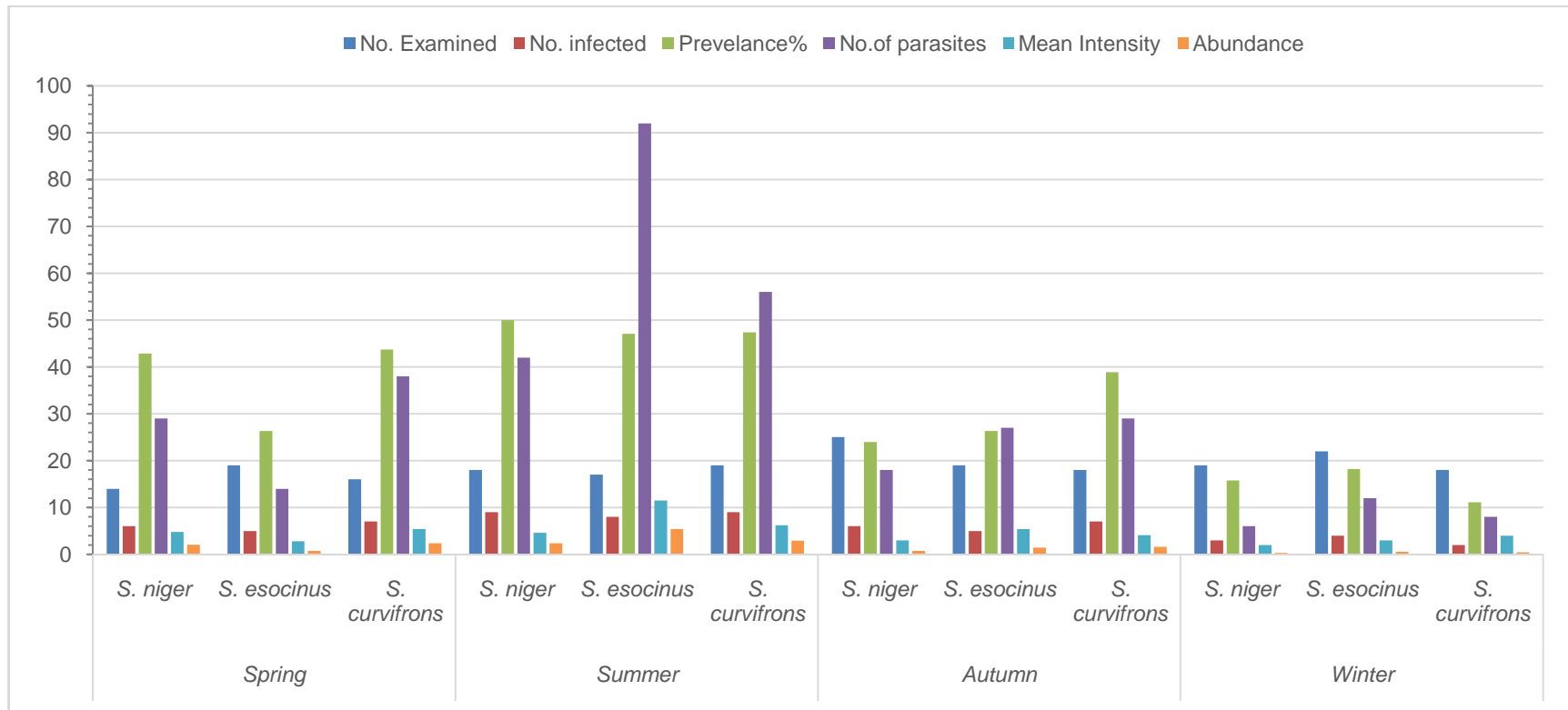


Fig. 21 : Seasonal prevalence of *Bothriocephalusacheilognathi* of *Schizothorax* spp.from Dal Lake

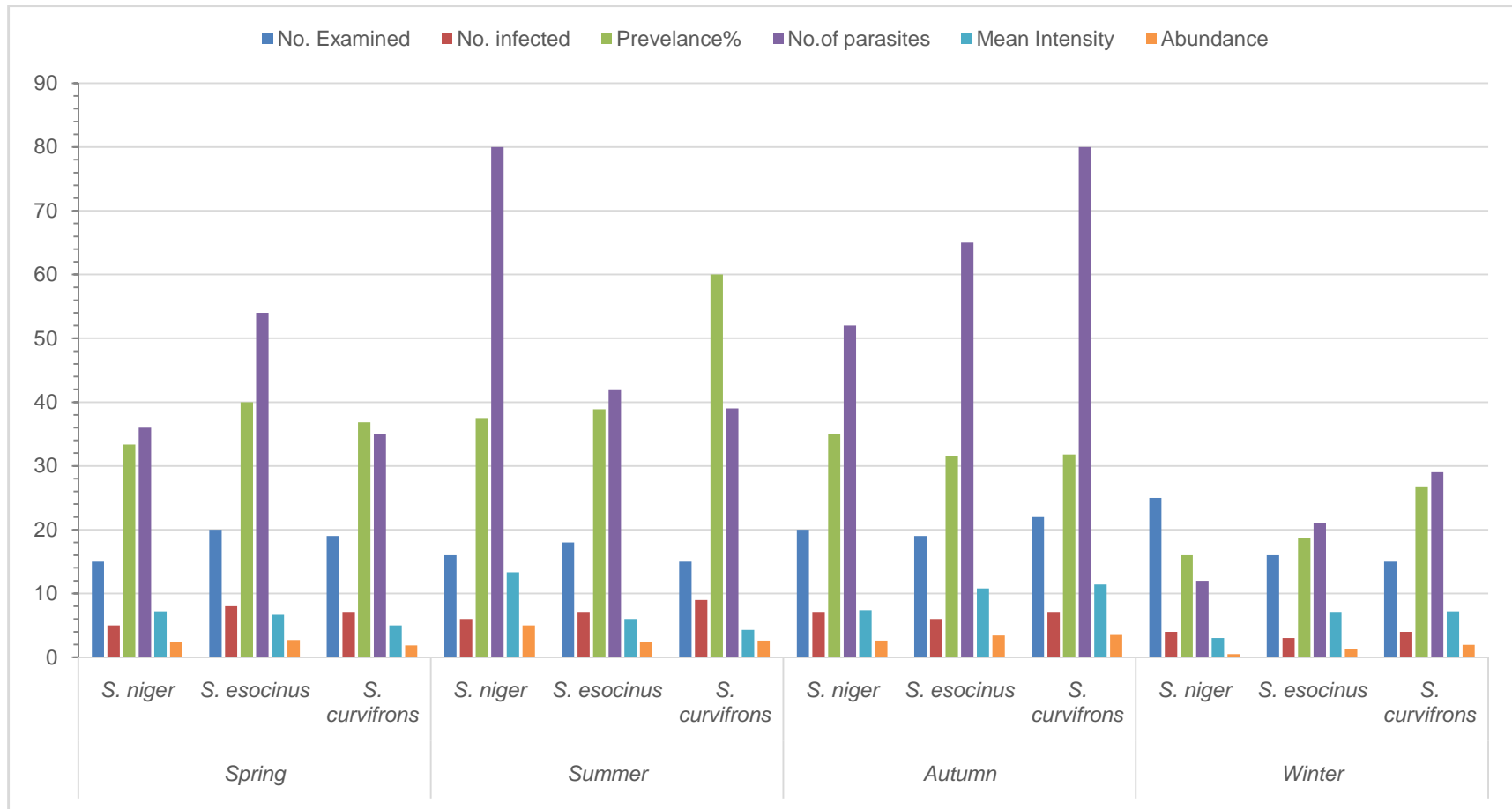


Fig. 22: Seasonal prevalence of *Bothriocephalusacheilognathi* of *Schizothorax* spp. from River Jehlum

Table 11: Gender-wise infection dynamics of *Bothriocephalus acheilognathi* recorded of *Schizothorax* spp. from Dal Lake and River Jehlum

Season	Host	Dal Lake							River Jehlum						
		No. Examined	No. infected	Prevalence (%)	No. of parasites	Mean Intensity	Abundance	P-value	No. Examined	No. infected	Prevalence (%)	No. of parasites	Mean Intensity	Abundance	P-value
<i>S. niger</i>	Male	43	13	30.23	87	6.6	2.3	> 0.05	33	17	51.51	123	7.2	3.72	< 0.01
	Female	33	9	27.27	66	7.3	1.9		43	9	20.93	79	8.7	1.83	
<i>S. esocinus</i>	Male	39	16	41.02	97	6	2.1	> 0.05	47	19	40.42	92	4.8	1.95	> 0.05
	Female	38	8	21.05	42	5.2	1.9		26	6	23.07	52	8.6	2	
<i>S. curvifrons</i>	Male	52	13	25	67	5.1	2	> 0.05	37	12	32.43	108	9	2.91	> 0.05
	Female	19	4	21.05	13	3.2	0.4		34	10	29.41	47	4.7	1.38	

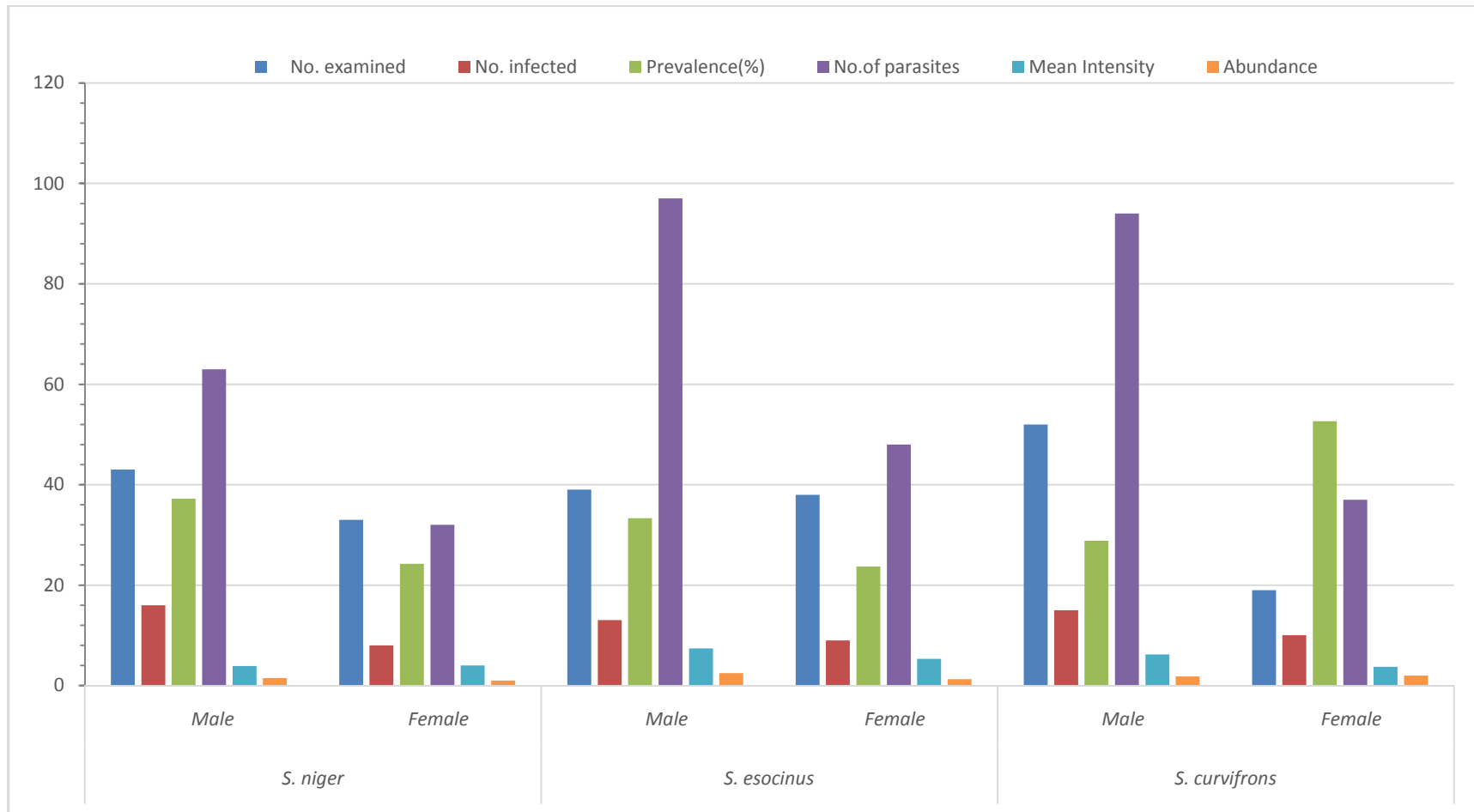


Fig. 23: Genderwise prevalence of *Bothriocephalus acheilognathi* of *Schizothorax* spp. from Dal Lake

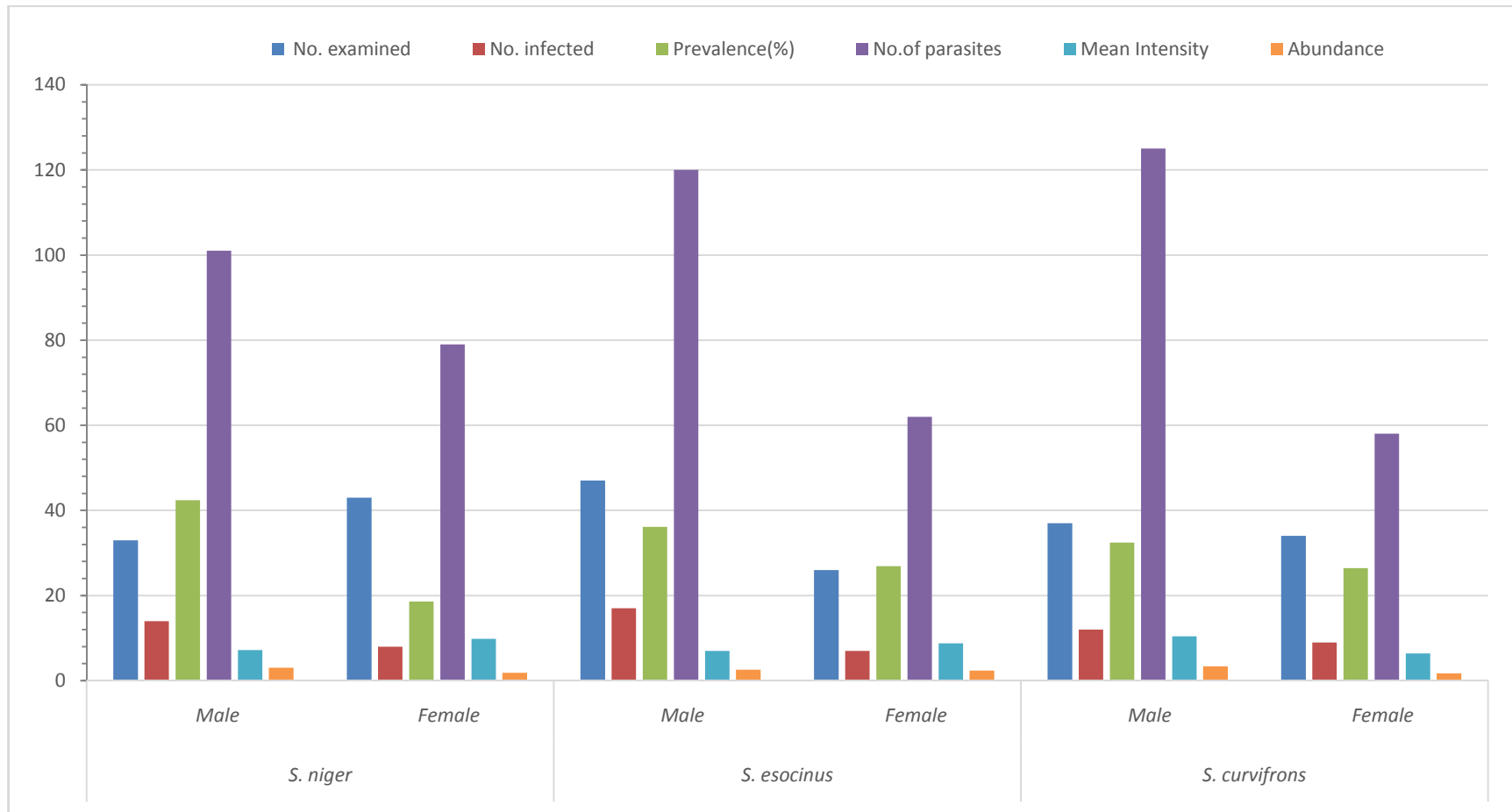


Fig. 24 : Genderwise prevalence of *Bothriocephalusacheilognathi* of *Schizothorax* spp. from River Jehlum

4.2.4 Prevalence of *Adenoscolex oreini* in Dal Lake and River Jhelum

4.2.4.1 Host-wise prevalence (Table 12 and Fig. 25&26)

Examination of 444 fish specimens revealed 144 (32.43%) were found to be infected with the *Adenoscolex oreini*. Out of 224 specimens examined from the Dal Lake 71 (31.69%) specimens were found infected with *Adenoscolex Oreini*. Host-wise distribution of the parasite was significantly varied ($p < 0.01$) which showed *S. niger* (31.57%), *S. esocinus* (28.57%) and *S. curvifrons* (35.21%). Out of 220 *Schizothorax* spp. examined from River Jhelum 73 (33.18%) were infected with *Adenoscolex oreini* which included *S. niger* (28.94%), *S. esocinus* (32.87%) and *S. curvifrons* (38.02%).

The mean intensity of *Adenoscolex oreini* in *S. niger*, *S. esocinus* and *S. curvifrons* in Dal Lake were 3.9, 6.5 and 5.2 having an abundance of 1.25, 1.88 and 1.84, respectively. Where as it was 8.18, 7.58 and 6.77 in the respective fishes of River Jhelum which had an abundance of 2.36, 2.49 and 2.57 respectively. The overall mean intensities of *Adenoscolex oreini* in *Schizothorax* spp. of Dal Lake and River Jhelum were 5.22 and 7.46 having an abundance of 1.65 and 2.4 respectively.

4.2.4.2 Seasonal prevalence (Table 13 and Fig. 27&28)

Seasonal estimation of *Adenoscolex oreini* infection revealed definite variation in the prevalence of infection in *Schizothorax* spp. with highest rate of infection in summer and lowest in winter. There was a gradual increase in the prevalence rate from spring to summer and decreased with the onset of autumn and least observed prevalence was observed during winter season. In Dal Lake during summer the overall prevalence of *Adenoscolex oreini* infection was highest (48.14%) and the species-wise distribution were *S. niger* (50.0%), *S. esocinus* (47.05%) and *S. curvifrons* (47.3%) while as during winter it was least (15.25%) with species-wise distribution as *S. niger* (15.78%), *S. esocinus* (18.18%) and *S. curvifrons* (11.11%). Contrary to this in River Jhelum during summer the

prevalence was 44.89% with species-wise distribution as *S. niger* (37.5%), *S. esocinus* (38.8%) and *S. curvifrons* (60.0%) and the least prevalence (19.6%) was found during the winter season with as *S. niger*(16.0%), *S. esocinus*(18.7%) and *S. curvifrons*(26.6%).

The mean intensities of the parasite during summer were 4.6, 11.5 and 6.2 in *S. niger*, *S. esocinus* and *S. curvifrons*, respectively with the abundance of the parasites as 2.3, 5.4 and 2.9 for the respective fishes in Dal Lake. Whereas during winter the mean intensities were 2, 3 and 4 with abundance of 0.3, 0.5 and 0.4 in the corresponding fishes. In river Jhelum the mean intensities of the parasite during summer were 13.3, 6.0 and 4.3 in *S. niger*, *S. esocinus* and *S. curvifrons* respectively with the abundance of the parasites as 5, 2.3 and 2.6, respectively. Whereas during winter the mean intensities were 3, 7 and 7.2 with abundance of 0.4, 1.3 and 1.9 in the corresponding fishes.

4.2.4.3 Gender-wise prevalence of *Adenoscolex oreini*(Table14 and Fig. 29&30)

Gender-wise distribution of *Adenoscolex oreini* in *Schizothorax* spp. of Dal Lake revealed that the overall prevalence of the parasite in males was 32.83% (*S. niger* 37.2%, *S. esocinus* 33.33% and *S. curvifrons* 28.84%) whereas it was 30% in females (*S. niger* 24.24%, *S. esocinus* 23.68% and *S. curvifrons* 52.63%). In River Jhelum the overall prevalence in males was 36.75% (*S. niger* 42.42%, *S. esocinus* 36.17% and *S. curvifrons* 32.4%) while as in females it was 23.3% (*S. niger* 18.6%, *S. esocinus* 26.92% and *S. curvifrons* 26.4%).

The overall mean intensities of the parasite in males and females of *Schizothorax* spp. were 5.7 and 4.3, with an abundance of 1.8 and 1.3 respectively in Dal Lake. In river Jhelum the overall mean intensities of the parasite in males and females of *Schizothorax* spp. were 8.0 and 8.2 with an abundance of 2.9 and 1.5, respectively.

Table 12: Overall prevalence of *Adenoscolex oreini* in various host species

Host	Dal Lake							River Jhelum						
	No. Examined	No. infected	Prevalence (%)	No. of parasites	Mean Intensity	Abundance	P-value	No. Examined	No. infected	Prevalence (%)	No. of parasites	Mean Intensity	Abundance	P-value
<i>S. niger</i>	76	24	31.57	95	3.95	1.25	< 0.01	76	22	28.94	180	8.18	2.36	< 0.01
<i>S. esocinus</i>	77	22	28.57	145	6.59	1.88	< 0.01	73	24	32.87	182	7.58	2.49	< 0.01
<i>S. curvifrons</i>	71	25	35.21	131	5.24	1.84	< 0.05	71	27	38.02	183	6.77	2.57	> 0.05
Total	224	71	31.69	371	5.22	1.65	< 0.01	220	73	33.18	545	7.46	2.47	< 0.01

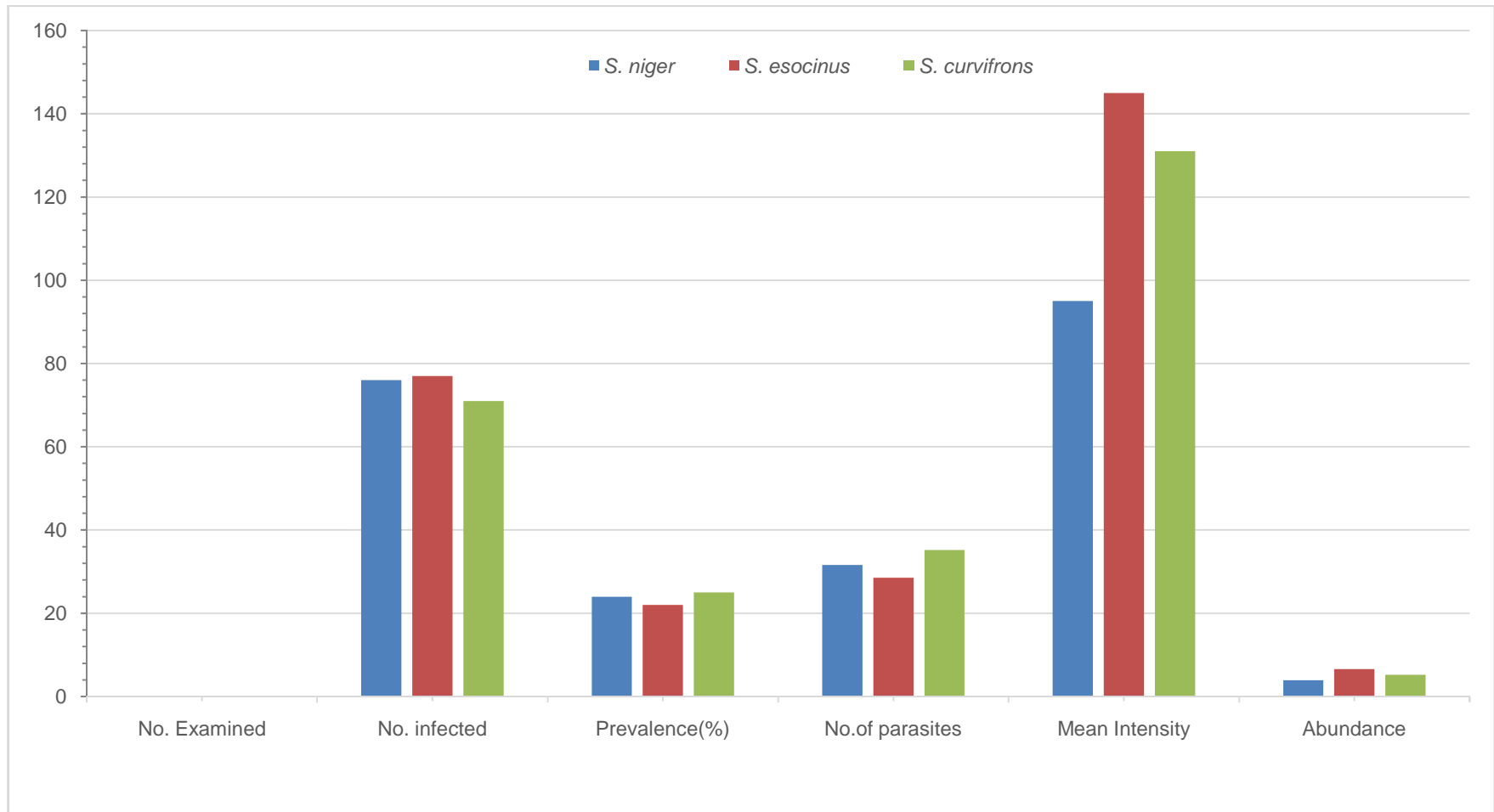


Fig. 25 : Prevalance of *Adenoscolex oreini* in various host species of Dal Lake

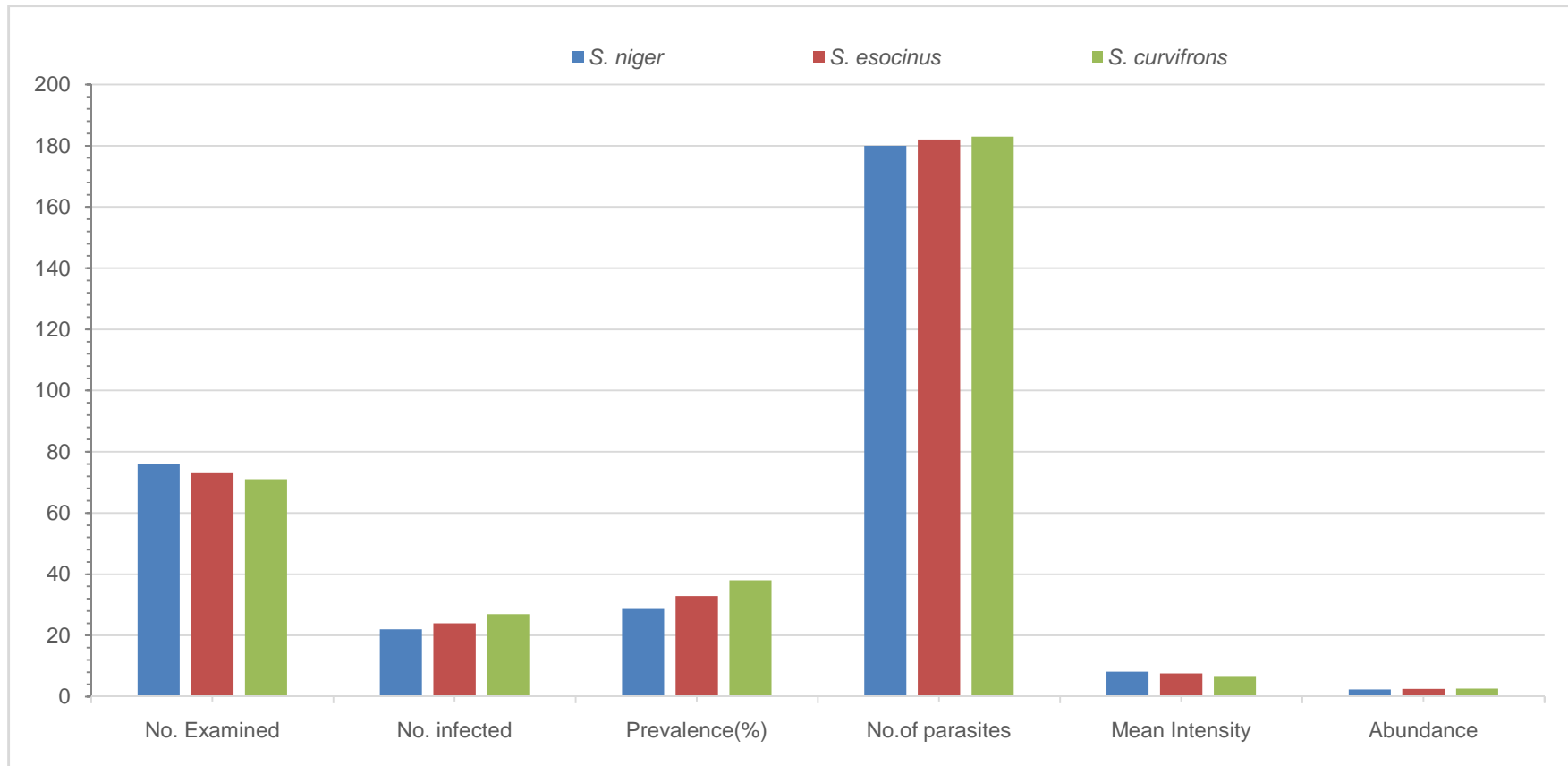


Fig. 26 : Prevalence of *Adenoscolex oreini* in various host species of River Jehlum

Table 13: Infection dynamics of *Adenoscolex oreini* recorded of *Schizothorax* spp. from Dal Lake and River Jehlum

Season	Host	Dal Lake							River Jehlum						
		No. Examined	No. infected	Prevalence (%)	No. of parasites	Mean Intensity	Abundance	P-value	No. Examined	No. infected	Prevalence (%)	No. of parasites	Mean Intensity	Abundance	P-value
Spring	<i>S. niger</i>	14	6	42.85	29	4.8	2.07	> 0.05	15	5	33.33	36	7.2	2.4	> 0.05
	<i>S. esocinus</i>	19	5	26.31	14	2.8	0.73	> 0.05	20	8	40	54	6.7	2.7	> 0.05
	<i>S. curvifrons</i>	16	7	43.75	38	5.4	2.37	> 0.05	19	7	36.84	35	5	1.84	> 0.05
Summer	<i>S. niger</i>	18	9	50	42	4.6	2.33	> 0.05	16	6	37.5	80	13.3	5	> 0.05
	<i>S. esocinus</i>	17	8	47.05	92	11.5	5.41	> 0.05	18	7	38.88	42	6	2.33	> 0.05
	<i>S. curvifrons</i>	19	9	47.36	56	6.2	2.94	> 0.05	15	9	60	39	4.3	2.6	> 0.05
Autumn	<i>S. niger</i>	25	6	24	18	3	0.72	<0.05	20	7	35	52	7.4	2.6	> 0.05
	<i>S. esocinus</i>	19	5	26.31	27	5.4	1.42	> 0.05	19	6	31.57	65	10.8	3.42	> 0.05
	<i>S. curvifrons</i>	18	7	38.88	29	4.1	1.61	> 0.05	22	7	31.81	80	11.4	3.63	> 0.05
Winter	<i>S. niger</i>	19	3	15.78	6	2	0.31	< 0.01	25	4	16	12	3	0.48	< 0.01
	<i>S. esocinus</i>	22	4	18.18	12	3	0.54	< 0.01	16	3	18.75	21	7	1.31	< 0.05
	<i>S. curvifrons</i>	18	2	11.11	8	4	0.44	< 0.01	15	4	26.66	29	7.2	1.93	> 0.05
	Total	224	71	31.69	371	5.2	1.65	< 0.01	220	73	33.18	545	7.4	2.47	< 0.01

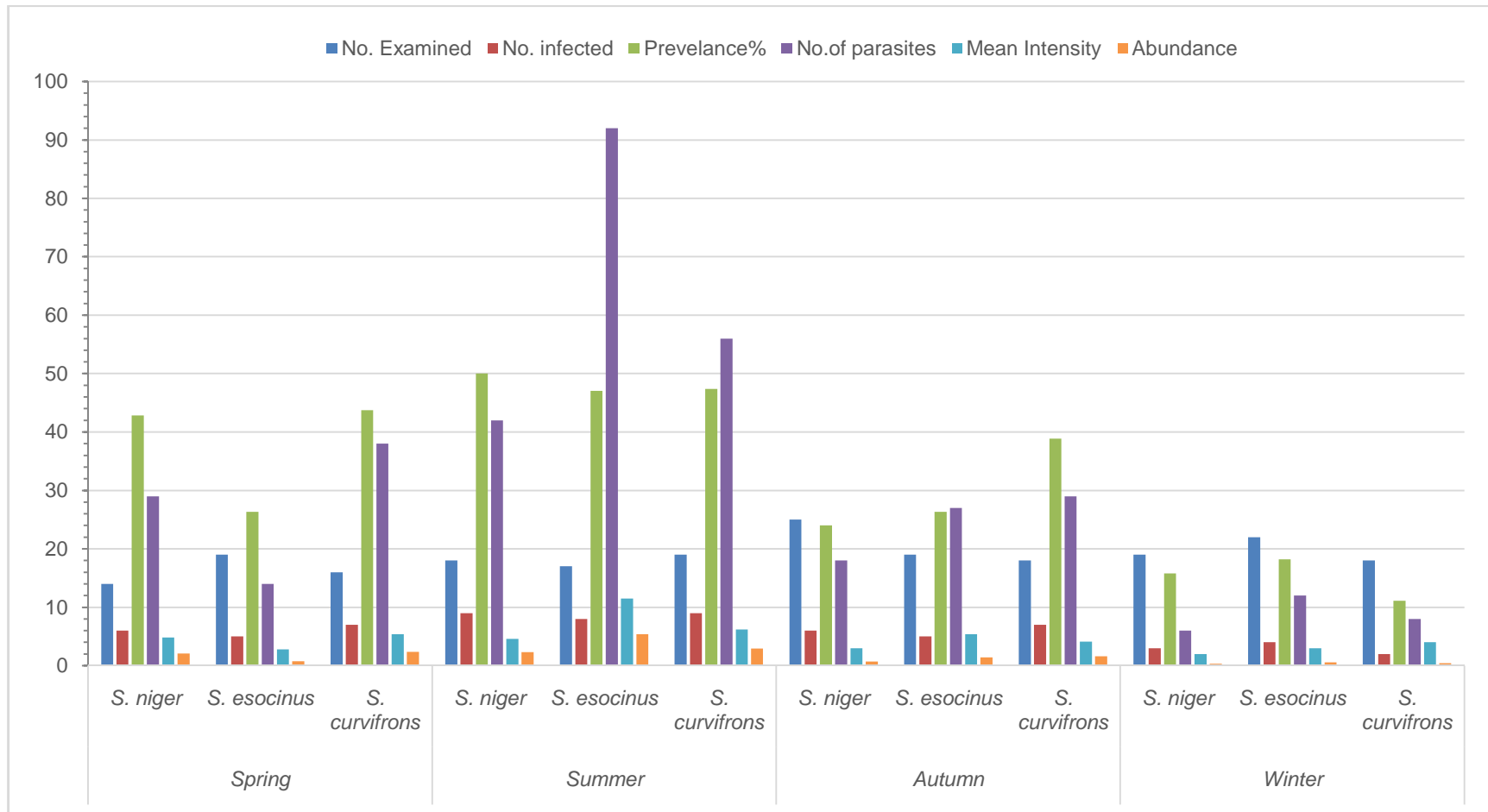


Fig. 27 : Seasonal prevalence of *Adenoscolex oreini* of *Schizothorax* spp. from Dal Lake

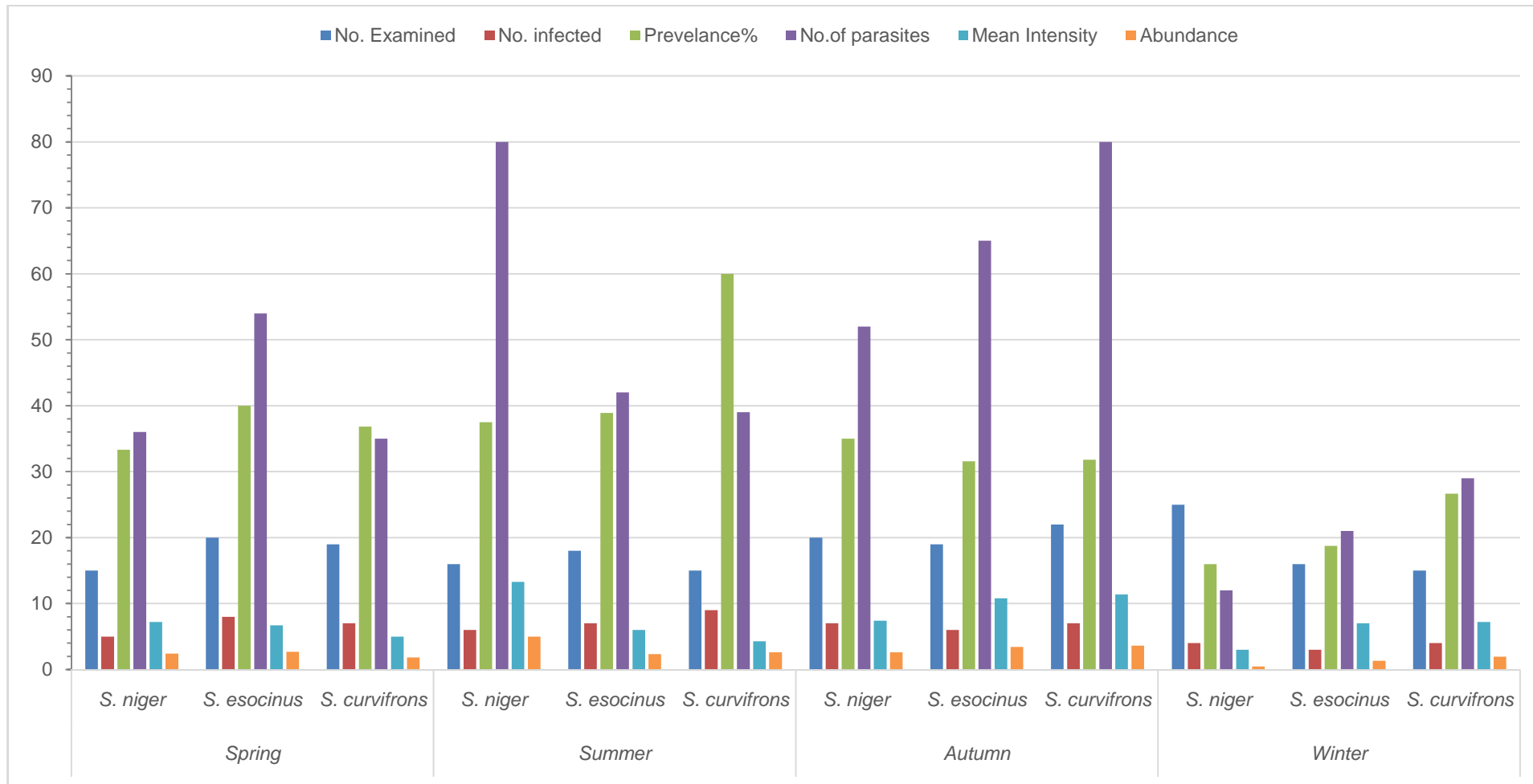


Fig. 28 : Seasonal prevalence of *Adenoscolex oreini* of *Schizothorax* spp. from River Jhelum

Table 14: Gender-wise infection dynamics of *Adenoscolex oreini* recorded of *Schizothorax* spp. from Dal Lake and River Jehlum

Season	Host	Dal Lake							River Jehlum						
		No. Examined	No. infected	Prevalence (%)	No. of parasites	Mean Intensity	Abundance	P-value	No. Examined	No. infected	Prevalence (%)	No. of parasites	Mean Intensity	Abundance	P-value
<i>S. niger</i>	Male	43	16	37.20	63	3.9	1.46		33	14	42.42	101	7.2	3.06	< 0.05
	Female	33	8	24.24	32	4	0.96	> 0.05	43	8	18.60	79	9.8	1.83	
<i>S. esocinus</i>	Male	39	13	33.33	97	7.4	2.48		47	17	36.17	120	7	2.55	> 0.05
	Female	38	9	23.68	48	5.3	1.26	> 0.05	26	7	26.92	62	8.8	2.38	
<i>S. curvifrons</i>	Male	52	15	28.84	94	6.2	1.80		37	12	32.43	125	10.4	3.37	> 0.05
	Female	19	10	52.63	37	3.7	1.94	> 0.05	34	9	26.47	58	6.4	1.70	

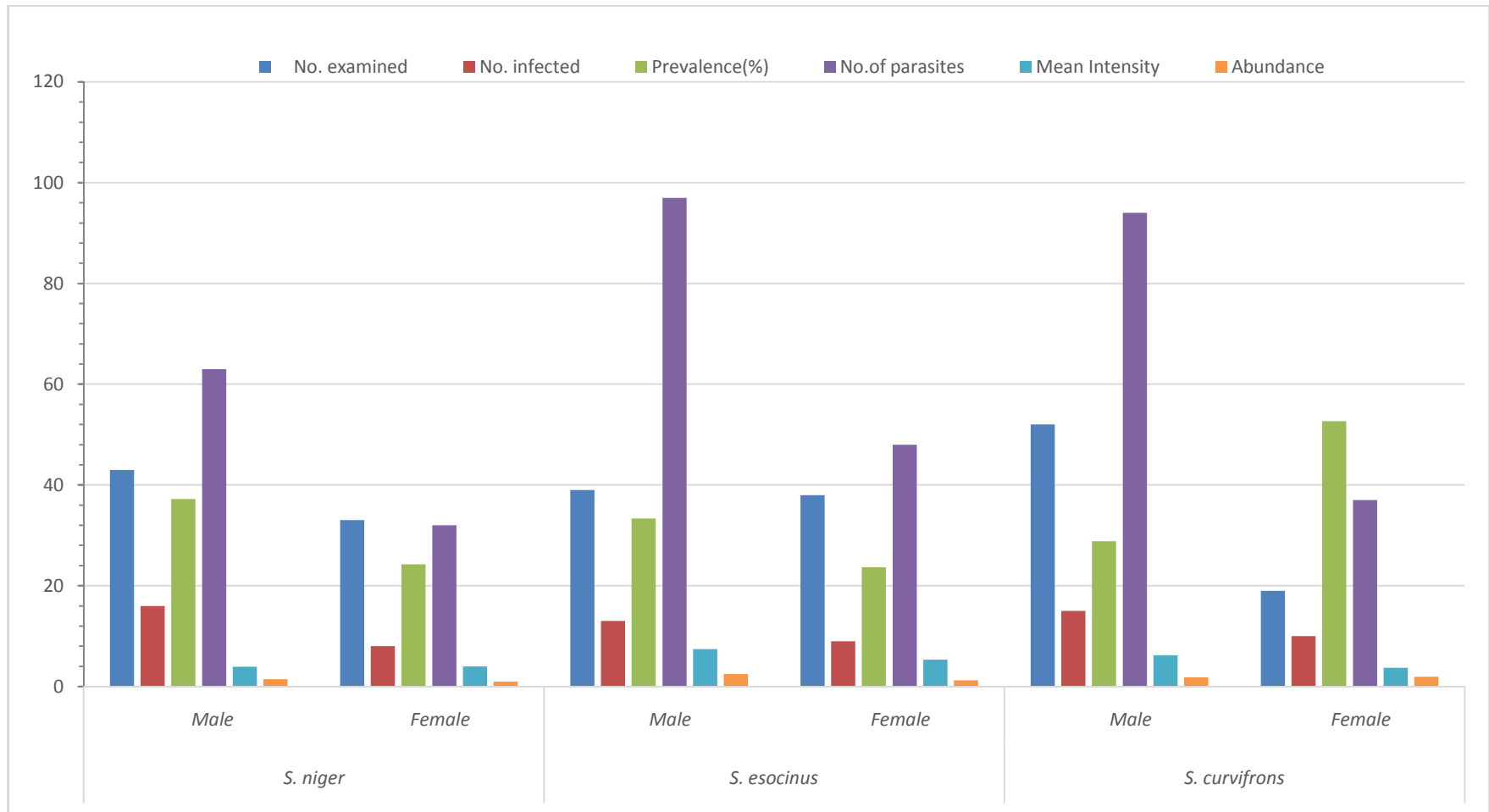


Fig. 29 : Genderwise prevalence of *Adenoscolex oreini* recorded of *Schizothorax* spp. from Dal Lake

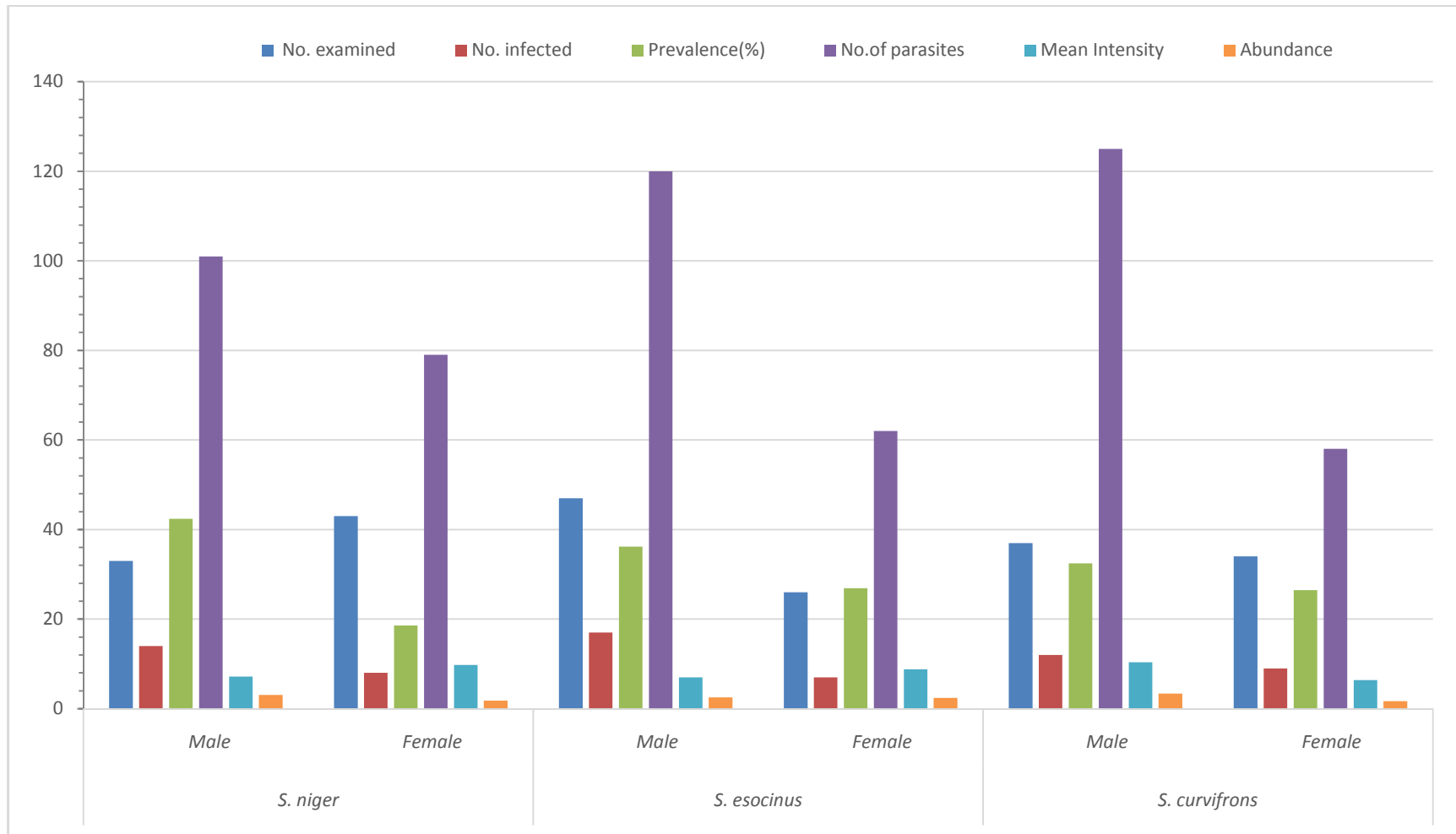


Fig. 30 : Genderwise prevalence of *Adenoscolex oreini* recorded of *Schizothorax* spp. from River Jehlum

4.2.5 Influence of sex and condition factor on the level of infection (Fig. 31 to 37)

Insignificant relationship existed between gender and helminth infection. Condition factors was found to be lower in infected fish than in uninfected fish in both water bodies.

Analysis of the condition factor of uninfected and infected *Schizothorax* spp. of Dal Lake and River Jhelum revealed significant differences ($p < 0.05$) with higher in River Jhelum. The condition factors for various *Schizothorax* spp. in the two water bodies by Mann-Whitney test revealed *S. niger* ($U=13$, $p < 0.01$), *S. esocinus* ($U=45$, $p > 0.05$) and for *S. curvifrons* ($U=34$, $p > 0.05$) of Dal lake While as in River Jhelum it was *S. niger* ($U=3$, $p < 0.01$), *S. esocinus* ($U=3$, $p < 0.01$) and for *S. curvifrons* ($U=16$, $p < 0.05$).

4.2.6 Correlation between prevalence and water quality in two water bodies (Table 15)

Temperature was the most important abiotic factor that affected the parasites at all life cycle stages. A positive correlation ($P < 0.01$) existed between water temperature and parasitic prevalence in Dal Lake and River Jhelum.

Prevalence of *P. kashmirensis* and *A. oreini* in the fishes of Dal Lake presented a significant negative correlation ($P < 0.01$) with dissolved oxygen whereas it showed insignificant negative correlation ($P > 0.05$) in all other cases of patterns of infection under various locations.

pH showed insignificant positive correlation ($P > 0.05$) with all parasitic infections. Prevalence of infections showed insignificant positive correlation ($P > 0.05$) with Carbon dioxide except for *P. kashmirensis* of River Jhelum and *B. acheilognathi* of Dal Lake and River Jhelum which showed significant positive correlation ($P < 0.05$).

Table-15: Correlation between environmental variables and prevalence of *Pomphorhynchus Kashmirensis*, *Adenoscolex oreini* and *Bothriocephalus acheilognathi*

Environmental variables	Prevalence of <i>Pomphorynchus kashmirensis</i>				Prevalence of <i>Adenoscolex oreini</i>				Prevalence of <i>Bothriocephalus acheilognathi</i>			
	Dal Lake		River Jhelum		Dal Lake		River Jhelum		Dal Lake		River Jhelum	
	Pearson's coef.	Spearman's rho	Pearson's coef.	Spearman's rho	Pearson's coef.	Spearman's rho	Pearson's coef.	Spearman's rho	Pearson's coef.	Spearman's rho	Pearson's coef.	Spearman's rho
Temperature	.882**	.853**	.907**	.907**	.802**	.699*	.810**	.592*	.722**	.664*	.922**	.729**
Oxygen	-.842**	-.825**	-.451	-.350	-.755**	-.643*	-.105	-.084	-.297	-.280	.017	.056
pH	.420	.587*	-.227	-.266	.487	.601*	-.111	-.182	.231	.231	.182	.364
Carbon dioxide	.682	.021	.628*	.522	-.007	-.035	.529	.322	.598*	.629*	.579*	.581*

*Correlation is significant at the 0.05 level (2-tailed)

**Correlation is significant at the 0.01 level (2-tailed)

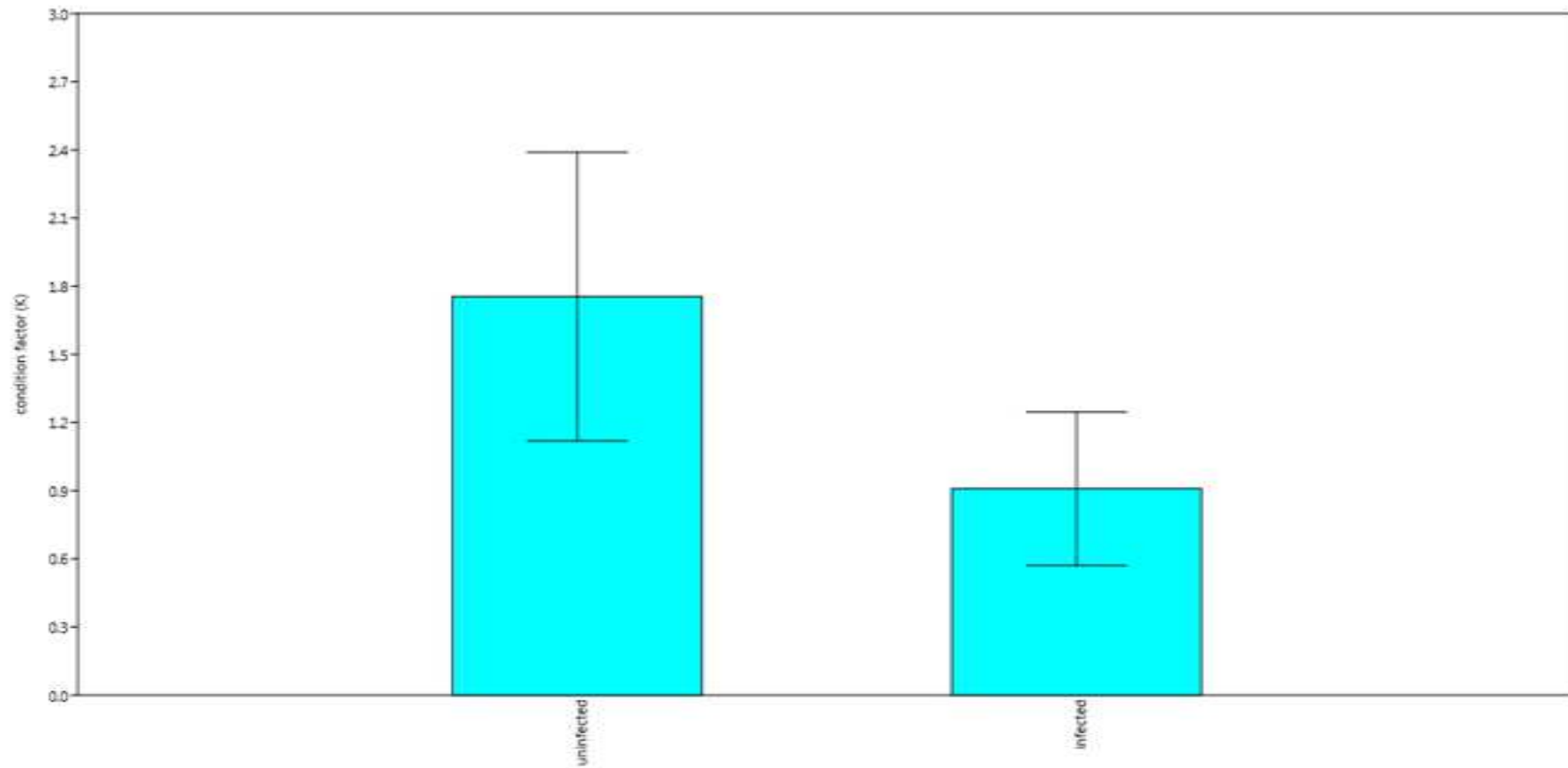


Fig 31. Condition factor of infected and uninfected *Schizothoraxniger* of Dal Lake

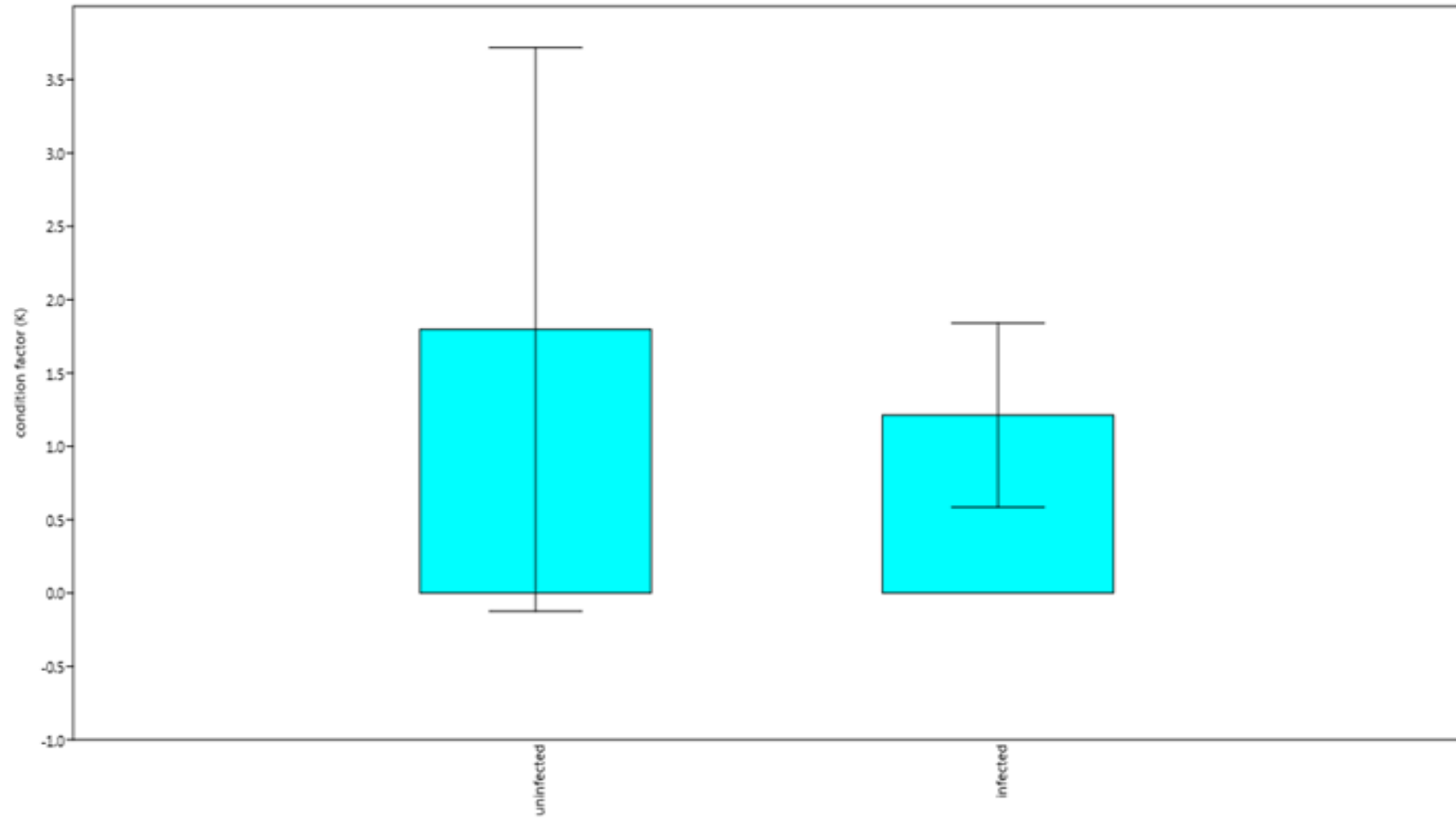


Fig 32. Condition factor of infected and uninfected *Schizothorax esocinus* of Dal Lake

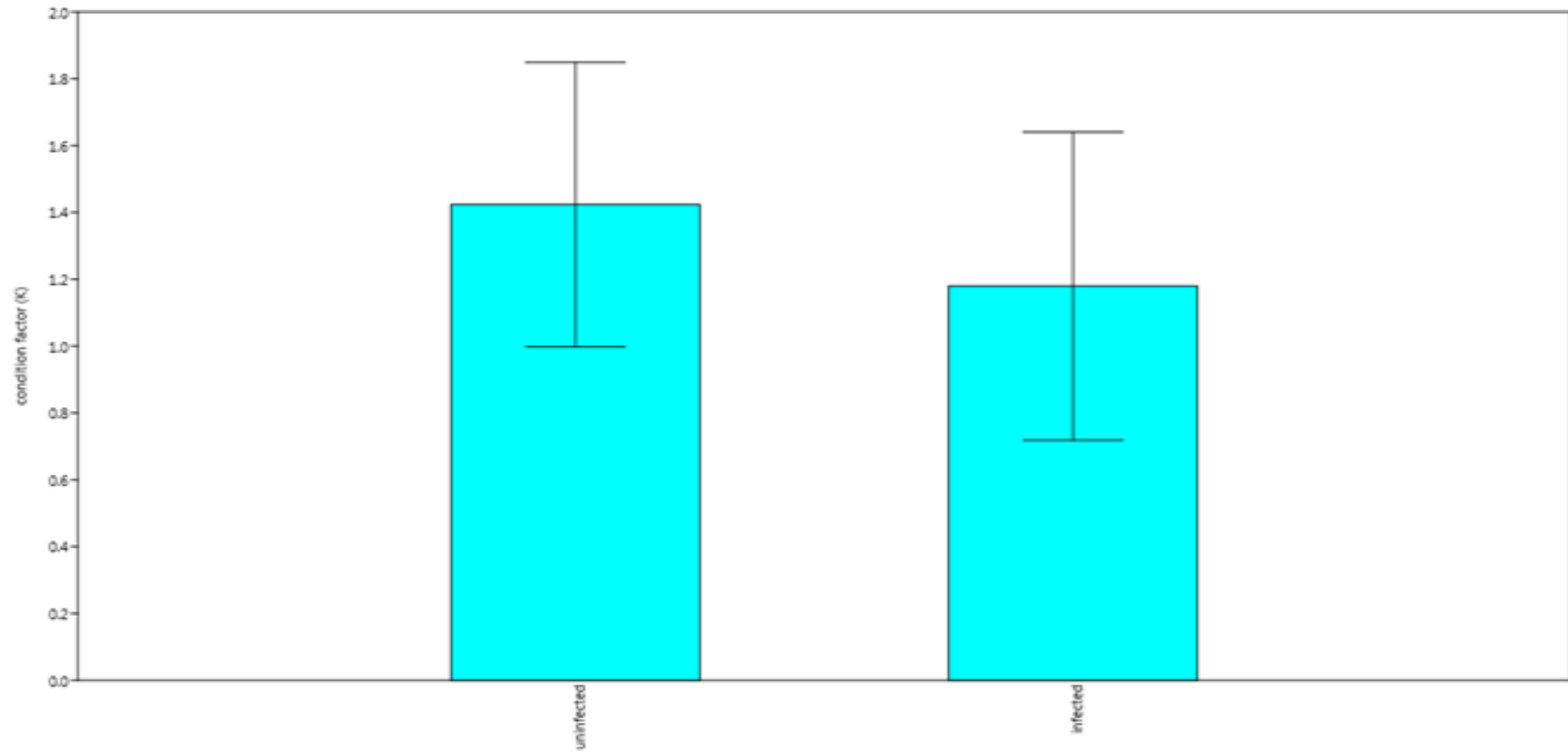


Fig 33. Condition factor of infected and uninfected *Schizothorax curvifrons* of Dal Lake

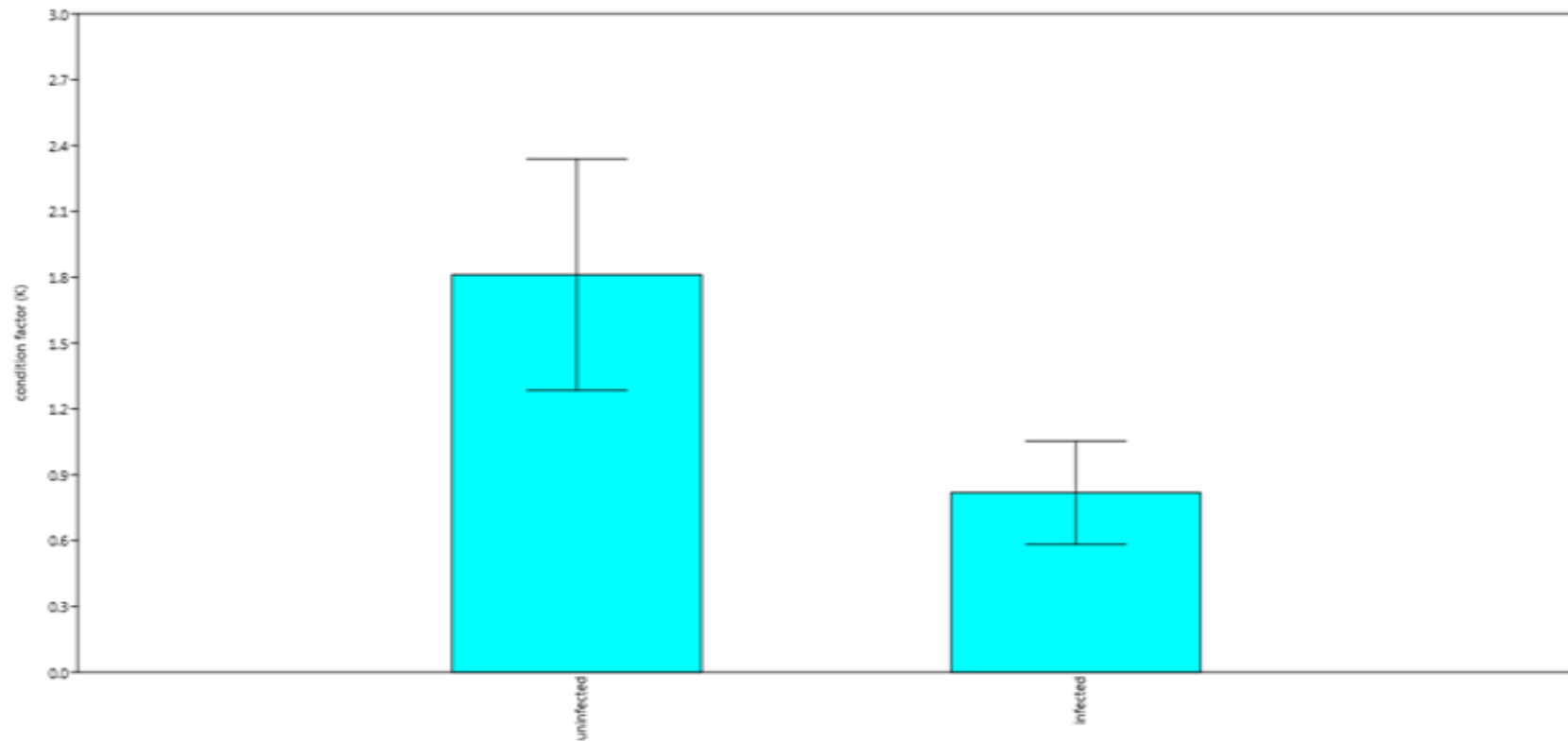


Fig 34. Condition factor of infected and uninfected *Schizothoraxniger* of River Jhelum

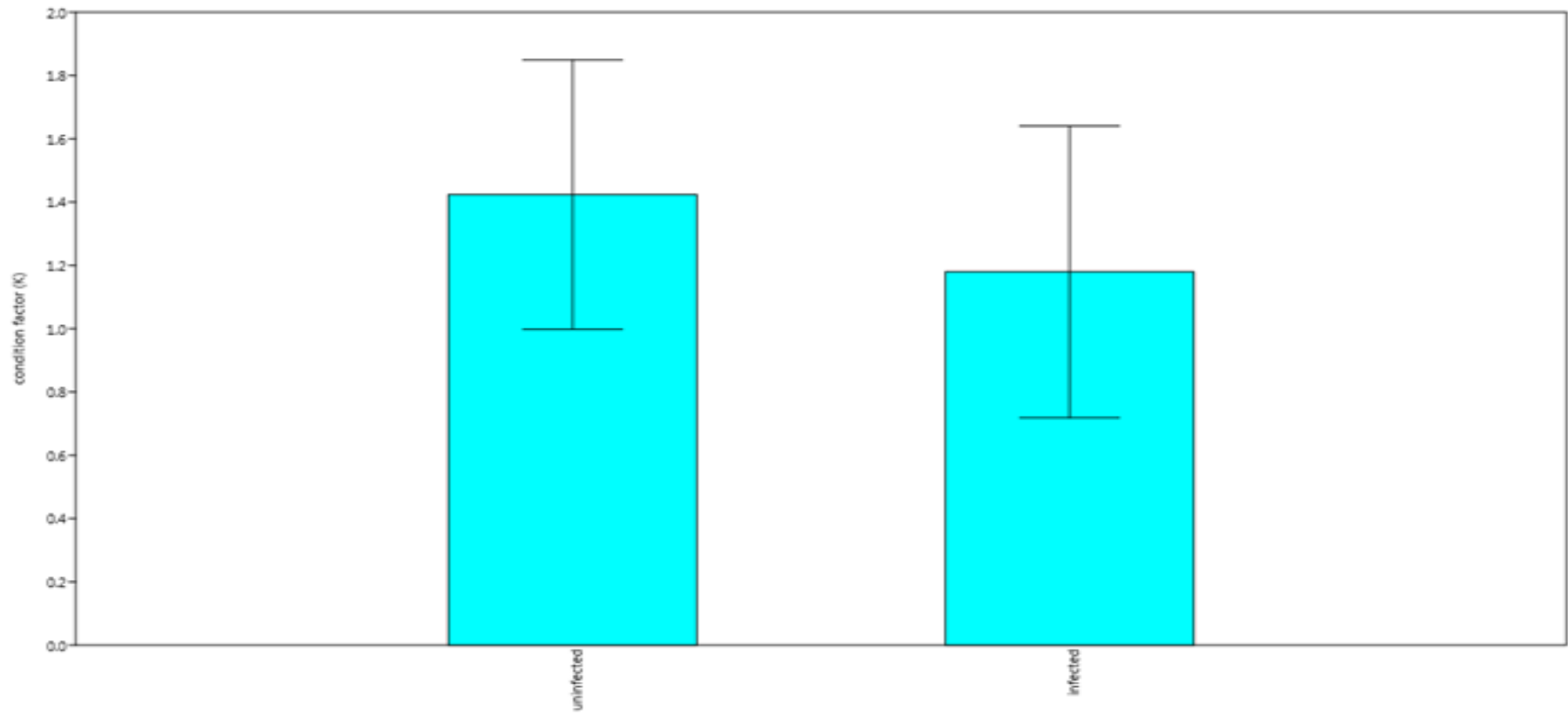


Fig. 35. Condition factor of infected and uninfected *Schizothorax esocinus* of River Jhelum

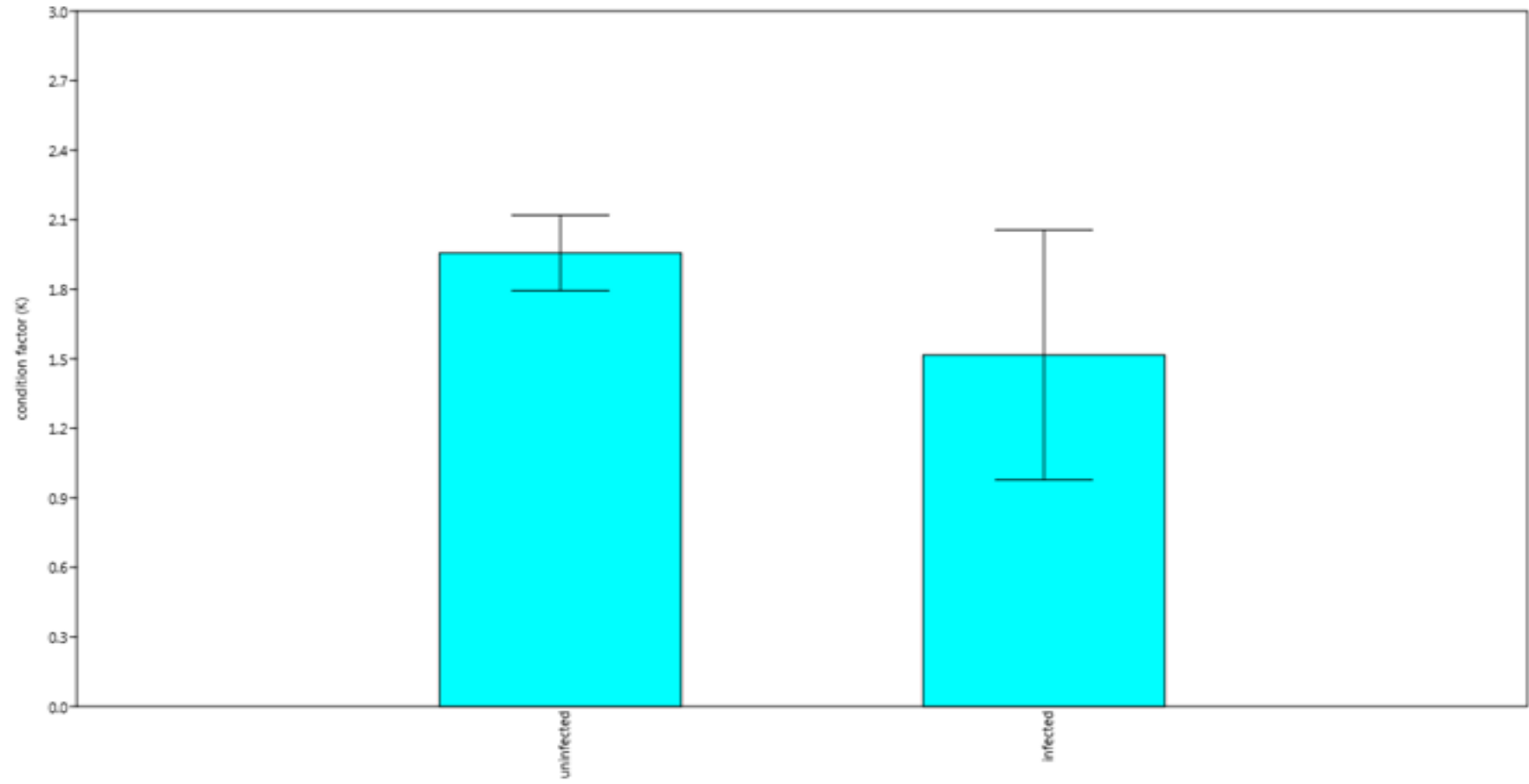


Fig. 36. Condition factor of infected and uninfected *Schizothorax curvifrons* of River Jhelum

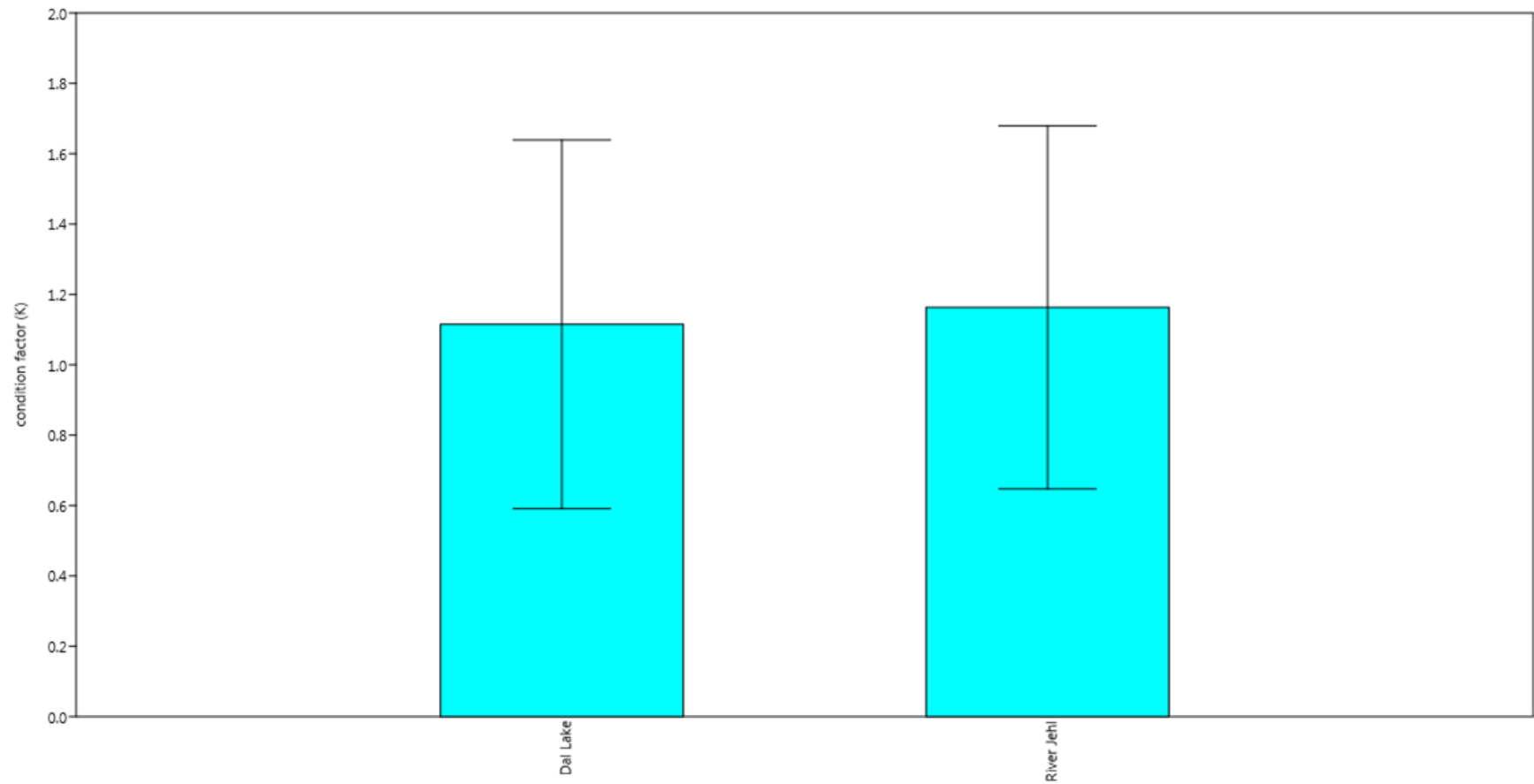


Fig. 37:Overall comparative analysis of condition factor of fish in two water bodies with different water quality

4.3 Metal concentration in water (Tables 16 to 22)

Results of the analysis of the heavy metals (Cu, Zn, Co, Ni, Mn, Cr, Al, Fe, Ca, Cd, Pb and Hg) in water samples, parasites and tissues of *Schizothorax niger* and their correlations are presented in the Tables. The water samples analyzed, during different seasons, were from seven different locations of the two water bodies, viz, Dalgate, Saidakadal, Hazratbal and Telbal of Dal Lake, and Zerobridge, Chattabal weir and Khannabal of River Jhelum.

We performed repeated measure test, ANOVA technique and post hoc test for seasonal comparison. Mean \pm SD of heavy metal concentrations in waters at the seven sites were tabulated in Table 2 to 7. The ranking of mean concentrations of the twelve metals at different sites of Dal Lake was as Dalgate site Fe > Ca > Mn > Al > Zn > Cd > Cu > Ni > Co > Cr; Saidakadal site as Fe > Mn > Ca > Al > Zn > Cd > Ni > Co > Cr > Cu; Hazratbal site as Al > Fe > Ca > Mn > Zn > Cd > Ni > Co > Cr > Cu; Telbal site as Fe > Al > Ca > Mn > Cu > Ni > Co > Zn > Cd > Cr. In River Jhelum the ranking of metals at different sites was Zerobridge, site as Ca > Fe > Al > Mn > Zn > Ni > Cu > Co > Cd > Cr; Chattabal weir site as Fe > Mn > Ca > Al > Co > Zn > Cd > Ni > Cu > Cr and Khannabal site as Fe > Ca > Al > Mn > Zn > Co > Cd > Cu > Cr > Ni. Applying one way ANOVA, showed highly significant difference ($p < 0.01$) between the seven investigated sites for all heavy metals concentration.

4.3.1 Copper (Cu)

The concentrations of copper in water samples from different stations varied significantly ($p < 0.05$) seasonally. Overall the average concentrations (Mean \pm SD) at Dalgate, Saidakadal, Hazratbal and Telbal of Dal Lake were 0.20 \pm 0.12 mg/l, 0.21 \pm 0.34 mg/l, 0.04 \pm 0.01 mg/l and 0.65 \pm 0.80 mg/l, respectively. At Zerobridge, Chattabal weir and Khannabal of River Jhelum these were 0.32 \pm 0.38 mg/l, 0.15 \pm 0.21 mg/l and 0.74 \pm 1.14 mg/l, respectively. Significant ($p < 0.05$) variation existed during different seasons at different locations.

Table 16: Descriptive statistics obtained by ANOVA to compare concentration (mg/l) of the selected heavy metals of the water samples collected along Dalgate of Dal Lake seasonally

Location	Seasons				Overall Mean	C.D
	Autumn (Mean \pm SD)	Winter (Mean \pm SD)	Spring (Mean \pm SD)	Summer (Mean \pm SD)		
Copper (Cu)	0.047 \pm 0.015	0.156 \pm 0.022	0.288 \pm 0.049	0.331 \pm 0.036	0.20 \pm 0.12	0.100
Zinc (Zn)	0.768 \pm 0.056	0.039 \pm 0.007	0.325 \pm 0.033	1.437 \pm 0.194	0.64 \pm 0.60	0.309
Cobalt (Co)	0.537 \pm 0.037	0.160 \pm 0.051	0.003 \pm 0.002	0.003 \pm 0.002	0.17 \pm 0.25	0.095
Nickel (Ni)	0.483 \pm 0.056	0.316 \pm 0.016	0.003 \pm 0.002	0.003 \pm 0.002	0.20 \pm 0.23	0.089
Manganese (Mn)	1.455 \pm 0.083	1.581 \pm 0.085	5.705 \pm 0.327	5.507 \pm 0.521	3.56 \pm 2.36	0.946
Chromium (Cr)	0.268 \pm 0.049	0.363 \pm 0.157	0.003 \pm 0.002	0.001 \pm 0.001	0.15 \pm 0.18	0.249
Aluminum (Al)	1.676 \pm 0.049	2.426 \pm 0.156	1.413 \pm 0.101	1.481 \pm 0.085	1.74 \pm 0.46	0.317
Iron (Fe)	6.501 \pm 0.205	12.315 \pm 1.026	12.530 \pm 0.626	11.978 \pm 0.534	10.8 \pm 2.89	2.013
Calcium (Ca)	1.855 \pm 0.040	4.097 \pm 0.213	5.584 \pm 0.401	4.192 \pm 0.435	3.93 \pm 1.54	0.953
Cadmium (Cd)	0.613 \pm 0.054	0.050 \pm 0.013	0.089 \pm 0.009	0.342 \pm 0.032	0.27 \pm 0.26	0.098
Lead (Pb)	BDL	BDL	BDL	BDL	BDL	-
Mercury (Hg)	BDL	BDL	BDL	BDL	BDL	-

CD=Critical difference; BDL=below detection limit; N.S= Non significant.

Table 17: Descriptive statistics obtained by ANOVA to compare concentration (mg/l) of the selected heavy metals of the water samples collected along Saidakadal of Dal Lake seasonally

Location	Seasons				Overall Mean	C.D
	Autumn (Mean ± SD)	Winter (Mean ± SD)	Spring (Mean ± SD)	Summer (Mean ± SD)		
Copper (Cu)	0.051 ± 0.013	0.025 ± 0.021	0.051 ± 0.010	0.730 ± 0.049	0.21±0.34	0.084
Zinc (Zn)	0.697 ± 0.012	0.396 ± 0.034	0.482 ± 0.033	1.016 ± 0.031	0.64±0.27	0.088
Cobalt (Co)	0.549 ± 0.038	0.378 ± 0.025	0.023 ± 0.011	0.014 ± 0.008	0.24±0.26	0.072
Nickel (Ni)	0.424 ± 0.074	0.053 ± 0.014	0.013 ± 0.008	0.004 ± 0.001	0.12±0.20	0.114
Manganese (Mn)	1.427 ± 0.075	4.379 ± 0.235	8.245 ± 0.194	2.857 ± 0.160	4.22±2.93	0.532
Chromium (Cr)	0.099 ± 0.030	0.291 ± 0.058	0.011 ± 0.006	0.005 ± 0.002	0.10±0.13	0.099
Aluminum (Al)	3.434 ± 0.061	1.769 ± 0.064	1.442 ± 0.036	2.639 ± 0.087	2.32±0.89	0.195
Iron (Fe)	6.698 ± 0.054	8.560 ± 0.544	8.945 ± 0.256	13.682 ± 0.369	9.47±2.97	1.070
Calcium (Ca)	1.365 ± 0.038	4.804 ± 0.239	4.705 ± 0.119	3.789 ± 0.088	3.66±1.60	0.429
Cadmium (Cd)	0.588 ± 0.031	0.475 ± 0.054	0.077 ± 0.010	0.063 ± 0.009	0.30±0.27	0.097
Lead (Pb)	BDL	BDL	BDL	BDL		-
Mercury (Hg)	BDL	BDL	BDL	BDL		-

CD=Critical difference; BDL=below detection limit; N.S= Non significant.

Table 18: Descriptive statistics obtained by ANOVA to compare concentration(mg/l) of the selected heavy metals of the water samples collected along Hazratbal basin of Dal Lake seasonally

Location	Seasons				Overall Mean	C.D
	Autumn (Mean ± SD)	Winter (Mean ± SD)	Spring(Mean ± SD)	Summer (Mean ± SD)		
Copper (Cu)	0.022 ± 0.012	0.046 ± 0.020	0.038 ± 0.006	0.063 ± 0.010	0.04±0.01	N.S.
Zinc (Zn)	0.556 ± 0.078	1.536 ± 0.186	0.062 ± 0.011	0.011 ± 0.006	0.54±0.70	0.305
Cobalt (Co)	0.555 ± 0.054	0.482 ± 0.070	0.006 ± 0.002	0.004 ± 0.001	0.26±0.29	0.133
Nickel (Ni)	0.325 ± 0.052	0.759 ± 0.061	0.183 ± 0.047	0.024 ± 0.015	0.32±0.31	0.142
Manganese (Mn)	1.433 ± 0.065	6.492 ± 0.673	4.719 ± 0.118	0.011 ± 0.007	3.16±2.96	1.037
Chromium (Cr)	0.254 ± 0.089	0.005 ± 0.002	0.002 ± 0.001	0.004 ± 0.001	0.06±0.12	0.135
Aluminum (Al)	2.533 ± 0.127	2.006 ± 0.256	6.851 ± 0.350	13.212 ± 1.181	6.15±5.18	1.912
Iron (Fe)	5.061 ± 0.471	2.373 ± 0.110	4.031 ± 0.293	5.665 ± 0.287	4.28±1.44	0.959
Calcium (Ca)	1.967 ± 0.228	5.094 ± 0.486	5.249 ± 0.326	4.155 ± 0.342	4.11±1.51	1.081
Cadmium (Cd)	0.717 ± 0.096	0.058 ± 0.011	0.378 ± 0.058	0.568 ± 0.079	0.43±0.28	0.207
Lead (Pb)	BDL	BDL	BDL	BDL		-
Mercury (Hg)	BDL	BDL	BDL	BDL		-

CD=Critical difference; BDL=below detection limit; N.S= Non significant.

Table 19: Descriptive statistics obtained by ANOVA to compare concentration(mg/l) of the selected heavy metals of the water samples collected along Telbal of Dal Lake seasonally

Location	Seasons				Overall Mean	C.D
	Autumn (Mean ± SD)	Winter (Mean ± SD)	Spring(Mean ± SD)	Summer (Mean ± SD)		
Copper (Cu)	0.036 ± 0.009	0.297 ± 0.067	0.439 ± 0.077	1.831 ± 0.234	0.65±0.80	0.386
Zinc (Zn)	0.517 ± 0.023	0.288 ± 0.078	0.271 ± 0.054	0.004 ± 0.002	0.27±0.20	0.148
Cobalt (Co)	0.494 ± 0.052	0.266 ± 0.048	0.379 ± 0.108	0.009 ± 0.003	0.28±0.20	0.196
Nickel (Ni)	0.683 ± 0.092	0.406 ± 0.018	0.987 ± 0.080	0.012 ± 0.009	0.52±0.41	0.187
Manganese (Mn)	1.657 ± 0.304	2.211 ± 0.197	5.337 ± 1.370	0.003 ± 0.001	2.30±2.23	2.142
Chromium (Cr)	0.071± 0.011	0.003 ± 0.001	0.002 ± 0.002	0.745 ± 0.057	0.20±0.36	0.088
Aluminum (Al)	0.488 ± 0.060	2.675 ± 0.239	6.054 ± 0.338	4.967 ± 0.502	3.54±2.47	0.988
Iron (Fe)	7.764 ± 0.262	4.869 ± 0.338	12.827 ± 1.062	32.178 ± 6.441	14.4±12.29	9.892
Calcium (Ca)	1.890 ± 0.126	3.427 ± 0.236	4.781 ± 0.159	3.868 ± 0.219	3.49±1.20	0.575
Cadmium (Cd)	0.441 ± 0.032	0.224 ± 0.033	0.067 ± 0.009	0.297 ± 0.018	0.25±0.15	0.076
Lead (Pb)	BDL	BDL	BDL	BDL		-
Mercury (Hg)	BDL	BDL	BDL	BDL		-

CD=Critical difference; BDL=below detection limit; N.S= Non significant.

Table 20: Descriptive statistics obtained by ANOVA to compare concentration (mg/l) of the selected heavy metals of the water samples collected along Zerobridge of River Jhelum seasonally

Location	Seasons				Overall Mean	C.D
	Autumn (Mean ± SD)	Winter (Mean ± SD)	Spring (Mean ± SD)	Summer (Mean ± SD)		
Copper (Cu)	0.005 ± 0.002	0.003 ± 0.001	0.535 ± 0.165	0.766 ± 0.077	0.32±0.38	0.276
Zinc (Zn)	0.422 ± 0.040	0.627 ± 0.056	0.077 ± 0.011	1.760 ± 0.345	0.72±0.72	0.532
Cobalt (Co)	0.656 ± 0.065	0.363 ± 0.041	0.058 ± 0.005	0.003 ± 0.001	0.27±0.30	0.116
Nickel (Ni)	0.537 ± 0.040	0.748 ± 0.037	1.274 ± 0.144	0.003 ± 0.002	0.64±0.52	0.233
Manganese (Mn)	1.446 ± 0.280	4.898 ± 0.340	0.019 ± 0.013	2.515 ± 0.105	2.21±2.05	0.685
Chromium (Cr)	0.244 ± 0.059	0.009 ± 0.005	0.016 ± 0.008	0.014 ± 0.007	0.07±0.11	0.091
Aluminum (Al)	2.519 ± 0.293	0.005 ± 0.002	5.131 ± 0.448	3.719 ± 0.263	2.84±2.17	0.902
Iron (Fe)	9.080 ± 0.429	2.564 ± 0.187	1.700 ± 0.218	1.459 ± 0.249	3.70±3.61	0.867
Calcium (Ca)	8.206 ± 0.583	8.013 ± 0.507	6.346 ± 0.273	1.659 ± 0.255	6.05±3.04	1.298
Cadmium (Cd)	0.406 ± 0.056	0.068 ± 0.013	0.023 ± 0.008	0.512 ± 0.050	0.25±0.24	0.115
Lead (Pb)	BDL	BDL	BDL	BDL		-
Mercury (Hg)	BDL	BDL	BDL	BDL		-

CD=Critical difference; BDL=below detection limit; N.S= Non significant.

Table 21: Descriptive statistics obtained by ANOVA to compare concentration (mg/l) of the selected heavy metals of the water samples collected along Chattabal weir of River Jhelum seasonally

Location	Seasons				Overall Mean	C.D
	Autumn (Mean ± SD)	Winter (Mean ± SD)	Spring (Mean ± SD)	Summer (Mean ± SD)		
Copper (Cu)	0.094 ± 0.063	0.003 ± 0.002	0.053 ± 0.010	0.468 ± 0.066	0.15±0.21	0.139
Zinc (Zn)	0.511± 0.044	0.438 ± 0.057	0.071 ± 0.013	0.789 ± 0.104	0.45±0.29	0.193
Cobalt (Co)	0.654 ± 0.042	1.880 ± 0.139	0.002 ± 0.001	0.003 ± 0.001	0.63±0.88	0.220
Nickel (Ni)	0.428 ± 0.041	0.429 ± 0.038	0.019 ± 0.007	0.006 ± 0.003	0.22±0.24	0.085
Manganese (Mn)	1.980 ± 0.377	9.480 ± 0.486	5.659 ± 0.186	2.034 ± 0.256	4.78±3.57	1.046
Chromium (Cr)	0.231 ± 0.028	0.002 ± 0.001	0.009 ± 0.004	0.018 ± 0.007	0.06±0.11	0.045
Aluminum (Al)	3.973 ± 0.444	2.096 ± 0.317	1.829 ± 0.284	4.718 ± 0.404	3.15±1.41	1.113
Iron (Fe)	2.581 ± 0.090	2.867 ± 0.195	11.459 ± 0.675	7.923 ± 2.495	6.20±4.27	3.921
Calcium (Ca)	2.488 ± 0.083	5.336 ± 0.412	4.187 ± 0.226	3.608 ± 0.407	3.90±1.18	0.949
Cadmium (Cd)	0.603 ± 0.022	0.347 ± 0.032	0.322 ± 0.036	0.341± 0.024	0.40±0.13	0.088
Lead (Pb)	BDL	BDL	BDL	BDL		-
Mercury (Hg)	BDL	BDL	BDL	BDL		-

CD=Critical difference; BDL=below detection limit; N.S= Non significant.

Table 22: Descriptive statistics obtained by ANOVA to compare concentration (mg/l) of the selected heavy metals of the water samples collected along Khannabal of River Jhelum seasonally

Location	Seasons				Overall Mean	C.D
	Autumn (Mean ± SD)	Winter (Mean ± SD)	Spring (Mean ± SD)	Summer (Mean ± SD)		
Copper (Cu)	0.012 ± 0.003	0.042 ± 0.014	2.420 ± 0.099	0.494 ± 0.054	0.74±1.14	0.172
Zinc (Zn)	0.495 ± 0.058	0.063 ± 0.007	1.811 ± 0.317	5.318 ± 0.373	1.92±2.38	0.745
Cobalt (Co)	0.805 ± 0.043	1.629 ± 0.389	0.004 ± 0.002	1.643± 0.376	1.02±0.78	0.820
Nickel (Ni)	0.521 ± 0.056	0.411 ± 0.071	0.006 ± 0.003	0.007 ± 0.002	0.23±0.26	0.137
Manganese (Mn)	2.676 ± 0.078	3.537 ± 0.252	3.344 ± 0.160	3.078 ± 0.187	3.15±0.37	0.546
Chromium (Cr)	0.364 ± 0.043	0.320 ± 0.108	0.003 ± 0.001	0.770 ± 0.072	0.36±0.31	0.207
Aluminum (Al)	4.525 ± 0.339	5.630 ± 0.283	1.996 ± 0.225	5.090 ± 0.524	4.31±1.60	1.091
Iron (Fe)	3.543 ± 0.389	13.801 ± 0.123	25.750 ± 1.375	7.615 ± 0.573	12.67±9.68	2.882
Calcium (Ca)	3.740 ± 0.863	7.526 ± 0.371	4.278 ± 0.298	3.478 ± 0.197	4.75±1.87	1.520
Cadmium (Cd)	0.399 ± 0.047	0.004 ± 0.002	1.999 ± 0.217	0.671 ± 0.087	0.76±0.86	0.361
Lead (Pb)	BDL	BDL	BDL	BDL	-	-
Mercury (Hg)	BDL	BDL	BDL	BDL	-	-

CD=Critical difference; BDL=below detection limit; N.S= Non significant.

Maximum concentration in Dal Lake was detected in summer at Telbal site (1.83 ± 0.23 mg/l) which was significantly ($p < 0.05$) higher than Saidakadal (0.73 ± 0.04 mg/l), Dalgate (0.33 ± 0.03 mg/l) and Hazratbal (0.06 ± 0.01 mg/l). Minimum concentration was detected in autumn at Saidakadal (0.05 ± 0.01 mg/l), Hazratbal (0.02 ± 0.01 mg/l), Dalgate (0.04 ± 0.01 mg/l), Telbal (0.03 ± 0.00 mg/l). During summer in River Jhelum maximum concentration was observed at Zerobridge (0.76 ± 0.07 mg/l) followed by Khannabal (0.49 ± 0.05 mg/l) and Chattabal weir (0.46 ± 0.06 mg/l). Minimum concentration was in autumn at Zerobridge (0.00 ± 0.00 mg/l) which was significantly ($p < 0.05$) lower value than at Khannabal (0.01 ± 0.00 mg/l) and Chattabal weir (0.09 ± 0.06 mg/l). However, concentrations in water samples from Hazratbal site show slight difference among seasons and did not vary significantly ($p > 0.05$). There was an increasing trend from autumn to summer at all the station. The minimum and maximum concentration recorded in River Jhelum and Dal Lake during different seasons was 0.00 to 0.76 mg/l and 0.02 to 1.83 mg/l, respectively which were higher than 1.00 mg/l, the Maximum Limit recommended by WHO/FEPA referred in Table-23.

4.3.2 Zinc (Zn)

The concentration of zinc in water among different stations varied significantly ($p < 0.05$). Overall the average concentration (Mean \pm SD) at Dalgate, Saidakadal, Hazratbal and Telbal of Dal Lake were 0.64 ± 0.60 mg/l, 0.64 ± 0.27 mg/l, 0.54 ± 0.70 mg/l and 0.27 ± 0.20 mg/l. At Zerobridge, Chattabal weir and Khannabal of River Jhelum these were 0.72 ± 0.72 mg/l, 0.45 ± 0.29 mg/l and 0.74 ± 1.14 mg/l, respectively. Significant ($p < 0.05$) variation existed overall concentrations in during different season at different stations. During summer the highest concentration of (5.31 ± 0.37 mg/l) was noted at Khannabal site followed by Zerobridge (1.76 ± 0.34 mg/l) and Chattabal Weir (0.78 ± 0.10 mg/l). Lowest concentration during spring was observed at Chattabal weir (0.07 ± 0.01 mg/l) followed by Zerobridge (0.07 ± 0.01). However, at Khannabal lowest concentration

(0.06±0.00) was found during winter. During summer in Dal lake highest concentration was at Dalgate (1.43±0.19 mg/l) and Saidakadal (1.01±0.03 mg/l). While during spring highest concentration was noted at Hazratbal (1.53±0.18) followed by Telbal (0.51±0.02 mg/l). Lowest concentrations during summer were at Telbal (0.00±0.00 mg/l) and Hazratbal (0.01±0.00 mg/l) while during winter lowest were recorded at Dalgate (0.03±0.00 mg/l) and Saidakadal (0.39±0.03 mg/l). The minimum and maximum concentration recorded in River Jhelum and Dal Lake during different seasons was 0.06 to 5.31 mg/l and 0.00 to 1.53, respectively, indicating that highest concentration more than the recommended value by WHO/FEPA 3.000 mg/l were found in River Jhelum than in Dal Lake (Table-23).

Table 23 : International guidelines for heavy metals in water and fish

Heavy Metals	Maximum Limit WHO (1984)/FEPA (mg/L) for water	Maximum Limit WHO (1984)/FEPA/ IAEA-407(ug/g) for fish
Copper(Cu)	0.84	3.28
Zinc(Zn)	4.65	30 (FAO, 1983)
Cobalt(Co)	0.004	-
Nickel(Ni)	0.1-0.2	0.60
Manganese (Mn)	0.050	0.5
Chromium(Cr)	0.05	0.73
Aluminum(Al)	-	-
Iron(Fe)	0.300	146 (Wyse <i>et al.</i> ,2003)
Calcium(Ca)	-	-
Cadmium (Cd)	0.003	0.18
Lead (Pb)	0.010	0.12
Mercury(Hg)	0.001	-

4.3.3 Cobalt (Co)

The concentration of cobalt was seen maximum during autumn season and minimum during summer season for all locations except Khannabal at which maximum concentration was in summer season. The average concentration (Mean±SD) at different locations, viz, Dalgate, Saidakadal, Hazratbal and Telbal of Dal Lake were 0.17 ± 0.25 mg/l, 0.24 ± 0.26 mg/l, 0.26 ± 0.29 mg/l and 0.28 ± 0.20 mg/l, respectively. In River Jhelum at Zerobridge, Chattabal weir and Khannabal the concentrations were 0.27 ± 0.30 mg/l, 0.63 ± 0.88 mg/l and 1.02 ± 0.78 mg/l, respectively. The concentrations during different seasons were significantly ($p<0.05$) different at various locations. Maximum concentration was detected in autumn for all locations with highest value in Hazratbal (0.55 ± 0.05 mg/l) which was significantly ($p<0.05$) higher than Saidakadal (0.54 ± 0.03 mg/l), Dalgate (0.53 ± 0.03 mg/l) and Telbal (0.49 ± 0.05 mg/l) of Dal Lake. In River Jhelum concentration at Zerobridge (0.65 ± 0.06 mg/l) was significantly ($p<0.05$) higher than Chattabal weir (0.65 ± 0.04 mg/l) where as at Khannabal maximum concentration was found in summer season (1.64 ± 0.37 mg/l). Significantly minimum ($p<0.05$) concentration during summer were found at all locations except Khannabal which revealed minimum concentration (0.004 ± 0.002 mg/l) during spring season. The minimum and maximum concentration recorded in River Jhelum and Dal Lake during different seasons was 0.002 to 1.64 mg/l and 0.003 to 0.55 were higher than 0.004 mg/l which is the Maximum Limit recommended by WHO/FEPA (Table8).

4.3.4 Nickel (Ni)

Overall the average concentrations (mean±sd) were 0.20 ± 0.23 mg/l, 0.12 ± 0.20 mg/l, 0.32 ± 0.31 mg/l, 0.52 ± 0.41 mg/l, 0.64 ± 0.52 mg/l, 0.22 ± 0.24 mg/l and 0.23 ± 0.26 mg/l at, Dalgate, Saidakadal, Hazratbal and Telbal of Dal Lake and Zerobridge, Chattabal weir and Khannabal of River Jhelum, respectively. The concentrations of nickel during different seasons varied significantly ($p<0.05$) at different locations. Minimum concentrations were found in summer season at all

locations. Maximum concentrations were observed in different seasons at different locations. Maximum level during autumn in River Jhelum were found at Khannabal (0.52 ± 0.05 mg/l) followed by Chattabal weir (0.42 ± 0.04 mg/l) and during spring at Zerobridge (1.27 ± 0.14 mg/l). In Dal Lake maximum levels were found at Dalgate (0.48 ± 0.05 mg/l) and Saidakadal (0.42 ± 0.07 mg/l) during autumn, at Hazratbal (0.75 ± 0.06 mg/l) during winter, and at Telbal (0.98 ± 0.08 mg/l) during spring. The minimum and maximum concentration recorded in River Jhelum and Dal Lake during different seasons was 0.003 to 1.27 mg/l and 0.003 to 0.98, respectively indicating that highest concentrations more than the recommended value by WHO/FEPA 0.1-0.2 mg/l were found in River Jhelum than in Dal Lake (Table-23).

4.3.5 Manganese (Mn)

The concentration of manganese in water differed significantly ($p < 0.05$) both location wise as well as seasonally. During spring highest concentrations in Dal Lake were noted at Saidakadal (8.24 ± 0.19 mg/l) followed by Dalgate (5.70 ± 0.32 mg/l) and Telbal (5.33 ± 1.37 mg/l) and highest level during winter was at Hazratbal (6.49 ± 0.67 mg/l). Lower concentrations were found during autumn at Saidakadal (1.42 ± 0.07 mg/l) followed by Dalgate (1.45 ± 0.08 mg/l) and during summer at Telbal (0.003 ± 0.001 mg/l) followed by Hazratbal (0.011 ± 0.007 mg/l). In River Jhelum concentrations were significantly ($p < 0.01$) higher during winter season at Chattabal weir (9.48 ± 0.48 mg/l) followed by Zerobridge (4.89 ± 0.34 mg/l) and Khannabal (3.53 ± 0.25 mg/l) site. Lower concentrations were detected during autumn at Chattabal weir (1.98 ± 0.37 mg/l) and Khannabal (2.67 ± 0.07 mg/l). During spring low level was at Zerobridge site (0.019 ± 0.013 mg/l). The average concentrations (Mean \pm SD) at different locations during different seasons at Dalgate, Saidakadal, Hazratbal and Telbal of Dal Lake were 3.56 ± 2.36 mg/l, 4.22 ± 2.93 mg/l, 3.16 ± 2.96 mg/l, 2.30 ± 2.23 mg/l whereas at Zerobridge, Chattabal weir and Khannabal of River Jhelum these were 2.21 ± 2.05 mg/l, 4.78 ± 3.57 mg/l and 3.15 ± 0.37 mg/l, respectively. The minimum and maximum concentrations in

River Jhelum and Dal Lake during different seasons were 0.01 to 9.48 mg/l and 0.003 to 8.24, respectively which were higher than 0.050 mg/l, the Maximum Limit recommended by WHO/FEPA referred in Table-23.

4.3.6 Chromium (Cr)

The concentration of chromium in water samples during different seasons were significantly ($p < 0.05$) varied at different locations. The average concentration (Mean \pm SD) at Dalgate, Saidakadal, Hazratbal and Telbal of Dal Lake were 0.15 \pm 0.18 mg/l, 0.10 \pm 0.13 mg/l, 0.06 \pm 0.12 mg/l and 0.20 \pm 0.36 mg/l, respectively when at Zerobridge, Chattabal weir and Khannabal of River Jhelum these were 0.07 \pm 0.11 mg/l, 0.06 \pm 0.11 mg/l and 0.36 \pm 0.31 mg/l, respectively. Maximum concentrations in Dal Lake during autumn were at Hazratbal (0.25 \pm 0.08 mg/l), during winter at Dalgate (0.36 \pm 0.15 mg/l) followed by Saidakadal (0.29 \pm 0.05 mg/l) and during summer at Telbal site (0.74 \pm 0.05). Minimum concentrations during summer were at Dalgate (0.003 \pm 0.002 mg/l), Hazratbal (0.004 \pm 0.001 mg/l) and Saidakadal (0.005 \pm 0.002 mg/l), and during winter at Telbal (0.003 \pm 0.001 mg/l). In River Jhelum maximum concentrations during autumn were at Zerobridge (0.244 \pm 0.059 mg/l) and Chattabal weir (0.23 \pm 0.02 mg/l) while as during summer it was at Khannabal (0.77 \pm 0.07 mg/l). Minimum concentrations during winter were at Zerobridge (0.009 \pm 0.005 mg/l) and Chattabal weir (0.002 \pm 0.001 mg/l) and during spring at Khannabal (0.003 \pm 0.001 mg/l). The minimum and maximum concentration recorded in River Jhelum and Dal Lake during different season was 0.002 to 0.7 mg/l and 0.003 to 0.7 were higher than 0.05 mg/l which is the Maximum Limit recommended by WHO/FEPA (Table-23).

4.3.7 Aluminum (Al)

The average concentration (Mean \pm SD) at Dalgate, Saidakadal, Hazratbal and Telbal of Dal Lake were 1.74 \pm 0.46 mg/l, 2.32 \pm 0.89 mg/l, 6.15 \pm 5.18 mg/l and 3.54 \pm 2.47 mg/l. At Zerobridge, Chattabal weir and Khannabal of River Jhelum

these were 2.84 ± 2.17 mg/l, 3.15 ± 1.41 mg/l and 4.31 ± 1.60 mg/l, respectively. The concentration of aluminum varied significantly ($p < 0.05$) at different locations seasonally. Highest concentrations were detected at Dalgate site (2.42 ± 0.15 mg/l) during winter, at Saidakadal (3.43 ± 0.06 mg/l) during autumn, at Hazratbal (13.21 ± 1.18 mg/l) during summer and at Telbal (6.05 ± 0.33 mg/l) during spring. Lowest concentrations also varied significantly ($p < 0.05$) and at Dalgate (1.41 ± 0.10 mg/l) and Saidakadal (1.44 ± 0.03 mg/l) during spring season were noted. Lowest concentrations were noted at Telbal (0.48 ± 0.06 mg/l) during autumn while at Hazratbal (2.00 ± 0.25 mg/l) during winter. In River Jhelum highest concentrations were observed at Khannabal (5.63 ± 0.28 mg/l) during winter, Chattabal weir (4.71 ± 0.40 mg/l) during summer and Zerobridge (5.13 ± 0.44 mg/l) during spring. Lowest concentrations during spring were at Chattabal weir (1.82 ± 0.28 mg/l) followed by Khannabal (1.99 ± 0.22 mg/l) while lowest value was recorded during winter at Zerobridge (0.005 ± 0.002 mg/l). The minimum and maximum concentrations recorded in River Jhelum and Dal Lake during different seasons was 0.005 to 5.63 mg/l and 0.48 to 13.21 mg/l which were higher than 0.0 mg/l which is the Maximum Limit recommended by WHO/FEPA (Table-23).

4.3.8 Iron (Fe)

The average concentrations (mean \pm sd) of iron in water samples from Dalgate, Saidakadal, Hazratbal and Telbal of Dal Lake were 10.8 ± 2.89 mg/l, 9.47 ± 2.97 mg/l, 4.28 ± 1.44 mg/l and 14.4 ± 12.29 mg/l, respectively. At Zerobridge, Chattabal weir and Khannabal of River Jhelum these were 3.70 ± 3.61 mg/l, 6.20 ± 4.27 mg/l and 12.67 ± 9.68 mg/l, respectively. The concentration of iron varied significantly ($p < 0.05$) among different locations seasonally. During autumn in River Jhelum lowest concentrations of (2.58 ± 0.09 mg/l) was noted at Chattabal weir followed by Khannabal (3.54 ± 25.7 mg/l) while during summer lowest concentration was at Zerobridge site (1.45 ± 0.24 mg/l). Highest concentrations during spring seasons were found at Khannabal (25.7 ± 1.37 mg/l) and Chattabal

weir (11.45 ± 0.67 mg/l) while at Zerobridge (9.08 ± 0.42 mg/l) highest level was found in autumn. In Dal Lake lowest level was detected at Dalgate (6.50 ± 0.20 mg/l) and Saidakadal (6.69 ± 0.05 mg/l) during autumn. Also lowest level was detected during winter at Hazratbal (2.37 ± 0.11 mg/l) and Telbal (4.86 ± 0.33 mg/l). Highest concentrations at Telbal (32.17 ± 6.44 mg/l), Saidakadal (13.68 ± 0.36 mg/l) and Hazratbal (5.66 ± 0.28 mg/l) were found during summer while during winter highest was recorded at Dalgate (12.53 ± 0.62 mg/l). The minimum and maximum concentrations recorded in River Jhelum and Dal Lake during different seasons were 1.45 to 25.7 mg/l and 2.3 to 32.1 mg/l and were higher than 0.300 mg/l which is the Maximum Limit recommended by WHO/FEPA (Table-23).

4.3.9 Calcium (Ca)

The concentration of calcium in water samples varied significantly ($p < 0.05$) from different locations seasonally. Overall the average concentrations (Mean \pm SD) were 3.93 ± 1.54 mg/l, 3.66 ± 1.60 mg/l, 4.11 ± 1.51 mg/l and 3.49 ± 1.20 mg/l, at Dalgate, Saidakadal, Hazratbal and Telbal of Dal Lake, and 6.05 ± 3.04 mg/l, 3.90 ± 1.18 mg/l and 4.75 ± 1.87 mg/l, at Zerobridge, Chattabal weir and Khannabal of River Jhelum, respectively. Maximum concentrations varied in different locations of Dal Lake with highest concentrations detected in spring at Dalgate (5.58 ± 0.40 mg/l), Hazratbal (5.24 ± 0.32 mg/l), Telbal (4.78 ± 0.15 mg/l) and Saidakadal (4.70 ± 0.11 mg/l). Minimum concentrations were detected in autumn at Saidakadal (0.06 ± 0.009 mg/l), Dalgate (1.85 ± 0.04 mg/l), Telbal (1.89 ± 0.12 mg/l) and Hazratbal (1.96 ± 0.22 mg/l). During winter in River Jhelum maximum concentrations were at Chattabal (5.33 ± 0.41 mg/l) followed by Khannabal (1.99 ± 0.21 mg/l). While during summer highest concentration was noted at Zerobridge (8.2 ± 0.58 mg/l) and at Chattabal weir (2.48 ± 0.08 mg/l) during autumn. Minimum concentration at Khannabal (0.004 ± 0.002 mg/l) and Zerobridge (1.65 ± 0.25 mg/l) observed during summer varied significantly ($p < 0.05$) from that of Chattabal weir (2.48 ± 0.08 mg/l) during autumn. The

minimum and maximum concentration recorded in River Jhelum and Dal Lake during different season was 0.004 to 5.3 mg/l and 0.06 to 5.58 mg/l.

4.3.10 Cadmium (Cd)

The concentration of cadmium in water samples varied significantly ($p < 0.05$) at different locations seasonally. The average concentration (mean) detected at different locations during different seasons at Dalgate, Saidakadal, Hazratbal and Telbal of Dal Lake were 0.27 ± 0.26 mg/l, 0.30 ± 0.27 mg/l, 0.43 ± 0.28 mg/l and 0.25 ± 0.15 mg/l while at Zerobridge, Chattabal weir and Khannabal of River Jhelum these were 0.25 ± 0.24 mg/l, 0.40 ± 0.13 mg/l and 0.76 ± 0.86 mg/l, respectively. The increase or decrease in concentrations of cadmium during different season vary significantly at different sites with highest concentrations detected during autumn at Hazratbal (0.71 ± 0.09 mg/l) site followed by Dalgate (0.61 ± 0.05 mg/l), Saidakadal (0.58 ± 0.03 mg/l) and Telbal (0.44 ± 0.03 mg/l). Lowest concentrations were detected during winter at Dalgate (0.05 ± 0.01 mg/l) and Hazratbal station (0.05 ± 0.01 mg/l). While during spring lowest concentration was noted at Telbal (0.06 ± 0.00 mg/l) and during summer lowest were recorded at Saidakadal (0.06 ± 0.00 mg/l). In River Jhelum during autumn highest concentrations were observed at Chattabal (0.60 ± 0.02 mg/l) and at Zerobridge (0.51 ± 0.05 mg/l) it was noted during summer while high level during spring season was noted at Khannabal (1.9 ± 0.21 mg/l). During winter lowest concentrations were detected at Khannabal (0.004 ± 0.002 mg/l). During spring lowest concentrations were noted at Zerobridge (0.023 ± 0.008 mg/l) and Chattabal weir (0.32 ± 0.03 mg/l). The minimum and maximum concentrations recorded in River Jhelum and Dal Lake during different seasons was 0.004 to 1.9 mg/l and 0.05 to 0.71 mg/l, higher than 0.003 mg/l which is the Maximum Limit recommended by WHO/FEPA (Table-23).

4.4 Metal concentration in fish tissue and parasites

The liver and muscle tissue pieces along with the parasites recovered

from the same *Schizothorax niger* were analyzed for the heavy metals. A total of 3 parasite species were recovered from *Schizothorax niger* from Dal Lake and River Jhelum. The parasites recovered were acanthocephalan parasite *Pomphorhynchus kashmirensis* and two intestinal cestodes, *Adenoscolex oreini* and *Bothriocephalus acheilognathi*. The concentrations of the metals in the tissues and the corresponding parasite are presented in the Tables 24 to 31.

The mean concentrations of the twelve heavy metals in the tissues of *Schizothorax niger* and their helminthes parasites in the two investigated water bodies are ranked in both infected and uninfected fishes. The mean concentrations of heavy metals for fish muscle, liver and *Adenoscolex* parasite of Dal Lake are ranked as Fe > Ca > Al > Mn > Zn > Cu > Ni > Cd > Cr > Co, Fe > Ca > Mn > Al > Cu > Zn > Cd > Ni > Co > Cr and Ca > Zn > Fe > Mn > Al > Cu > Co > Ni > Cd > Cr and of River Jhelum are ranked as Ca > Fe > Al > Zn > Cu > Cr > Ni > Mn > Co > Cd, Ca > Fe > Al > Zn > Cu > Mn > Ni > Co > Cd > Cr and Ca > Fe > Cr > Mn > Al > Zn > Cu > Cd > Ni > Co. The overall mean concentration of the twelve metals for fish muscle, liver and *Bothriocephalus* parasite of Dal Lake are ranked as Fe > Ca > Zn > Al > Cr > Ni > Cu > Mn > Cd > Co, Fe > Ca > Cu > Zn > Al > Mn > Ni > Cd > Cr > Co and Fe > Ca > Al > Zn > Mn > Cr > Cu > Ni > Cd > Co and of River Jhelum are ranked as Ca > Fe > Zn > Al > Cr > Mn > Ni > Cu > Co > Cd, Fe > Ca > Zn > Cu > Al > Mn > Ni > Cr > Cd > Co and Fe > Ca > Zn > Al > Cu > Mn > Cr > Ni > Co > Cd. The overall mean concentration of the twelve metals of Dal Lake for fish muscle, liver and *Pomphorhynchus* parasite are ranked as Cd > Ca > Cr > Zn > Cu > Mn > Al > Ni > Co > Cd, Ca > Fe > Zn > Al > Cu > Mn > Cr > Ni > Co > Cd and Ca > Fe > Mn > Cr > Cu > Ni > Zn > Al > Cd > Co and of River Jhelum are ranked as Zn > Fe > Ca > Cu > Cr > Al > Mn > Co > Ni > Cd, Ca > Fe > Zn > Al > Mn > Ni > Cu > Co > Cr > Cd and Ca > Fe > Mn > Cr > Al > Cu > Ni > Zn > Co > Cd.

Compared to the metal concentrations of muscle and liver tissues of infected fishes and the tissues concentrations of uninfected fishes from Dal Lake

were ranked as Fe > Ca > Zn > Cu > Ni > Cd > Al > Mn > Co > Cr and Fe > Cd > Cu > Ca > Al > Mn > Zn > Ni > Co > Cr and for River Jhelum as Ca > Fe > Al > Zn > Mn > Ni > Cr > Cu > Co > Cd and Fe > Ca > Al > Cu > Zn > Mn > Cd > Ni > Co > Cr, respectively.

Highly significant ($P < 0.01$) effects of the type of metal and the site of location were seen on their concentration in the tissues and parasites. Two factors interaction revealed high significance ($P < 0.01$) in the case of (heavy metal*site), significance ($P < 0.05$) in the case of (Fish*heavy metal) and non significance in the case of (Fish*site) while, three factors interaction (fish*site*heavy metal) showed high significance ($P < 0.01$) on the concentration of heavy metals in the infected fishes at the two sites.

4.4.1 Seasonal effect on the comparative concentrations of the heavy metals in tissues of the *Schizothoraxniger* and *Adenoscolex oreini* collected from Dal Lake (Table 24)

The highest concentration of copper was seen in fish liver (1.74 ± 0.009 $\mu\text{g/g}$) followed by *Adenoscolex oreini* (1.59 ± 2.78 $\mu\text{g/g}$) and fish muscle (1.25 ± 1.26 $\mu\text{g/g}$).

Zinc was seen more in *Adenoscolex* (6.60 ± 11.8 $\mu\text{g/g}$) than in fish muscle (1.30 ± 0.84 $\mu\text{g/g}$) and fish liver (1.16 ± 0.14 $\mu\text{g/g}$).

Cobalt was seen in *Adenoscolex* (0.54 ± 0.50 $\mu\text{g/g}$) more followed by fish muscle (0.38 ± 0.39 $\mu\text{g/g}$) and fish liver (0.09 ± 0.04 $\mu\text{g/g}$).

Nickel was seen higher in fish muscle (0.80 ± 0.90 $\mu\text{g/g}$) followed by fish liver (0.55 ± 0.01 $\mu\text{g/g}$) and *Adenoscolex* (0.52 ± 0.63 $\mu\text{g/g}$).

Manganese was seen in high in *Adenoscolex* (3.01 ± 3.32 $\mu\text{g/g}$) followed by fish liver (2.55 ± 0.01 $\mu\text{g/g}$) and fish muscle (1.69 ± 2.20 $\mu\text{g/g}$).

Chromium was seen more concentrated in fish muscle (0.74 ± 1.20 $\mu\text{g/g}$) compared to *Adenoscolex* (0.005 ± 0.006 $\mu\text{g/g}$) and fish liver (0.06 ± 0.081 $\mu\text{g/g}$).

Table 24: Seasonal effect on the comparative concentrations ($\mu\text{g/g}$) of the heavy metals in tissues of the *Schizothoraxniger* and *Adenoscolex oreini* collected from Dal Lake

Metals	Tissues																	
	<i>Adenoscolex oreini</i>						Liver						Muscle*					
	Autumn	Winter	Spring	Summer	Overall mean	CD	Autumn	Winter	Spring	Summer	Overall mean	CD	Autumn	Winter	Spring	Summer	Overall mean	CD
	Mean \pm SD	Mean \pm SD	Mean \pm SD	Mean \pm SD			Mean \pm SD	Mean \pm SD	Mean \pm SD	Mean \pm SD			Mean \pm SD	Mean \pm SD	Mean \pm SD	Mean \pm SD		
Copper (Cu)	0.033 \pm 0.018	0.005 \pm 0.002	0.079 \pm 0.022	6.254 \pm 0.387	1.59 \pm 2 .78	0.587	0.006 \pm 0.004	0.007 \pm 0.004	2.726 \pm 0.045	4.246 \pm 0.091	1.74 \pm 0 .009	0.154	0.014 \pm 0.004	0.168 \pm 0.023	2.957 \pm 0.155	1.871 \pm 0.036	1.25 \pm 1 .26	0.243
Zinc (Zn)	2.062 \pm 0.414	0.739 \pm 0.087	0.010 \pm 0.007	23.623 \pm 6.103	6.60 \pm 1 1.8	9.250	1.550 \pm 0.064	1.603 \pm 0.022	0.002 \pm 0.002	1.497 \pm 0.024	1.16 \pm 0 .14	0.109	1.252 \pm 0.053	2.141 \pm 0.056	0.009 \pm 0.004	1.821 \pm 0.054	1.30 \pm 0 .84	0.142
Cobalt (Co)	0.490 \pm 0.135	0.412 \pm 0.092	0.006 \pm 0.002	1.286 \pm 0.058	0.54 \pm 0 .50	0.261	0.188 \pm 0.018	0.003 \pm 0.002	0.003 \pm 0.001	0.196 \pm 0.023	0.09 \pm 0 .04	0.044	0.263 \pm 0.006	0.257 \pm 0.013	1.031 \pm 0.018	0.005 \pm 0.002	0.38 \pm 0 .39	0.035
Nickel (Ni)	0.574 \pm 0.124	1.477 \pm 0.095	0.030 \pm 0.017	0.005 \pm 0.002	0.52 \pm 0 .63	0.238	0.155 \pm 0.007	0.902 \pm 0.009	1.141 \pm 0.059	0.011 \pm 0.005	0.55 \pm 0 .01	0.092	0.275 \pm 0.029	0.699 \pm 0.045	2.257 \pm 0.089	0.003 \pm 0.002	0.80 \pm 0 .90	0.157
Manganese (Mn)	0.064 \pm 0.011	0.005 \pm 0.002	5.138 \pm 0.590	6.865 \pm 0.942	3.01 \pm 3 .32	1.681	0.053 \pm 0.005	5.329 \pm 0.142	1.305 \pm 0.049	3.538 \pm 0.142	2.55 \pm 0 .01	0.313	0.099 \pm 0.027	1.381 \pm 0.145	0.009 \pm 0.004	5.277 \pm 0.084	1.69 \pm 2 .20	0.256
Chromium (Cr)	0.004 \pm 0.002	0.006 \pm 0.004	0.006 \pm 0.003	0.005 \pm 0.003	0.005 \pm 0.006	N.S.	0.228 \pm 0.037	0.010 \pm 0.006	0.001 \pm 0.000	0.002 \pm 0.001	0.06 \pm 0 .081	0.056	0.197 \pm 0.024	0.008 \pm 0.004	2.779 \pm 0.094	0.011 \pm 0.006	0.74 \pm 1 .20	0.147
Aluminum (Al)	1.607 \pm 0.295	0.003 \pm 0.002	0.399 \pm 0.096	9.109 \pm 0.544	2.82 \pm 3 .95	0.947	1.696 \pm 0.095	1.023 \pm 0.254	2.566 \pm 0.346	5.477 \pm 1.562	2.21 \pm 0 .21	2.454	0.566 \pm 0.031	0.533 \pm 0.019	3.424 \pm 0.174	7.194 \pm 0.130	2.92 \pm 2 .80	0.333
Iron (Fe)	5.326 \pm 0.182	0.560 \pm 0.069	7.965 \pm 0.385	6.514 \pm 0.535	5.09 \pm 2 .93	1.040	27.55 \pm 14.76	66.68 \pm 10.77	23.76 \pm 1.703	16.25 \pm 2.028	33.7 \pm 2 .21	27.928	6.971 \pm 0.075	14.628 \pm 0.449	18.654 \pm 0.174	77.19 \pm 0.142	29.3 \pm 2 8.6	0.768
Calcium (Ca)	10.329 \pm 0.390	28.864 \pm 2.029	34.336 \pm 1.785	0.007 \pm 0.002	18.38 \pm 14.47	4.129	7.222 \pm 1.158	2.490 \pm 0.151	9.399 \pm 7.273	19.856 \pm 5.560	10.15 \pm 0.19	N.S.	24.58 \pm 0.193	6.034 \pm 0.019	9.852 \pm 0.221	74.92 \pm 3.636	28.8 \pm 2 8.4	5.515
Cadmium (Cd)	0.010 \pm 0.003	0.020 \pm 0.012	0.044 \pm 0.015	0.024 \pm 0.012	0.02 \pm 0 .026	N.S.	0.536 \pm 0.467	1.773 \pm 0.409	0.553 \pm 0.387	2.524 \pm 0.444	1.13 \pm 0 .027	1.294	0.042 \pm 0.007	0.005 \pm 0.005	0.030 \pm 0.010	3.059 \pm 0.475	0.78 \pm 1 .43	0.719
Lead (Pb)	BDL	BDL	BDL	BDL			BDL	BDL	BDL	BDL			BDL	BDL	BDL	BDL		
Mercury (Hg)	BDL	BDL	BDL	BDL			BDL	BDL	BDL	BDL			BDL	BDL	BDL	BDL		

Table 25: Seasonal effect on the comparative concentrations ($\mu\text{g/g}$) of the heavy metals in tissues of the *Schizothoraxniger* and *Adenoscolex oreini* collected from River Jhelum

Metals	Tissues																	
	<i>Adenoscolex oreini</i>						Liver						Muscle*					
	Autumn	Winter	Spring	Summer	Overall mean	CD	Autumn	Winter	Spring	Summer	Overall mean	CD	Autumn	Winter	Spring	Summer	Overall mean	CD
Mean \pm SD	Mean \pm SD	Mean \pm SD	Mean \pm SD	Mean \pm SD			Mean \pm SD	Mean \pm SD	Mean \pm SD	Mean \pm SD			Mean \pm SD	Mean \pm SD	Mean \pm SD	Mean \pm SD		
Copper (Cu)	0.069 \pm 0.033	0.327 \pm 0.064	0.070 \pm 0.006	3.481 \pm 0.124	0.98 \pm 1.48	0.217	1.171 \pm 0.294	0.286 \pm 0.281	1.228 \pm 0.202	0.617 \pm 0.164	0.83 \pm 0.060	0.729	0.002 \pm 0.002	0.001 \pm 0.001	2.612 \pm 0.654	0.805 \pm 0.621	0.86 \pm 1.42	1.363
Zinc (Zn)	1.425 \pm 0.085	0.347 \pm 0.038	0.020 \pm 0.008	22.842 \pm 0.678	6.15 \pm 9.92	1.035	1.347 \pm 0.058	1.208 \pm 0.303	0.321 \pm 0.316	1.221 \pm 0.256	1.07 \pm 0.054	0.772	1.089 \pm 0.246	1.912 \pm 0.143	0.402 \pm 0.400	2.083 \pm 0.521	1.49 \pm 1.00	1.083
Cobalt (Co)	0.355 \pm 0.045	0.029 \pm 0.005	0.472 \pm 0.133	0.040 \pm 0.011	0.22 \pm 0.24	0.213	0.185 \pm 0.039	0.057 \pm 0.017	0.162 \pm 0.160	0.780 \pm 0.068	0.280 \pm 0.031	0.270	0.906 \pm 0.440	0.101 \pm 0.099	0.145 \pm 0.041	0.019 \pm 0.019	0.159 \pm 0.19	0.685
Nickel (Ni)	0.523 \pm 0.041	0.597 \pm 0.039	0.014 \pm 0.004	0.005 \pm 0.001	0.28 \pm 0.28	0.086	0.601 \pm 0.039	0.580 \pm 0.133	0.042 \pm 0.011	0.080 \pm 0.078	0.33 \pm 0.04	0.240	0.213 \pm 0.055	0.526 \pm 0.083	1.976 \pm 0.360	0.516 \pm 0.512	0.80 \pm 0.95	0.959
Manganese (Mn)	0.391 \pm 0.016	0.014 \pm 0.006	6.806 \pm 0.273	19.756 \pm 0.298	6.74 \pm 8.20	0.611	0.459 \pm 0.124	0.851 \pm 0.068	0.588 \pm 0.054	0.360 \pm 0.065	0.57 \pm 0.026	0.249	0.118 \pm 0.033	0.461 \pm 0.074	0.102 \pm 0.099	0.451 \pm 0.117	0.30 \pm 0.24	0.262
Chromium (Cr)	0.230 \pm 0.039	0.018 \pm 0.005	44.438 \pm 2.429	0.018 \pm 0.008	11.1 \pm 19.85	3.673	0.143 \pm 0.037	0.002 \pm 0.002	0.004 \pm 0.002	0.147 \pm 0.145	0.04 \pm 0.037	N.S.	0.260 \pm 0.068	1.847 \pm 0.407	0.688 \pm 0.371	0.495 \pm 0.038	0.82 \pm 0.84	0.840
Aluminum (Al)	1.848 \pm 0.062	0.030 \pm 0.014	0.205 \pm 0.014	24.404 \pm 1.023	6.62 \pm 10.61	1.549	1.040 \pm 0.214	1.468 \pm 0.310	1.007 \pm 0.735	4.990 \pm 1.154	1.68 \pm 0.12	2.145	0.582 \pm 0.006	0.106 \pm 0.104	0.288 \pm 0.069	13.642 \pm 3.372	4.47 \pm 7.42	5.101
Iron (Fe)	17.552 \pm 0.563	0.645 \pm 0.094	25.616 \pm 0.434	24.686 \pm 1.011	17.12 \pm 10.35	1.874	8.208 \pm 1.388	4.679 \pm 1.943	15.29 \pm 1.28	15.758 \pm 1.883	11.05 \pm 0.08	4.994	7.527 \pm 2.369	2.397 \pm 0.651	15.08 \pm 3.351	13.69 \pm 1.168	9.47 \pm 6.66	6.525
Calcium (Ca)	1.751 \pm 0.102	0.253 \pm 0.029	72.718 \pm 1.591	37.632 \pm 0.630	28.08 \pm 30.62	2.592	7.000 \pm 1.375	20.02 \pm 18.44	86.07 \pm 12.06	28.66 \pm 7.468	35.84 \pm 0.073	35.246	22.174 \pm 2.357	5.779 \pm 4.390	9.596 \pm 2.089	65.44 \pm 13.331	28.85 \pm 30.3	21.748
Cadmium (Cd)	0.061 \pm 0.010	0.513 \pm 0.069	0.043 \pm 0.012	0.193 \pm 0.121	0.20 \pm 0.24	0.212	0.157 \pm 0.074	0.341 \pm 0.085	0.047 \pm 0.044	0.842 \pm 0.592	0.19 \pm 0.012	N.S.	15.09 \pm 15.028	0.007 \pm 0.004	0.050 \pm 0.015	0.540 \pm 0.130	0.19 \pm 0.277	N.S.
Lead (Pb)	BDL	BDL	BDL	BDL			BDL	BDL	BDL	BDL			BDL	BDL	BDL	BDL		
Mercury (Hg)	BDL	BDL	BDL	BDL			BDL	BDL	BDL	BDL			BDL	BDL	BDL	BDL		

CD=Critical difference; BDL=below detection limit; N.S= Non significant.

Table 26: Seasonal effect on the comparative concentrations ($\mu\text{g/g}$) of the heavy metals in tissues of the *Schizothoraxniger* and *Bothriocephalusacheilognathi* collected from Dal Lake

Metals	Tissues																	
	<i>Adenoscolex oreini</i>						Liver						Muscle*					
	Autumn	Winter	Spring	Summer	Overall mean	CD	Autumn	Winter	Spring	Summer	Overall mean	CD	Autumn	Winter	Spring	Summer	Overall mean	CD
Mean \pm SD	Mean \pm SD	Mean \pm SD	Mean \pm SD	Mean \pm SD			Mean \pm SD	Mean \pm SD	Mean \pm SD	Mean \pm SD			Mean \pm SD	Mean \pm SD	Mean \pm SD	Mean \pm SD		
Copper (Cu)	0.356 \pm 0.042	1.071 \pm 0.150	1.180 \pm 0.185	0.293 \pm 0.031	0.72 \pm 0. 48	0.369	5.765 \pm 1.464	9.040 \pm 2.057	0.887 \pm 0.067	1.769 \pm 1.077	4.27 \pm 0. 20	4.151	0.002 \pm 0.002	0.829 \pm 0.016	0.030 \pm 0.010	0.002 \pm 0.002	0.21 \pm 0. 36	0.029
Zinc (Zn)	2.411 \pm 0.304	9.610 \pm 0.588	2.091 \pm 0.235	3.285 \pm 0.062	4.34 \pm 3. 23	1.065	5.550 \pm 0.571	2.719 \pm 0.411	1.902 \pm 0.678	3.676 \pm 0.925	3.76 \pm 0. 084	2.034	3.107 \pm 0.045	10.814 \pm 0.350	4.390 \pm 0.024	4.435 \pm 0.117	5.68 \pm 3. 10	0.564
Cobalt (Co)	0.006 \pm 0.002	0.053 \pm 0.012	0.005 \pm 0.002	0.005 \pm 0.002	0.017 \pm 0. .02	0.020	0.011 \pm 0.004	0.006 \pm 0.005	0.001 \pm 0.001	0.018 \pm 0.016	0.005 \pm 0. .009	N.S.	0.001 \pm 0.001	0.176 \pm 0.018	0.001 \pm 0.001	0.001 \pm 0.001	0.04 \pm 0. 079	0.027
Nickel (Ni)	0.046 \pm 0.017	0.738 \pm 0.073	0.494 \pm 0.043	0.010 \pm 0.003	0.321 \pm 0. .32	0.131	0.117 \pm 0.049	0.265 \pm 0.061	0.087 \pm 0.051	0.548 \pm 0.252	0.18 \pm 0. 02	N.S.	0.271 \pm 0.022	0.551 \pm 0.008	0.050 \pm 0. .012	0.037 \pm 0.006	0.22 \pm 0. 21	0.040
Manganese (Mn)	5.135 \pm 0.227	3.885 \pm 0.265	3.524 \pm 0.251	0.544 \pm 0.130	3.27 \pm 1. 78	0.678	1.772 \pm 0.222	1.853 \pm 0.438	0.163 \pm 0.023	0.034 \pm 0.013	1.03 \pm 0. 14	0.743	0.148 \pm 0.013	0.555 \pm 0.012	0.045 \pm 0.002	0.064 \pm 0.002	0.20 \pm 0. 21	0.026
Chromium (Cr)	0.551 \pm 0.039	0.463 \pm 0.062	5.959 \pm 0.428	0.261 \pm 0.054	1.80 \pm 2. 50	0.661	0.031 \pm 0.027	0.156 \pm 0.023	0.130 \pm 0.017	0.222 \pm 0.196	0.08 \pm 0. 00	N.S.	0.449 \pm 0.021	0.373 \pm 0.015	0.314 \pm 0.005	0.066 \pm 0.025	0.30 \pm 0. 15	0.056
Aluminum (Al)	1.661 \pm 0.396	0.754 \pm 0.064	6.446 \pm 0.911	17.376 \pm 0.588	6.55 \pm 6. 88	1.748	0.957 \pm 0.080	1.027 \pm 0.331	2.535 \pm 0.183	12.88 \pm 10.14	1.72 \pm 0. 04	N.S.	0.563 \pm 0.014	0.442 \pm 0.007	3.401 \pm 0.115	7.581 \pm 0.058	2.99 \pm 2. 97	0.197
Iron (Fe)	9.048 \pm 0.303	3.563 \pm 0.415	42.366 \pm 5.536	48.557 \pm 2.951	25.88 \pm 2. 1.31	9.517	45.642 \pm 3.147	28.938 \pm 4.328	10.634 \pm 0.440	9.505 \pm 0.262	25.83 \pm 3. .56	8.128	53.40 \pm 0.577	46.83 \pm 1.418	6.248 \pm 0.015	3.569 \pm 0.084	27.51 \pm 23.3	2.318
Calcium (Ca)	3.982 \pm 0.266	8.197 \pm 0.163	19.429 \pm 2.410	50.931 \pm 4.703	20.63 \pm 1. 9.62	8.003	9.536 \pm 1.232	3.924 \pm 0.383	8.280 \pm 5.785	24.61 \pm 6.138	12.10 \pm 0. .51	12.900	2.491 \pm 0.032	6.483 \pm 0.025	3.453 \pm 0.054	42.36 \pm 0.280	13.69 \pm 17.04	0.436
Cadmium (Cd)	0.007 \pm 0.003	0.030 \pm 0.023	0.005 \pm 0.002	0.208 \pm 0.138	0.06 \pm 0. 16	N.S	0.180 \pm 0.042	0.067 \pm 0.027	0.038 \pm 0.036	0.417 \pm 0.262	0.10 \pm 0. 09	N.S	0.182 \pm 0.005	0.008 \pm 0.004	0.004 \pm 0.003	0.238 \pm 0.011	0.10 \pm 0. 10	0.020
Lead (Pb)	BDL	BDL	BDL	BDL			BDL	BDL	BDL	BDL			BDL	BDL	BDL	BDL		
Mercury (Hg)	BDL	BDL	BDL	BDL			BDL	BDL	BDL	BDL			BDL	BDL	BDL	BDL		

CD=Critical difference; BDL=below detection limit; N.S= Non significant.

Table 27: Seasonal effect on the comparative concentrations ($\mu\text{g/g}$) of the heavy metals in tissues of the *Schizothoraxniger* and *Bothriocephalus acheilognathi* collected from River Jhelum

Metals	Tissues																	
	<i>Adenoscolex oreini</i>						Liver						Muscle*					
	Autumn	Winter	Spring	Summer	Overall mean	CD	Autumn	Winter	Spring	Summer	Overall mean	CD	Autumn	Winter	Spring	Summer	Overall mean	CD
	Mean \pm SD	Mean \pm SD	Mean \pm SD	Mean \pm SD			Mean \pm SD	Mean \pm SD	Mean \pm SD	Mean \pm SD			Mean \pm SD	Mean \pm SD	Mean \pm SD	Mean \pm SD		
Copper (Cu)	0.513 \pm 0.105	0.009 \pm 0.004	0.557 \pm 0.057	3.945 \pm 0.122	1.25 \pm 1.61	0.259	4.593 \pm 1.318	7.554 \pm 1.892	0.066 \pm 0.064	8.788 \pm 8.380	3.29 \pm 0.11	N.S.	0.126 \pm 0.123	0.033 \pm 0.012	0.015 \pm 0.013	0.045 \pm 0.012	0.02 \pm 0.029	N.S.
Zinc (Zn)	1.413 \pm 0.072	0.335 \pm 0.052	4.315 \pm 0.110	6.152 \pm 0.234	3.05 \pm 2.38	0.413	38.920 \pm 3.303	21.168 \pm 3.604	5.516 \pm 0.879	1.573 \pm 0.396	18.90 \pm 0.66	7.534	1.428 \pm 0.383	7.305 \pm 1.346	5.284 \pm 0.817	1.601 \pm 0.653	3.93 \pm 3.08	2.641
Cobalt (Co)	0.491 \pm 0.082	0.015 \pm 0.006	0.032 \pm 0.015	0.007 \pm 0.002	0.13 \pm 0.22	0.126	0.002 \pm 0.001	0.002 \pm 0.002	0.001 \pm 0.001	0.006 \pm 0.003	0.002 \pm 0.002	N.S.	0.155 \pm 0.150	0.001 \pm 0.001	0.000 \pm 0.000	0.055 \pm 0.016	0.01 \pm 0.031	N.S.
Nickel (Ni)	0.522 \pm 0.068	0.808 \pm 0.094	0.452 \pm 0.114	0.007 \pm 0.002	0.44 \pm 0.33	0.245	0.052 \pm 0.045	0.288 \pm 0.072	0.050 \pm 0.046	0.424 \pm 0.147	0.15 \pm 0.01	0.266	0.375 \pm 0.097	0.370 \pm 0.070	0.062 \pm 0.059	0.064 \pm 0.017	0.21 \pm 0.021	0.203
Manganese (Mn)	0.188 \pm 0.054	0.498 \pm 0.055	3.506 \pm 0.160	0.588 \pm 0.147	1.19 \pm 1.39	0.349	0.995 \pm 0.148	1.199 \pm 0.300	0.024 \pm 0.022	0.095 \pm 0.040	0.62 \pm 0.11	0.511	0.203 \pm 0.045	0.631 \pm 0.046	0.173 \pm 0.112	0.176 \pm 0.040	0.29 \pm 0.023	0.205
Chromium (Cr)	0.068 \pm 0.009	0.341 \pm 0.036	0.399 \pm 0.140	3.869 \pm 0.244	1.16 \pm 1.63	0.429	0.004 \pm 0.003	0.005 \pm 0.004	0.030 \pm 0.010	0.177 \pm 0.175	0.01 \pm 0.006	N.S.	0.452 \pm 0.081	0.465 \pm 0.025	0.086 \pm 0.084	0.248 \pm 0.062	0.31 \pm 0.020	0.203
Aluminum (Al)	5.054 \pm 0.250	0.419 \pm 0.040	0.546 \pm 0.082	0.367 \pm 0.074	1.59 \pm 2.06	0.417	0.784 \pm 0.113	0.652 \pm 0.261	1.816 \pm 0.062	10.578 \pm 8.691	1.23 \pm 0.059	N.S.	0.774 \pm 0.128	1.436 \pm 0.893	0.842 \pm 0.108	2.214 \pm 0.342	1.44 \pm 1.17	N.S.
Iron (Fe)	8.859 \pm 0.284	0.426 \pm 0.050	1.957 \pm 0.359	16.262 \pm 0.807	6.87 \pm 6.51	1.404	37.33 \pm 7.154	8.906 \pm 0.055	12.815 \pm 4.039	23.390 \pm 3.873	22.4 \pm 1.15	13.733	35.927 \pm 8.318	12.624 \pm 7.399	3.672 \pm 0.392	6.796 \pm 0.926	14.9 \pm 7.16	16.900
Calcium (Ca)	1.539 \pm 0.097	0.389 \pm 0.037	0.546 \pm 0.041	20.454 \pm 0.998	5.73 \pm 8.79	1.518	6.853 \pm 0.913	2.889 \pm 0.454	5.999 \pm 4.918	18.798 \pm 4.702	9.02 \pm 0.32	10.402	2.584 \pm 1.164	2.872 \pm 0.441	2.568 \pm 0.233	45.13 \pm 1.715	15.74 \pm 23.78	16.312
Cadmium (Cd)	0.238 \pm 0.037	0.052 \pm 0.015	0.021 \pm 0.009	0.238 \pm 0.038	0.13 \pm 0.11	0.085	0.002 \pm 0.002	0.002 \pm 0.002	0.006 \pm 0.004	0.202 \pm 0.196	0.003 \pm 0.004	N.S.	11.20 \pm 11.199	0.001 \pm 0.001	0.002 \pm 0.002	0.017 \pm 0.004	0.006 \pm 0.008	N.S.
Lead (Pb)	BDL	BDL	BDL	BDL			BDL	BDL	BDL	BDL			BDL	BDL	BDL	BDL		
Mercury (Hg)	BDL	BDL	BDL	BDL			BDL	BDL	BDL	BDL			BDL	BDL	BDL	BDL		

CD=Critical difference; BDL=below detection limit; N.S= Non significant.

Table 28: Seasonal effect on the comparative concentrations ($\mu\text{g/g}$) of the heavy metals in tissues of the *Schizothoraxniger* and *Pomphorhynchus Kashmirensis* collected from Dal Lake

Metals	Tissues																	
	<i>Adenoscolex oreini</i>						Liver						Muscle*					
	Autumn	Winter	Spring	Summer	Overall mean	CD	Autumn	Winter	Spring	Summer	Overall mean	CD	Autumn	Winter	Spring	Summer	Overall mean	CD
Mean \pm SD	Mean \pm SD	Mean \pm SD	Mean \pm SD	Mean \pm SD			Mean \pm SD	Mean \pm SD	Mean \pm SD	Mean \pm SD			Mean \pm SD	Mean \pm SD	Mean \pm SD	Mean \pm SD		
Copper (Cu)	0.035 \pm 0.007	0.207 \pm 0.014	0.153 \pm 0.053	2.451 \pm 0.129	0.71 \pm 1.04	0.212	1.210 \pm 0.303	0.008 \pm 0.004	0.050 \pm 0.046	0.476 \pm 0.217	0.44 \pm 0.029	0.567	3.242 \pm 0.051	0.531 \pm 0.015	0.009 \pm 0.004	0.523 \pm 0.009	1.07 \pm 1.30	0.082
Zinc (Zn)	0.427 \pm 0.082	0.439 \pm 0.055	0.499 \pm 0.139	0.449 \pm 0.078	0.45 \pm 0.19	N.S.	1.402 \pm 0.047	1.285 \pm 0.317	0.018 \pm 0.009	0.111 \pm 0.064	0.75 \pm 0.060	0.494	2.298 \pm 0.039	2.266 \pm 0.023	1.513 \pm 0.026	1.324 \pm 0.020	1.85 \pm 0.45	0.085
Cobalt (Co)	0.068 \pm 0.012	0.535 \pm 0.105	0.038 \pm 0.017	0.109 \pm 0.035	0.18 \pm 0.23	0.170	0.441 \pm 0.113	0.002 \pm 0.001	0.071 \pm 0.069	0.426 \pm 0.075	0.24 \pm 0.099	0.229	0.040 \pm 0.015	0.153 \pm 0.014	0.007 \pm 0.003	0.008 \pm 0.005	0.051 \pm 0.065	0.031
Nickel (Ni)	0.463 \pm 0.059	0.009 \pm 0.004	0.295 \pm 0.030	1.108 \pm 0.162	0.46 \pm 0.45	0.264	0.310 \pm 0.117	0.583 \pm 0.146	0.060 \pm 0.043	0.343 \pm 0.017	0.31 \pm 0.05	0.292	0.263 \pm 0.015	0.439 \pm 0.013	0.015 \pm 0.011	0.009 \pm 0.006	0.18 \pm 0.18	0.035
Manganese (Mn)	0.672 \pm 0.075	0.008 \pm 0.004	4.676 \pm 0.247	1.044 \pm 0.105	1.59 \pm 1.88	0.421	0.394 \pm 0.141	0.584 \pm 0.154	0.223 \pm 0.072	0.424 \pm 0.054	0.40 \pm 0.02	N.S.	1.436 \pm 0.013	1.253 \pm 0.016	0.002 \pm 0.002	0.047 \pm 0.015	0.68 \pm 0.680	0.038
Chromium (Cr)	0.014 \pm 0.007	0.016 \pm 0.010	1.238 \pm 0.124	1.938 \pm 0.116	0.80 \pm 0.86	0.257	0.191 \pm 0.048	0.011 \pm 0.006	0.144 \pm 0.133	0.773 \pm 0.194	0.21 \pm 0.017	0.364	1.352 \pm 0.089	9.819 \pm 97.701	0.235 \pm 0.089	0.065 \pm 0.030	2.86 \pm 4.76	N.S.
Aluminum (Al)	0.363 \pm 0.039	0.532 \pm 0.043	0.009 \pm 0.004	0.917 \pm 0.196	0.45 \pm 0.39	0.310	1.286 \pm 0.312	0.048 \pm 0.006	0.061 \pm 0.034	3.408 \pm 3.253	0.45 \pm 0.063	N.S.	0.202 \pm 0.051	0.402 \pm 0.045	0.113 \pm 0.078	0.461 \pm 0.113	0.31 \pm 0.204	0.231
Iron (Fe)	7.566 \pm 0.078	4.758 \pm 0.582	28.998 \pm 2.878	21.860 \pm 1.761	15.79 \pm 10.85	5.178	24.988 \pm 7.862	42.50 \pm 10.628	0.076 \pm 0.032	17.30 \pm 16.74	17.82 \pm 0.64	N.S.	53.13 \pm 13.188	37.76 \pm 6.871	6.761 \pm 6.117	1.057 \pm 0.109	24.71 \pm 27.56	24.311
Calcium (Ca)	29.568 \pm 4.142	7.640 \pm 0.492	68.328 \pm 5.655	77.499 \pm 4.371	45.75 \pm 30.37	12.512	87.40 \pm 18.623	142.04 \pm 27.83	42.100 \pm 9.120	13.61 \pm 3.49	71.15 \pm 1.30	52.747	22.53 \pm 5.367	14.46 \pm 2.962	28.38 \pm 4.105	48.05 \pm 5.003	30.8 \pm 5.08	13.477
Cadmium (Cd)	0.020 \pm 0.010	0.014 \pm 0.008	0.016 \pm 0.008	0.890 \pm 0.354	0.23 \pm 0.53	0.536	0.040 \pm 0.010	0.055 \pm 0.012	0.043 \pm 0.015	0.307 \pm 0.290	0.04 \pm 0.034	N.S.	10.26 \pm 10.263	0.009 \pm 0.006	0.003 \pm 0.002	0.007 \pm 0.003	0.005 \pm 0.007	N.S.
Lead (Pb)	BDL	BDL	BDL	BDL			BDL	BDL	BDL	BDL			BDL	BDL	BDL	BDL		
Mercury (Hg)	BDL	BDL	BDL	BDL			BDL	BDL	BDL	BDL			BDL	BDL	BDL	BDL		

CD=Critical difference; BDL=below detection limit; N.S.= Non significant.

Table 29: Seasonal effect on the comparative concentrations ($\mu\text{g/g}$) of the heavy metals in tissues of the *Schizothoraxniger* and *Pomphorhynchus Kashmirensis* collected from River Jhelum

Metals	Tissues																	
	<i>Adenoscolex oreini</i>						Liver						Muscle*					
	Autumn	Winter	Spring	Summer	Overall mean	CD	Autumn	Winter	Spring	Summer	Overall mean	CD	Autumn	Winter	Spring	Summer	Overall mean	CD
Mean \pm SD	Mean \pm SD	Mean \pm SD	Mean \pm SD	Mean \pm SD			Mean \pm SD	Mean \pm SD	Mean \pm SD	Mean \pm SD			Mean \pm SD	Mean \pm SD	Mean \pm SD	Mean \pm SD		
Copper (Cu)	0.008 \pm 0.003	0.055 \pm 0.017	0.067 \pm 0.016	1.476 \pm 0.158	0.40 \pm 0.65	0.242	0.693 \pm 0.176	0.007 \pm 0.004	0.182 \pm 0.124	0.734 \pm 0.088	0.40 \pm 0.083	0.352	0.932 \pm 0.044	0.642 \pm 0.067	0.060 \pm 0.012	1.608 \pm 0.050	0.81 \pm 0.58	0.144
Zinc (Zn)	0.051 \pm 0.012	0.010 \pm 0.004	0.002 \pm 0.001	0.429 \pm 0.054	0.12 \pm 0.19	0.083	0.961 \pm 0.017	0.855 \pm 0.184	0.320 \pm 0.104	0.258 \pm 0.068	0.63 \pm 0.03	0.336	4.337 \pm 0.609	2.284 \pm 0.502	0.772 \pm 0.075	5.579 \pm 0.698	3.24 \pm 1.84	1.597
Cobalt (Co)	0.014 \pm 0.003	0.375 \pm 0.157	0.014 \pm 0.004	0.029 \pm 0.018	0.10 \pm 0.22	0.239	0.365 \pm 0.038	0.003 \pm 0.002	0.007 \pm 0.005	0.660 \pm 0.033	0.25 \pm 0.08	0.076	0.047 \pm 0.006	0.164 \pm 0.017	0.001 \pm 0.000	0.795 \pm 0.061	0.25 \pm 0.33	0.096
Nickel (Ni)	0.323 \pm 0.041	0.018 \pm 0.006	0.279 \pm 0.053	0.699 \pm 0.070	0.32 \pm 0.26	0.147	0.356 \pm 0.055	0.584 \pm 0.065	0.002 \pm 0.001	0.820 \pm 0.071	0.44 \pm 0.12	0.168	0.002 \pm 0.002	0.537 \pm 0.080	0.121 \pm 0.021	0.208 \pm 0.008	0.21 \pm 0.22	0.126
Manganese (Mn)	0.480 \pm 0.026	0.237 \pm 0.038	2.808 \pm 0.254	0.335 \pm 0.036	0.96 \pm 1.12	0.394	0.372 \pm 0.036	0.071 \pm 0.013	0.060 \pm 0.013	1.519 \pm 0.119	0.50 \pm 0.08	0.191	0.062 \pm 0.011	1.540 \pm 0.022	0.002 \pm 0.002	0.054 \pm 0.009	0.41 \pm 0.66	0.040
Chromium (Cr)	0.011 \pm 0.006	0.574 \pm 0.075	0.673 \pm 0.127	0.544 \pm 0.041	0.45 \pm 0.30	0.231	0.310 \pm 0.052	0.066 \pm 0.011	0.005 \pm 0.002	0.383 \pm 0.029	0.19 \pm 0.11	0.092	0.364 \pm 0.040	0.807 \pm 0.046	0.073 \pm 0.011	1.853 \pm 0.038	0.77 \pm 0.69	0.111
Aluminum (Al)	0.294 \pm 0.019	0.694 \pm 0.091	0.326 \pm 0.046	0.512 \pm 0.056	0.45 \pm 0.20	0.179	1.979 \pm 0.011	0.051 \pm 0.006	0.047 \pm 0.003	0.085 \pm 0.004	0.54 \pm 0.023	0.020	0.584 \pm 0.079	0.723 \pm 0.019	0.382 \pm 0.086	1.230 \pm 0.145	0.72 \pm 0.37	0.284
Iron (Fe)	5.855 \pm 0.201	2.253 \pm 0.429	27.690 \pm 1.943	20.908 \pm 0.949	14.17 \pm 10.98	3.346	19.218 \pm 0.279	55.58 \pm 0.549	3.491 \pm 0.115	9.617 \pm 0.122	21.97 \pm 0.62	0.965	43.748 \pm 0.704	23.104 \pm 0.338	18.77 \pm 0.316	12.310 \pm 0.141	24.4 \pm 2.10	1.291
Calcium (Ca)	37.576 \pm 3.647	6.804 \pm 0.371	76.292 \pm 3.333	84.174 \pm 1.997	51.21 \pm 32.39	8.077	66.988 \pm 0.465	18.932 \pm 0.252	0.934 \pm 0.040	33.316 \pm 0.506	30.0 \pm 1.03	1.108	22.690 \pm 0.365	9.760 \pm 0.333	5.438 \pm 0.169	16.480 \pm 0.172	13.59 \pm 6.75	0.832
Cadmium (Cd)	0.016 \pm 0.006	0.003 \pm 0.002	0.009 \pm 0.003	0.061 \pm 0.013	0.02 \pm 0.02	0.022	0.002 \pm 0.001	0.003 \pm 0.002	0.003 \pm 0.003	0.264 \pm 0.055	0.06 \pm 0.002	0.083	0.003 \pm 0.002	0.008 \pm 0.004	0.007 \pm 0.005	0.020 \pm 0.009	0.009 \pm 0.012	N.S.
Lead (Pb)	BDL	BDL	BDL	BDL			BDL	BDL	BDL	BDL			BDL	BDL	BDL	BDL		
Mercury (Hg)	BDL	BDL	BDL	BDL			BDL	BDL	BDL	BDL			BDL	BDL	BDL	BDL		

CD=Critical difference; BDL=below detection limit; N.S= Non significant.

Table 30: Seasonal effect on the comparative concentrations ($\mu\text{g/g}$) of the heavy metals in tissues of the *Schizothoraxniger* without parasites collected from Dal Lake

Metals	Tissues											
	Liver						Muscle*					
	Autumn Mean \pm SD	Winter Mean \pm SD	Spring Mean \pm SD	Summer Mean \pm SD	Overall mean	CD	Autumn Mean \pm SD	Winter Mean \pm SD	Spring Mean \pm SD	Summer Mean \pm SD	Overall mean	CD
Copper (Cu)	0.900 \pm 0.026	0.073 \pm 0.009	5.156 \pm 0.072	2.380 \pm 0.083	2.12 \pm 1. 98	0.171	0.005 \pm 0.002	0.008 \pm 0.005	4.394 \pm 0.126	0.746 \pm 0.030	1.28 \pm 1. 87	0.196
Zinc (Zn)	0.898 \pm 0.041	0.260 \pm 0.027	2.152 \pm 0.061	1.229 \pm 0.079	1.13 \pm 0. 70	0.168	1.137 \pm 0.052	5.960 \pm 0.028	1.680 \pm 0.044	2.439 \pm 0.145	2.80 \pm 1. 93	0.246
Cobalt (Co)	0.603 \pm 0.029	0.642 \pm 0.013	0.006 \pm 0.003	0.204 \pm 0.014	0.36 \pm 0. 27	0.053	0.505 \pm 0.026	0.545 \pm 0.033	0.055 \pm 0.008	0.007 \pm 0.004	0.27 \pm 0. 25	0.064
Nickel (Ni)	0.004 \pm 0.002	3.106 \pm 0.071	0.812 \pm 0.019	0.063 \pm 0.059	0.99 \pm 1. 29	0.143	0.325 \pm 0. 167	2.897 \pm 0.110	1.556 \pm 0.039	0.002 \pm 0.001	1.19 \pm 1. 18	0.308
Manganese (Mn)	0.484 \pm 0.039	0.009 \pm 0.007	3.937 \pm 0.125	1.211 \pm 0.073	1.41 \pm 1. 56	0.226	0.022 \pm 0.010	0.002 \pm 0.001	0.009 \pm 0.004	2.107 \pm 0.064	0.53 \pm 0. 93	0.098
Chromium (Cr)	0.054 \pm 0.015	0.003 \pm 0.002	0.006 \pm 0.004	0.001 \pm 0.001	0.01 \pm 0. 02	0.024	0.365 \pm 0.163	0.003 \pm 0.001	0.005 \pm 0.002	0.004 \pm 0.003	0.09 \pm 0. 23	0.246
Aluminum (Al)	3.082 \pm 0.052	2.283 \pm 0.174	1.143 \pm 112.4	1.344 \pm 0.184	1.96 \pm 0. 89	N.S.	1.334 \pm 0. 098	0.478 \pm 0.046	0.026 \pm 0.024	1.391 \pm 0.112	0.80 \pm 0. 61	0.237
Iron (Fe)	17.197 \pm 3.922	21.25 \pm 0.708	44.430 \pm 5.315	25.884 \pm 5.775	28.08 \pm 12.7	13.308	6.140 \pm 0.085	35.52 \pm 0.465	10.88 \pm 0.608	25.85 \pm 0.810	19.60 \pm 12.08	1.690
Calcium (Ca)	4.245 \pm 3.801	1.423 \pm 0.215	6.388 \pm 2.134	0.240 \pm 0.237	2.10 \pm 3. 40	N.S.	14.84 \pm 0.366	2.400 \pm 0.118	31.55 \pm 1.652	29.46 \pm 0.553	19.56 \pm 12.26	2.698
Cadmium (Cd)	0.133 \pm 0.034	3.720 \pm 0.696	1.553 \pm 0.756	7.560 \pm 1.762	3.65 \pm 3. 63	3.085	0.188 \pm 0.017	0.025 \pm 0.011	1.622 \pm 0.114	2.243 \pm 0.077	1.01 \pm 0. 97	0.210
Lead (Pb)	BDL	BDL	BDL	BDL			BDL	BDL	BDL	BDL		
Mercury (Hg)	BDL	BDL	BDL	BDL			BDL	BDL	BDL	BDL		

CD=Critical difference; BDL=below detection limit; N.S= Non significant.

Table 31: Seasonal effect on the comparative concentrations ($\mu\text{g/g}$) of the heavy metals in tissues of the *Schizothoraxniger* without parasites collected from River Jhelum

Metals	Tissues											
	Liver						Muscle*					
	Autumn Mean \pm SD	Winter Mean \pm SD	Spring Mean \pm SD	Summer Mean \pm SD	Overall mean	CD	Autumn Mean \pm SD	Winter Mean \pm SD	Spring Mean \pm SD	Summer Mean \pm SD	Overall mean	CD
Copper (Cu)	0.191 \pm 0.048	5.779 \pm 1.390	2.823 \pm 1.044	0.954 \pm 0.196	2.47 \pm 2. 81	2.646	0.323 \pm 0.165	0.002 \pm 0.002	2.482 \pm 0.072	0.284 \pm 0.028	0.77 \pm 1. 037	0.276
Zinc (Zn)	2.126 \pm 0.355	2.522 \pm 0.135	0.470 \pm 0.468	1.031 \pm 0.261	1.56 \pm 1. 06	0.993	1.355 \pm 0. 070	5.670 \pm 0.109	1.290 \pm 0.178	4.614 \pm 0.125	3.23 \pm 2. 01	0.383
Cobalt (Co)	0.590 \pm 0.159	0.140 \pm 0.074	0.011 \pm 0.008	0.146 \pm 0.037	0.17 \pm 0. 16	0.271	0.572 \pm 0.107	0.002 \pm 0.001	0.001 \pm 0.001	0.235 \pm 0.016	0.20 \pm 0. 26	0.163
Nickel (Ni)	0.593 \pm 0.109	0.713 \pm 0.072	0.763 \pm 0.017	0.143 \pm 0.139	0.54 \pm 0. 33	0.290	0.126 \pm 0.011	3.917 \pm 0.064	0.921 \pm 0.043	0.003 \pm 0.002	1.24 \pm 1. 62	0.118
Manganese (Mn)	0.383 \pm 0.098	1.322 \pm 0.246	0.818 \pm 0.155	0.637 \pm 0.039	0.81 \pm 0. 44	0.467	0.062 \pm 0.007	3.398 \pm 0.146	2.785 \pm 0.126	0.279 \pm 0.022	1.63 \pm 1. 53	0.294
Chromium (Cr)	0.372 \pm 0.053	0.109 \pm 0.108	0.004 \pm 0.002	0.006 \pm 0.005	0.09 \pm 0. 17	0.183	0.171 \pm 0.009	2.701 \pm 0.116	0.003 \pm 0.002	0.264 \pm 0.022	0.78 \pm 1. 14	0.180
Aluminum (Al)	1.824 \pm 0.458	0.402 \pm 0.399	0.590 \pm 0.148	9.362 \pm 2.178	3.62 \pm 4. 76	3.426	1.369 \pm 0.094	0.013 \pm 0.007	0.222 \pm 0.031	34.066 \pm 0.997	8.91 \pm 1 4.94	1.514
Iron (Fe)	20.43 \pm 2.278	9.654 \pm 3.540	38.24 \pm 7.916	25.39 \pm 4.412	23.90 \pm 14.4	15.108	5.339 \pm 0. 116	7.480 \pm 0.109	30.890 \pm 0.684	7.319 \pm 0.145	12.75 \pm 10.80	1.084
Calcium (Ca)	4.798 \pm 4.056	1.912 \pm 0.367	37.42 \pm 8.839	34.61 \pm 2.401	20.2 \pm 1 9.90	15.156	17.702 \pm 0.307	6.338 \pm 0.328	36.998 \pm 0.263	53.216 \pm 1.129	28.56 \pm 18.47	1.879
Cadmium (Cd)	6.590 \pm 6.320	0.250 \pm 0.018	0.058 \pm 0.054	0.750 \pm 0.189	0.35 \pm 0. 34	N.S	0.164 \pm 0.011	0.001 \pm 0.001	0.000 \pm 0.000	0.414 \pm 0.048	0.14 \pm 0. 180	0.075
Lead (Pb)	BDL	BDL	BDL	BDL			BDL	BDL	BDL	BDL		
Mercury (Hg)	BDL	BDL	BDL	BDL			BDL	BDL	BDL	BDL		

CD=Critical difference; BDL=below detection limit; N.S= Non significant.

Aluminum was seen in fish muscle ($2.92 \pm 2.80 \mu\text{g/g}$) more followed by *Adenoscolex* ($2.82 \pm 3.95 \mu\text{g/g}$) and fish liver ($2.21 \pm 0.21 \mu\text{g/g}$).

The highest concentration of iron was seen in fish liver ($33.7 \pm 2.21 \mu\text{g/g}$) followed by fish muscle ($29.36 \pm 28.6 \mu\text{g/g}$) and *Adenoscolex* ($5.09 \pm 2.93 \mu\text{g/g}$).

Calcium was more in fish muscle ($28.84 \pm 28.4 \mu\text{g/g}$) followed by *Adenoscolex* ($18.38 \pm 14.47 \mu\text{g/g}$) and fish liver ($10.15 \pm 0.19 \mu\text{g/g}$).

Cadmium was seen high in fish liver ($1.13 \pm 0.027 \mu\text{g/g}$) followed by fish muscle ($0.78 \pm 1.43 \mu\text{g/g}$) and *Adenoscolex* ($0.02 \pm 0.026 \mu\text{g/g}$).

4.4.2 Seasonal effect on the comparative concentrations of the heavy metals in tissues of the *Schizothorax niger* and *Adenoscolex oreini* collected from River Jhelum (Table 25)

The highest concentration of copper was seen in *Adenoscolex* ($0.98 \pm 1.48 \mu\text{g/g}$) followed by fish muscle ($0.86 \pm 1.42 \mu\text{g/g}$) and fish liver ($0.83 \pm 0.060 \mu\text{g/g}$).

Zinc was seen more in *Adenoscolex* ($6.15 \pm 9.92 \mu\text{g/g}$) followed by fish muscle ($1.49 \pm 1.00 \mu\text{g/g}$) and fish liver ($1.07 \pm 0.054 \mu\text{g/g}$).

The highest concentration of cobalt was seen in *Adenoscolex* ($0.22 \pm 0.24 \mu\text{g/g}$) followed by fish liver ($0.28 \pm 0.031 \mu\text{g/g}$) and fish muscle ($0.15 \pm 0.19 \mu\text{g/g}$).

Nickel was seen in fish liver ($0.33 \pm 0.04 \mu\text{g/g}$) more followed by fish muscle ($0.80 \pm 0.95 \mu\text{g/g}$) and *Adenoscolex* ($0.28 \pm 0.28 \mu\text{g/g}$).

Highest concentration of manganese was seen in *Adenoscolex* ($6.74 \pm 8.20 \mu\text{g/g}$) followed by fish liver ($0.57 \pm 0.026 \mu\text{g/g}$) and fish muscle ($0.30 \pm 0.24 \mu\text{g/g}$).

Chromium was seen more in *Adenoscolex* ($11.17 \pm 19.85 \mu\text{g/g}$) followed by fish muscle ($0.82 \pm 0.84 \mu\text{g/g}$) and fish liver ($0.04 \pm 0.037 \mu\text{g/g}$).

Aluminum was seen in *Adenoscolex* ($6.62 \pm 10.61 \mu\text{g/g}$) more than fish muscle ($4.47 \pm 7.42 \mu\text{g/g}$) and fish liver ($1.68 \pm 0.12 \mu\text{g/g}$).

Iron was seen more in *Adenoscolex* ($17.12 \pm 10.35 \mu\text{g/g}$) followed by fish liver ($11.05 \pm 0.08 \mu\text{g/g}$) and fish muscle ($9.47 \pm 6.66 \mu\text{g/g}$).

The highest concentration of calcium was seen in fish liver (35.84 ± 0.073 $\mu\text{g/g}$) followed by fish muscle (28.85 ± 30.32 $\mu\text{g/g}$) and *Adenoscolex* (28.08 ± 30.62 $\mu\text{g/g}$).

Cadmium was seen high in *Adenoscolex* (0.20 ± 0.24 $\mu\text{g/g}$) followed by fish liver (0.19 ± 0.012 $\mu\text{g/g}$) and fish muscle (0.19 ± 0.277 $\mu\text{g/g}$).

4.4.3 Seasonal effect on the comparative concentrations of the heavy metals in tissues of the *Schizothorax niger* and *Bothriocephalusacheilognathi* collected from Dal Lake (Table 26)

The highest concentration of copper was seen in fish liver (4.27 ± 0.20 $\mu\text{g/g}$) followed by *Bothriocephalus* (0.72 ± 0.48 $\mu\text{g/g}$) and fish muscles (0.21 ± 0.36 $\mu\text{g/g}$).

Zinc was seen more in fish muscles (5.68 ± 3.10 $\mu\text{g/g}$) followed by *Bothriocephalus* (4.34 ± 3.23 $\mu\text{g/g}$) and fish liver (3.76 ± 0.084 $\mu\text{g/g}$).

Cobalt was seen more in fish liver (0.005 ± 0.009 $\mu\text{g/g}$) followed by *Bothriocephalus* (0.017 ± 0.02 $\mu\text{g/g}$) and fish muscle (0.04 ± 0.079 $\mu\text{g/g}$).

Nickel was seen in *Bothriocephalus* (0.321 ± 0.32 $\mu\text{g/g}$) followed by fish muscle (0.22 ± 0.21 $\mu\text{g/g}$) and fish liver (0.18 ± 0.02 $\mu\text{g/g}$).

Manganese was seen more in *Bothriocephalus* (3.27 ± 1.78 $\mu\text{g/g}$) followed by fish liver (1.03 ± 0.14 $\mu\text{g/g}$) and fish muscle (0.20 ± 0.21 $\mu\text{g/g}$).

The highest concentration of chromium was seen in *Bothriocephalus* (1.80 ± 2.50 $\mu\text{g/g}$) followed by fish muscle (0.30 ± 0.15 $\mu\text{g/g}$) and fish liver (0.08 ± 0.00 $\mu\text{g/g}$).

Aluminum was seen more in *Bothriocephalus* (6.55 ± 6.88 $\mu\text{g/g}$) followed by fish muscle (2.99 ± 2.97 $\mu\text{g/g}$) and fish liver (1.72 ± 0.04 $\mu\text{g/g}$).

Iron was seen more in fish muscle (27.51 ± 23.3 $\mu\text{g/g}$) followed by fish liver (25.83 ± 3.56 $\mu\text{g/g}$) and *Bothriocephalus* (25.88 ± 21.31 $\mu\text{g/g}$).

The highest concentration of calcium was seen

in *Bothriocephalus* ($20.63 \pm 19.62 \mu\text{g/g}$) followed by fish muscle ($13.69 \pm 17.04 \mu\text{g/g}$) and fish liver ($12.10 \pm 0.51 \mu\text{g/g}$).

Cadmium was seen high in fish liver ($0.10 \pm 0.09 \mu\text{g/g}$) followed by fish muscle ($0.10 \pm 0.10 \mu\text{g/g}$) and *Bothriocephalus* ($0.06 \pm 0.16 \mu\text{g/g}$).

4.4.4 Seasonal effect on the comparative concentrations of the heavy metals in tissues of the *Schizothorax niger* and *Bothriocephalus acheilognathi* collected from River Jhelum (Table 27)

The highest concentration of copper was seen in fish liver ($3.29 \pm 0.11 \mu\text{g/g}$) followed by *Bothriocephalus* ($1.25 \pm 1.61 \mu\text{g/g}$) and fish muscle ($0.02 \pm 0.029 \mu\text{g/g}$).

Zinc was seen in fish liver ($18.90 \pm 0.66 \mu\text{g/g}$) followed by fish muscle ($3.93 \pm 3.08 \mu\text{g/g}$) and *Bothriocephalus* ($3.05 \pm 2.38 \mu\text{g/g}$).

Cobalt was seen in *Bothriocephalus* ($0.13 \pm 0.22 \mu\text{g/g}$) followed by fish muscle ($0.01 \pm 0.031 \mu\text{g/g}$) and fish liver ($0.002 \pm 0.002 \mu\text{g/g}$).

Nickel was seen more in *Bothriocephalus* ($0.44 \pm 0.33 \mu\text{g/g}$) followed by fish muscle ($0.21 \pm 0.21 \mu\text{g/g}$) and fish liver ($0.15 \pm 0.01 \mu\text{g/g}$).

Manganese was seen more in *Bothriocephalus* ($1.19 \pm 1.39 \mu\text{g/g}$) followed by fish liver ($0.62 \pm 0.11 \mu\text{g/g}$) and fish muscle ($0.29 \pm 0.23 \mu\text{g/g}$).

Chromium was seen more in *Bothriocephalus* ($1.16 \pm 1.63 \mu\text{g/g}$) followed by fish muscle ($0.31 \pm 0.20 \mu\text{g/g}$) and fish liver ($0.01 \pm 0.006 \mu\text{g/g}$).

Aluminum was seen more in *Bothriocephalus* ($1.59 \pm 2.06 \mu\text{g/g}$) followed by fish muscle ($1.44 \pm 1.17 \mu\text{g/g}$) and fish liver ($1.23 \pm 0.059 \mu\text{g/g}$).

The highest concentration of iron was seen in fish liver ($22.4 \pm 1.15 \mu\text{g/g}$) followed by fish muscle ($14.9 \pm 17.16 \mu\text{g/g}$) and *Bothriocephalus* ($6.87 \pm 6.51 \mu\text{g/g}$).

Calcium was seen more in fish muscle ($15.74 \pm 23.78 \mu\text{g/g}$) followed by fish liver ($9.02 \pm 0.32 \mu\text{g/g}$) and *Bothriocephalus* ($5.73 \pm 8.79 \mu\text{g/g}$).

The highest concentration of Cadmium was seen in *Bothriocephalus* ($0.13 \pm 0.11 \mu\text{g/g}$) followed by fish muscle ($0.006 \pm 0.008 \mu\text{g/g}$) and fish liver ($0.003 \pm 0.004 \mu\text{g/g}$).

4.4.5 Seasonal effect on the comparative concentrations of the heavy metals in tissues of the *Schizothoraxniger* and *Pomphorhynchus Kashmirensis* collected from Dal Lake (Table 28)

The highest concentration of copper was seen in fish muscle ($1.07 \pm 1.30 \mu\text{g/g}$) followed by *Pomphorhynchus* ($0.71 \pm 1.04 \mu\text{g/g}$) and fish liver ($0.44 \pm 0.029 \mu\text{g/g}$).

Zinc was seen more in fish muscle ($1.85 \pm 0.45 \mu\text{g/g}$) followed by fish liver ($0.75 \pm 0.060 \mu\text{g/g}$) and *Pomphorhynchus* ($0.45 \pm 0.19 \mu\text{g/g}$).

Cobalt was seen more in fish liver ($0.24 \pm 0.099 \mu\text{g/g}$) followed by *Pomphorhynchus* ($0.18 \pm 0.23 \mu\text{g/g}$) and fish muscle ($0.051 \pm 0.065 \mu\text{g/g}$).

Nickel was seen more in *Pomphorhynchus* ($0.46 \pm 0.45 \mu\text{g/g}$) followed by fish liver ($0.31 \pm 0.05 \mu\text{g/g}$) and fish muscle ($0.18 \pm 0.18 \mu\text{g/g}$).

The highest concentration of manganese was seen in *Pomphorhynchus* ($1.59 \pm 1.88 \mu\text{g/g}$) followed by fish muscle ($0.68 \pm 0.680 \mu\text{g/g}$) and fish liver ($0.40 \pm 0.02 \mu\text{g/g}$).

Chromium was seen more in fish muscle ($2.86 \pm 4.86 \mu\text{g/g}$) followed by *Pomphorhynchus* ($0.80 \pm 0.86 \mu\text{g/g}$) and fish liver ($0.21 \pm 0.017 \mu\text{g/g}$).

Aluminum was seen more in fish liver ($0.45 \pm 0.063 \mu\text{g/g}$) followed by *Pomphorhynchus* ($0.45 \pm 0.39 \mu\text{g/g}$) and fish muscle ($0.31 \pm 0.204 \mu\text{g/g}$).

The highest concentration of iron was seen in fish muscle ($24.71 \pm 27.56 \mu\text{g/g}$) followed by fish liver ($17.82 \pm 0.64 \mu\text{g/g}$) and *Pomphorhynchus* ($15.79 \pm 10.85 \mu\text{g/g}$).

Concentration of calcium was more in fish liver ($71.15 \pm 11.30 \mu\text{g/g}$) followed by *Pomphorhynchus* ($45.75 \pm 30.37 \mu\text{g/g}$) and fish muscle ($30.8 \pm 15.08 \mu\text{g/g}$).

Cadmium was more in *Pomphorhynchus* (0.23 ± 0.53 $\mu\text{g/g}$) followed by fish liver (0.04 ± 0.034 $\mu\text{g/g}$) and fish muscle (0.005 ± 0.007 $\mu\text{g/g}$).

4.4.6 Seasonal effect on the comparative concentrations of the heavy metals in tissues of the *Schizothoraxniger* and *Pomphorhynchus Kashmirensis* collected from River Jhelum (Table 29)

The highest concentration of copper was seen in fish muscle (0.81 ± 0.58 $\mu\text{g/g}$) followed by *Pomphorhynchus* (0.40 ± 0.65 $\mu\text{g/g}$) and fish liver (0.40 ± 0.083 $\mu\text{g/g}$).

Concentration of zinc was more seen in fish muscle (3.24 ± 1.84 $\mu\text{g/g}$) followed by fish liver (0.63 ± 0.03 $\mu\text{g/g}$) and *Pomphorhynchus* (0.12 ± 0.19 $\mu\text{g/g}$).

Cobalt was seen more in fish liver (0.25 ± 0.08 $\mu\text{g/g}$) followed by fish muscle (0.25 ± 0.33 $\mu\text{g/g}$) and *Pomphorhynchus* (0.10 ± 0.22 $\mu\text{g/g}$).

The highest concentration of nickel was seen in fish liver (0.44 ± 0.12 $\mu\text{g/g}$) followed by *Pomphorhynchus* (0.32 ± 0.26 $\mu\text{g/g}$) and fish muscle (0.21 ± 0.22 $\mu\text{g/g}$).

Manganese was seen more in *Pomphorhynchus* (0.96 ± 1.12 $\mu\text{g/g}$) followed by fish liver (0.50 ± 0.08 $\mu\text{g/g}$) and fish muscle (0.41 ± 0.66 $\mu\text{g/g}$).

Chromium was seen more in fish muscle (0.77 ± 0.69 $\mu\text{g/g}$) followed by *Pomphorhynchus* (0.45 ± 0.30 $\mu\text{g/g}$) and fish liver (0.19 ± 0.11 $\mu\text{g/g}$).

Aluminum was seen more in fish muscle (0.72 ± 0.37 $\mu\text{g/g}$) followed by fish liver (0.54 ± 0.023 $\mu\text{g/g}$) and *Pomphorhynchus* (0.45 ± 0.20 $\mu\text{g/g}$).

Iron was seen more in fish muscle (24.4 ± 12.10 $\mu\text{g/g}$) followed by fish liver (21.97 ± 0.62 $\mu\text{g/g}$) and *Pomphorhynchus* (14.17 ± 10.98 $\mu\text{g/g}$).

The highest concentration of calcium was seen in *Pomphorhynchus* (51.21 ± 32.39 $\mu\text{g/g}$) followed by fish liver (30.0 ± 1.03 $\mu\text{g/g}$) and fish muscle (13.59 ± 6.75 $\mu\text{g/g}$).

Cadmium was seen more in fish liver (0.06 ± 0.002 $\mu\text{g/g}$) followed by

Pomphorhynchus (0.02±0.02 µg/g) and fish muscle (0.009±0.012 µg/g).

4.4.7 Seasonal effect on the comparative concentrations of the heavy metals in tissues of the *Schizothoraxniger* without parasite from Dal Lake (Table 30)

Copper (1.28±1.87 µg/g), zinc (2.80±1.93 µg/g), nickel (1.19±1.18 µg/g), chromium (0.09±0.23 µg/g) and calcium (19.56±12.26 µg/g) were high in muscle than the respective liver 2.12±1.98 µg/g, 1.13±0.70 µg/g, 0.99±1.29 µg/g, 0.01±0.02 µg/g and 2.10±3.40 µg/g of the *Schizothorax niger*. On the other hand cobalt, manganese, aluminum, iron and cadmium levels were 0.36±0.27 µg/g, 1.41±1.56 µg/g, 1.96±0.89 µg/g, 28.08±12.7 µg/g, and 3.65±3.63 µg/g which were higher in liver than their respective muscle tissue 0.27±0.25 µg/g, 0.53±0.93 µg/g, 0.80±0.61 µg/g, 19.60±12.08 µg/g and 1.01±0.97 µg/g.

4.4.8 Seasonal effect on the comparative concentrations of the heavy metals in tissues of the *Schizothoraxniger* without parasite from River Jhelum (Table 31)

Copper (2.47±2.81 µg/g), chromium (0.09±0.17 µg/g), iron (23.90±14.4 µg/g) and cadmium (0.35±0.34 µg/g) were high in livers than the respective muscles 0.77±1.037 µg/g, 0.78±1.14 µg/g, 12.75±10.80 µg/g, 0.14±0.180 µg/g of the *Schizothorax niger*. On the other hand muscle tissues were having high levels of zinc (3.23±2.01 µg/g), cobalt (0.20±0.26 µg/g), nickel (1.24±1.62 µg/g), manganese (1.63±1.53 µg/g), aluminum (8.91±14.94 µg/g) and calcium (28.56±18.47 µg/g) than livers 1.56±1.06 µg/g, 0.17±0.16 µg/g, 0.54±0.33 µg/g, 0.81±0.44 µg/g, 3.62±4.76 µg/g and 20.2±19.90 µg/g respectively.

4.5 Minimum and maximum concentration recorded in Dal Lake and River Jhelum in fish muscle during different season in the infected fishes for various metals viz-a-viz recommended levels (Table 23)

In Dal Lake the maximum concentration of copper (1.25 µg/g), zinc (5.69 µg/g), iron (29.3 µg/g) in the muscles of infected fishes were still lower than the maximum limit recommended by WHO/FEPA/IAEA-407 whereas on the other the concentration of nickel (0.80 µg/g), manganese (1.69 µg/g), cadmium (0.78 µg/g) and chromium (2.86 µg/g) were higher than the permissible limits.

However, with regard to cobalt (0.38 µg/g), aluminum (2.99 µg/g) and calcium (30.8 µg/g) the reference values for permissible limit were not available.

In River Jhelum the maximum concentration of nickel (0.80 µg/g), manganese (0.41 µg/g), cadmium (0.19 µg/g) and chromium (0.82 µg/g) were higher than the permissible limits. Whereas on the other the concentration of copper (0.86 µg/g), zinc (3.93 µg/g), iron (24.4 µg/g) in the muscles of infected fishes were still lower than the Maximum Limit recommended by WHO/FEPA/IAEA-407. However, with regard to cobalt (0.25 µg/g), aluminum (4.47 µg/g) and calcium (28.8 µg/g) the reference values for permissible limit were not available.

4.6 Correlation studies

The correlation tables are presented in Tables 1 to 96 (Appendices).

4.6.1 Correlation among metal concentrations of water, fish tissues, and parasites, and physicochemical parameters of Dal Lake

During summer Al and Cu concentrations in livers of the fishes, infected with *Adenoscolex* revealed negative correlation ($p < 0.05$, $r = -0.922$ and $p < 0.05$, $r = -0.930$) respectively, with water. Al and Cr concentrations in livers of fishes, infected with *Bothriocephalus*, showed negative correlation ($p < 0.01$, $r = -0.973$ and $p < 0.05$, $r = -0.900$) respectively. Cd concentration in muscle of fishes infected with *Bothriocephalus* was found negatively correlated ($p < 0.05$, $r = -0.955$) in water levels.

Water temperature depicted significant positive correlation with Al concentration ($p < 0.05$, $r = 0.927$) and Ca concentration ($p < 0.05$, $r = 0.957$) in water. Free carbon dioxide seems to be positively correlated with Fe concentrations ($p < 0.05$, $r = 0.914$) and negatively with dissolved oxygen ($p < 0.05$, $r = -0.953$) and pH ($p < 0.05$, $r = -0.905$). Free carbon dioxide had positive correlation with Zn concentration ($p < 0.05$, $r = 0.885$).

Cu concentration in *Pomphorhynchus* was found positively correlated

with temperature ($p < 0.05$, $r = 0.949$). Fe concentration in liver of fishes infected with *Pomphorhynchus* was positively correlated with temperature ($p < 0.01$, $r = 0.988$). Cd concentration in the liver infected with *Pomphorhynchus* was found positively correlated with, dissolved oxygen ($p < 0.05$, $r = 0.883$) and free carbon dioxide ($p < 0.05$, $r = 0.902$).

During spring Cu concentration in *Pomphorhynchus* was negatively correlated ($p < 0.05$, $r = -0.897$) with water levels. *Bothriocephalus* had shown negative correlation with Ni concentration of water ($p < 0.01$, $r = -0.995$). Mn concentration in liver of fishes infected with *Adenoscolex* showed negative correlation ($p < 0.05$, $r = -0.890$) with water levels. Cr concentration in liver of fishes infected with *Bothriocephalus* showed negative correlation ($p < 0.05$, $r = -0.947$) with water. Fe concentration of *Adenoscolex* was found negatively correlated with water level ($p < 0.05$, $r = -0.929$). Cd concentration had positive correlation in the muscle tissue of fishes infected with *Pomphorhynchus* ($p < 0.05$, $r = 0.958$).

pH showed negative correlation with Cu concentration ($p < 0.05$, $r = -0.933$) whereas water temperature was positively correlated with Mn concentration ($p < 0.05$, $r = 0.879$) of water. Dissolved oxygen was found positively correlated with Al concentration of water ($p < 0.05$, $r = 0.939$). Free carbon dioxide depicted positive correlation with Cd concentration ($p < 0.05$, $r = 0.918$) in water levels.

Cu concentration in Liver of fishes infected with *Adenoscolex* was found to be positively correlated with temperature ($p < 0.05$, $r = 0.916$) whereas *Pomphorhynchus* with pH ($p < 0.05$, $r = 0.883$) of water. Zn concentration in *Bothriocephalus* showed positive correlation with pH ($p < 0.05$, $r = 0.885$) of water. Mn concentration in Liver of fish infected with *Adenoscolex* was found positively correlated with pH ($p < 0.01$, $r = 0.885$) of water. Muscle tissue of uninfected fish had positive correlation with free carbon dioxide ($p < 0.05$, $r = 0.892$). Cr concentration in *Pomphorhynchus* showed positive correlation with pH ($p < 0.01$, $r =$

0.970). Al concentration in Liver of fish infected with *Bothriocephalus* showed positive correlation with free carbon dioxide ($p < 0.01$, $r = -0.973$). Ca concentration in liver infected with *Pomphorhynchus* was positively correlated with pH ($p < 0.01$, $r = 0.961$).

During autumn Zn concentration in liver of fishes infected with *Adenoscolex* and muscle of fishes infected with *Bothriocephalus* revealed significant negative correlation ($p < 0.05$, $r = -0.947$ and $p < 0.05$, $r = -0.958$) respectively, with water. Ni concentration in muscle of fishes infected with *Bothriocephalus* was observed to have a high positive significant correlation ($p < 0.01$, $r = 0.991$) with water. Cr concentration in muscle of fishes infected with *Bothriocephalus* showed positive correlation ($p < 0.01$, $r = 0.991$) with water. Ca concentration was negatively correlated with liver of fishes infected with *Pomphorhynchus* ($p < 0.05$, $r = -0.949$) and Cd concentration was found negatively correlated with muscle of fishes infected with *Bothriocephalus* ($p < 0.05$, $r = -0.893$) in water.

Water temperature had positive correlation with Zn concentration ($p < 0.01$, $r = 0.982$) and negative correlation with Fe concentration ($p < 0.05$, $r = -0.952$), Ca concentration ($p < 0.05$, $r = -0.936$) and Cd concentration ($p < 0.05$, $r = -0.902$) in water levels also, free carbon dioxide was positively correlated with Cd concentration ($p < 0.05$, $r = 0.911$).

Cu concentration in *Adenoscolex* parasite revealed significant positive correlation ($p < 0.05$, $r = 0.909$) with water temperature. Zn concentration in the muscle tissue of fish infected with *Bothriocephalus* was positively correlated with temperature ($p < 0.05$, $r = 0.929$). Co concentration in the liver infected with *Pomphorhynchus* depicted positive correlation with free carbon dioxide ($p < 0.05$, $r = 0.893$). Ni concentration in liver of uninfected fish was positively correlated with free carbon dioxide ($p < 0.05$, $r = 0.936$). Mn concentration in *Pomphorhynchus* depicted significant positive correlation with free carbon dioxide ($p < 0.05$, $r = 0.938$) and the liver of same fish infected with *Pomphorhynchus* had positive

correlation with muscle tissue ($p < 0.01$, $r = 0.892$). Fe concentration in the liver infected with *Pomphorhynchus* showed positive correlation with pH ($p < 0.01$, $r = 0.976$). Ca concentration in muscle tissue infected with *Pomphorhynchus* was positively correlated with temperature ($p < 0.05$, $r = 0.891$) and pH ($p < 0.05$, $r = -0.931$). Cd concentration in the muscle tissue infected with *Bothriocephalus* showed positive correlation with temperature ($p < 0.05$, $r = 0.943$) and pH ($p < 0.05$, $r = 0.923$).

During winter Co concentration in muscle tissues of fishes infected with *Adenoscolex* depicted significant positive correlation ($p < 0.05$, $r = 0.909$) with water. Positive correlation existed between the Mn concentration of liver of fishes infected with *Pomphorhynchus* and water ($p < 0.01$, $r = 0.977$) and negative correlation between liver of fish infected with *Pomphorhynchus* and water ($p < 0.05$, $r = -0.937$). Zn concentration in liver of fishes infected with *Pomphorhynchus* was negatively correlated ($p < 0.05$, $r = -0.890$) with water. Al concentration had positive correlation between muscle of fishes infected with *Pomphorhynchus* ($p < 0.05$, $r = 0.939$) and water levels.

Water temperature depicted positive correlation with Mn concentration ($p < 0.01$, $r = 0.972$) in water. Free carbon dioxide was found positively correlated with Zn concentration ($p < 0.05$, $r = 0.885$) in water.

Zn concentration found in liver infected with *Bothriocephalus* had positive correlation with pH ($p < 0.05$, $r = 0.952$). Ni concentration in *Pomphorhynchus* depicted significant positive correlation with free carbon dioxide ($p < 0.05$, $r = 0.889$). Mn concentration in Liver of fish, infected with *Bothriocephalus*, depicted significant positive correlation with dissolved oxygen ($p < 0.05$, $r = 0.883$) and pH ($p < 0.01$, $r = 0.997$) of water. Liver of fish, infected with *Pomphorhynchus*, was positively correlated with temperature ($p < 0.01$, $r = 0.968$) and dissolved oxygen ($p < 0.05$, $r = 0.924$). Cr concentration in Muscle tissue of fish, infected with *Bothriocephalus*, was positively correlated ($p < 0.05$, $r = 0.938$) with water temperature. Fe concentration in Liver of fishes infected with

Pomphorhynchus showed strong positive correlation with temperature ($r= 0.980$), dissolved oxygen ($r= 0.967$) and pH ($r= 0.904$). Ca concentration in *Adenoscolex* was positively correlated with pH ($p<0.05$, $r=-0.939$). Cd concentration in Muscle tissue of fishes infected with *Pomphorhynchus* depicted positive correlation with dissolved oxygen ($p<0.05$, $r= 0.902$).

4.6.2 Correlation among metal concentrations of water, fish tissues, and parasites, and physicochemical parameters of River Jhelum

During summer Cu concentration in muscle of fishes infected with *Bothriocephalus* showed positive correlation ($p<0.05$, $r= 0.933$) with water. Zn concentration of *Adenoscolex* was found negatively correlated ($p<0.05$, $r=-0.992$) with water. Mn concentration in liver of fishes infected with *Pomphorhynchus* showed positive correlation ($p<0.05$, $r= 0.939$) with water. Cr concentration in muscle of fishes infected with *Adenoscolex* showed positive correlation ($p<0.05$, $r= 0.957$) with water. Al concentration in liver of fishes infected with *Adenoscolex* was found negatively correlated ($p<0.05$, $r=-0.906$) with water levels.

Water temperature was found positively correlated with Mn concentration ($p<0.05$, $r= 0.940$) and Cr concentration ($p<0.01$, $r= 0.982$) of water. Dissolved O₂ showed positive correlation with Zn concentration ($p<0.05$, $r= 0.924$) of water. Free CO₂ showed positive correlation Cu concentration ($p<0.05$, $r= 0.939$), Mn concentration ($p<0.05$, $r= 0.919$) and Al concentration ($p<0.01$, $r= 0.960$) of water.

Cu concentration in *Adenoscolex* was found positively correlated with dissolved oxygen ($p<0.01$, $r= 0.970$). Co concentration in tissue infected with *Adenoscolex* showed positive correlation with temperature ($p<0.01$, $r= 0.968$) and tissue infected *Pomphorhynchus* showed positive correlation with dissolved oxygen ($p<0.01$, $r= 0.966$). Mn concentration in Liver infected with *Pomphorhynchus* showed positive correlation with temperature ($p<0.05$, $r=0.902$). Cr concentration in tissue infected with *Adenoscolex* was found positively correlated with temperature ($p<0.05$, $r= 0.918$) and tissue infected

with *Bothriocephalus* showed positive correlation with free carbon dioxide ($p < 0.05, r = -0.911$). Fe concentration in *Bothriocephalus* depicted positive with dissolved oxygen ($p < 0.05, r = 0.907$). Cd concentration in *Adenoscolex* showed positive correlation with temperature ($p < 0.01, r = 0.951$).

During spring Zn concentration in the liver of fishes infected with *Pomphorhynchus* was negatively correlated ($p < 0.05, r = -0.957$) with water and muscle of fishes infected with *Bothriocephalus* was positively correlated ($p < 0.05, r = 0.924$) with water. Co concentration in the liver of fishes infected with *Bothriocephalus* depicted positive correlation ($p < 0.01, r = 0.968$) with water. Muscle of fishes infected with *Adenoscolex* ($p < 0.05, r = -0.983$) and *Bothriocephalus* ($p < 0.05, r = 0.935$) showed negative correlation with water. Ca concentration of muscles of fishes infected with *Bothriocephalus* was found positively correlated ($p < 0.05, r = 0.888$) and also Cd concentration of liver of fishes infected with *Pomphorhynchus* had positive correlation ($p < 0.01, r = 0.988$) with water levels.

Water temperature was found negatively correlated with Zn concentration ($p < 0.05, r = -0.946$) and showed positive correlation with Ni concentration ($p < 0.01, r = 0.963$) in water. Dissolved O_2 was negatively correlated with Cu concentration ($p < 0.05, r = -0.928$) and Al concentration ($p < 0.05, r = -0.907$) of water.

Cu concentration in *Adenoscolex* was found positively correlated with pH ($p < 0.05, r = 0.948$). Zn concentration in Liver infected with *Pomphorhynchus* showed positive correlation with temperature ($p < 0.01, r = 0.984$). Co concentration in Liver infected with *Pomphorhynchus* was negatively correlated with dissolved oxygen ($p < 0.05, r = -0.916$).

During autumn Ca concentration of liver of fishes infected with *Bothriocephalus* was negatively correlated ($p < 0.01, r = -0.974$) with water. Al concentration of *Pomphorhynchus* had positive correlation ($p < 0.05, r = 0.933$).

with water. The concentration in the liver of fishes, infected with *Pomphorhynchus*, was negatively correlated ($p < 0.01$, $r = -0.961$) with water. Mn concentration in muscle tissue of fishes infected with *Bothriocephalus* had positive correlation ($p < 0.01$, $r = 0.986$) with water. Cu concentration of *Pomphorhynchus* had negative correlation ($p < 0.01$, $r = -0.886$) with water.

Water temperature showed positive correlation with Zn concentration ($p < 0.01$, $r = 0.972$) and negative correlation with Mn concentration ($p < 0.01$, $r = -0.978$), Al concentration ($p < 0.05$, $r = -0.945$), Ca concentration ($p < 0.05$, $r = -0.942$). Dissolved O_2 depicted negative correlation with Zn concentration ($p < 0.05$, $r = -0.948$), Co concentration ($p < 0.05$, $r = -0.897$), Mn concentration ($p < 0.01$, $r = -0.971$), Al concentration ($p < 0.05$, $r = -0.945$) and Ca concentration ($p < 0.05$, $r = -0.885$).

Cu concentration in Liver infected with *Bothriocephalus* and *Adenoscolex* was found positively correlated with pH ($p < 0.01$, $r = 0.969$) ($p < 0.05$, $r = 0.929$), respectively. Co concentration in Liver infected with *Bothriocephalus* ($p < 0.05$, $r = 0.968$) showed positively significant correlation with pH ($p < 0.05$, $r = 0.919$). Ni concentration in Liver infected with *Bothriocephalus* showed negative correlation with temperature ($p < 0.05$, $r = -0.914$) and dissolved oxygen ($p < 0.05$, $r = -0.896$). Mn concentration in *Pomphorhynchus* was found positively correlated with pH ($p < 0.01$, $r = 0.923$). Cr concentration in *Bothriocephalus* was found positively correlated with pH ($p < 0.05$, $r = 0.880$). Al concentration in tissue infected with *Bothriocephalus* showed positive correlation with pH ($p < 0.05$, $r = 0.941$). Fe concentration in *Bothriocephalus* depicted positive correlation with dissolved oxygen ($p < 0.05$, $r = 0.950$).

During winter Al concentration in liver of fishes infected with *Bothriocephalus* was positively correlated ($p < 0.05$, $r = 0.936$) with water. Cd concentration in muscle tissue of fish infected with *Pomphorhynchus* was positively correlated ($p < 0.05$, $r = 0.898$) and Cr concentration depicted positive correlation of *Pomphorhynchus* ($p < 0.05$, $r = 0.908$) with water.

Water temperature was found positively correlated with Mn concentration ($p < 0.05$, $r = 0.918$) and negatively with Ca concentration ($p < 0.05$, $r = -0.921$). Free CO₂ had been positively correlated with Co concentration ($p < 0.05$, $r = 0.886$).

Cu concentration in Liver infected with *Bothriocephalus* was positively correlated with dissolved oxygen ($p < 0.01$, $r = 0.982$), Liver infected with *Pomphorhynchus* showed positive correlation with pH ($p < 0.05$, $r = 0.894$) and *Pomphorhynchus* was found positively correlated with pH ($p < 0.05$, $r = 0.919$). Zn concentration in Liver infected with *Adenoscolex* was positively correlated with dissolved oxygen ($p < 0.01$, $r = 0.961$). Co concentration tissue infected with *Bothriocephalus* showed positive correlation with pH ($p < 0.05$, $r = -0.924$). Ni concentration in tissue infected with *Pomphorhynchus* was found positively correlated with free carbon dioxide ($p < 0.05$, $r = 0.978$). Mn concentration in tissue infected with *Pomphorhynchus* was found positively correlated with temperature ($p < 0.01$, $r = 0.960$).

4.7 Histopathology of parasitic infections of *Schizothorax niger*

4.7.1 *Pomphorhynchus kashmirensis* infection:

Clinically the fishes showed grayish or yellowish discoloration with various degrees of emaciation (Fig. 38). Viscera looked reddish pink on opening the abdomen (Fig. 39). The intestines contained excessive mucous production and the acanthocephalan parasites had pierced the intestinal wall and were firmly attached (Fig. 40). Numerous acanthocephalan parasites were recovered from the intestine (Fig. 41). Liver was friable and comparatively pale in colour. The intensity of histopathological lesions varied in various seasons with the level of parasitic infections.

4.7.1.1 Spring

Intestine: Both lamina epithelia and lamina propria showed infiltration of inflammatory cells (Fig 42). The proboscis of the *Pomphorhynchus* was

embedded in the intestinal mucosa surrounded by the thickened connective tissue capsule infiltrated with inflammatory cells (Fig 43). Goblet cells hyperplasia was seen in lamina epithelia while eosinophilic granule cells were seen in lamina propria. Desquamation of epithelial cells was seen. Goblet cells revealed positivity of acid mucopolysaccharides (Fig 44).

Liver: Revealed cellular swelling, vacuolar degeneration and nuclear pyknosis (Fig. 45).

4.7.1.2 Summer

Intestine: Severe enteritis was seen characterized by infiltration of inflammatory cells in lamina propria and desquamated lamina epithelialis (Fig 46). The inflammation had extended the muscular tunic of the intestine (Fig 47). Sometimes the desquamation was so acute that the villi were completely denuded. Acid mucopolysaccharide was seen in the disrupted goblet cells (Fig 48).

Liver: Cells revealed swelling, vacuolar degeneration and kupffer cell hyperplasia (Fig. 49).

4.7.1.3 Autumn

Intestine: Severe enteritis was evident characterized by filtration of inflammatory cells in lamina propria and severe desquamation of lamina epithelium (Fig 50). Eosinophilic granulocytes were seen in lamina propria (Fig 51). Blood capillaries revealed congestion. Goblet cells revealed hyperplasia with presence of acid mucopolysaccharide (Fig 52).

Liver: It continued to reveal cellular swelling vacuolar degeneration and congestion (Fig. 53).

4.7.1.4 Winter

Intestine: *Pomphorhynchus* infection mostly coexisted with *Adenoscolex* infection during winters (Fig 54). The lamina epithelium was severely desquamated and there were heavy infiltration of inflammatory cells. Villi were sometimes

Pomphorhynchus kashmirensis infection in *Schizothorax niger*



Fig. 38: *S. niger* revealing gray and yellow discoloration of skin.



Fig 39: *S. niger* revealing reddish pink discoloration of abdominal viscera.

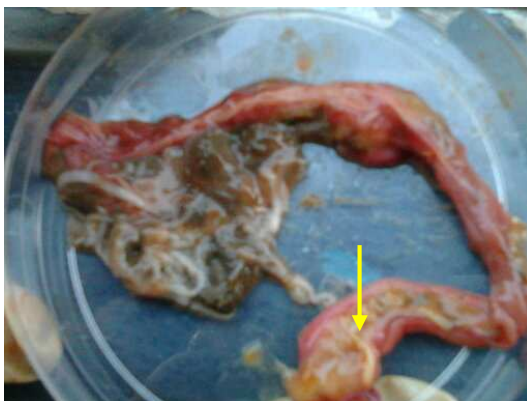


Fig. 40: Intestinal contents showing excessive mucous production. *Pomphorynchus* had penetrated the wall of intestine which was reddish in colour

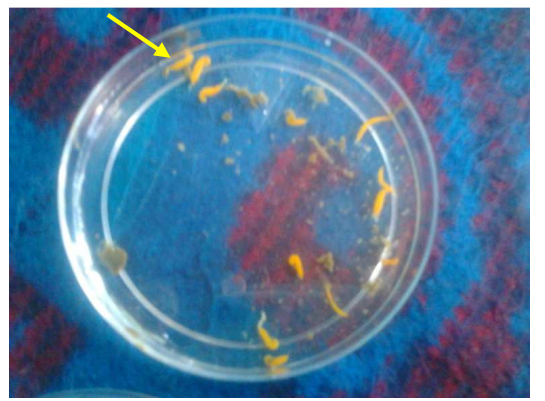


Fig. 41: *Pomphorynchus* recovered from the intestine of *S. niger*.

Spring season

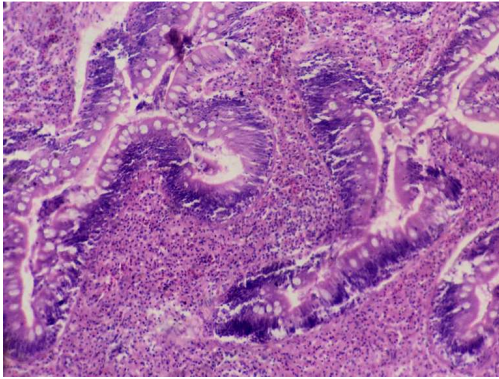


Fig 42. Both lamina epithelialis and lamina propria showed infiltration of inflammatory cells. **H & E X 35.**

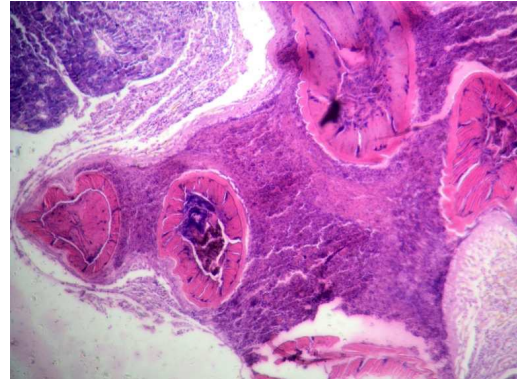


Fig 43. Section of presoma inserted into the intestinal was surrounded by connective tissue capsule. **H & E X 18.**

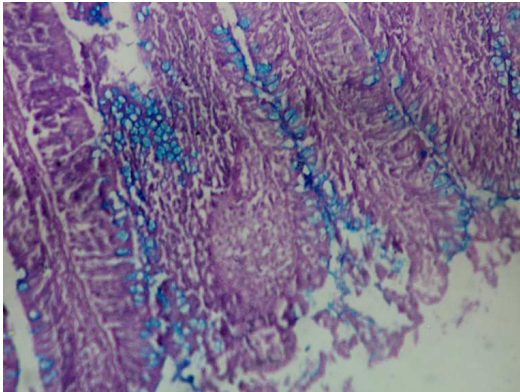


Fig 44. Goblet cell hyperplasia with positivity of acid mucopolysaccharides. **Alcian blue PAS staining X 33.**

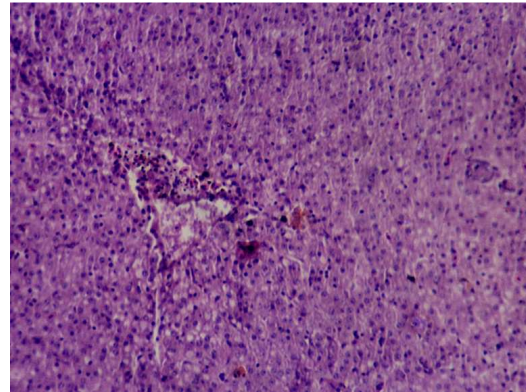


Fig 45. Liver showed cellular swelling, vacuolar degeneration and nuclear pyknosis. **H & E X 61.**

Summer season

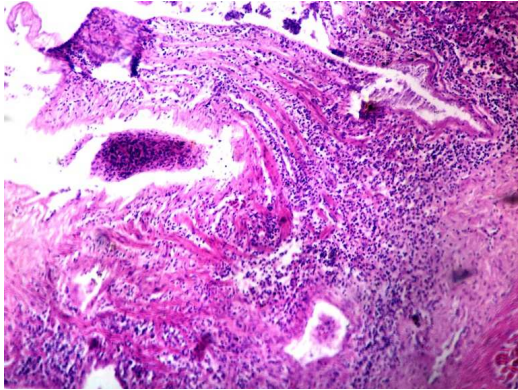


Fig 46. Severe enteritis characterized by infiltration of inflammatory cells in lamina propria and desquamated lamina epithelialis. **H & E X 30.**

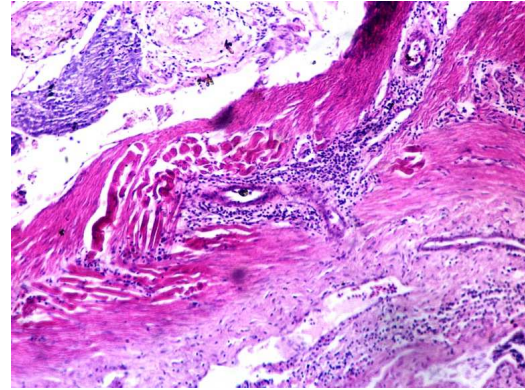


Fig 47. The inflammation had extended the muscular tunic of the intestine. **H & E X 35.**

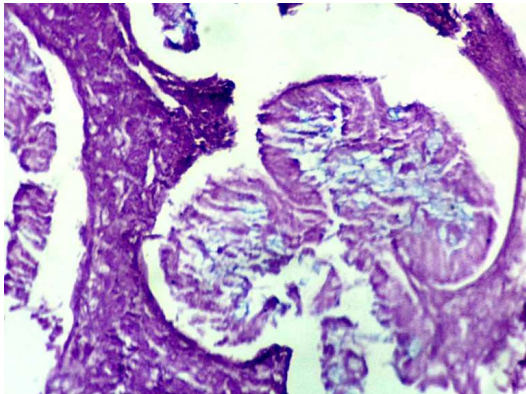


Fig 48. Acid mucopolysaccharide was seen in the disrupted goblet cells. **Alcian blue PAS staining X 65.**

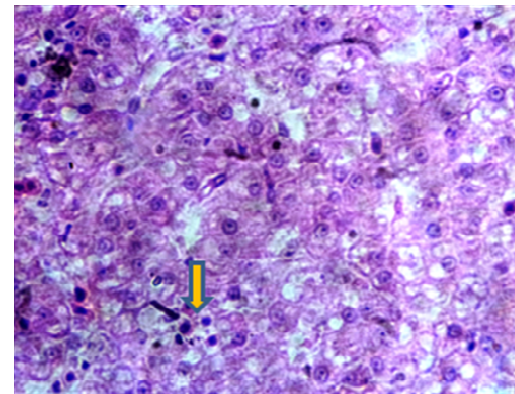


Fig 49. Liver cells revealed swelling, vacuolar degeneration and kupffer cell hyperplasia (arrow). **H & E X 160.**

Autumn season

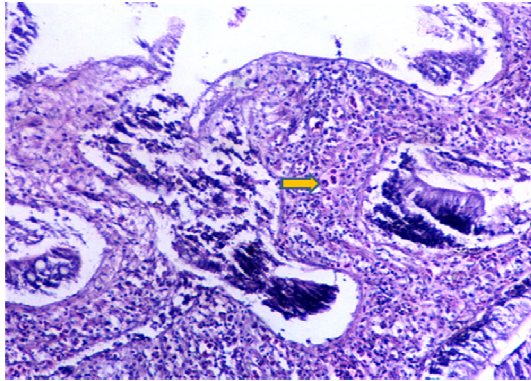


Fig 50. Intestine: Severe enteritis and severe desquamation of lamina epithelialis. Eosinophilic granulocytes were seen in lamina propria(arrow). **H & E X 56.**

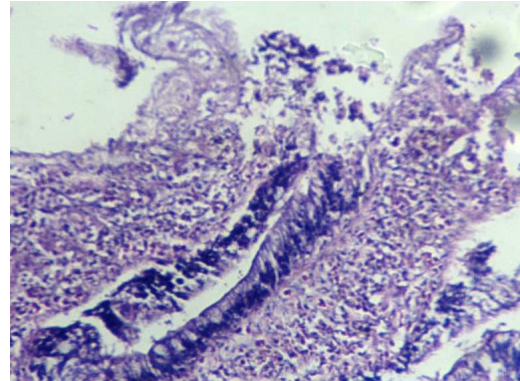


Fig 51. Intestine: severe desquamation of lamina epithelialis. **H & E X 56.**

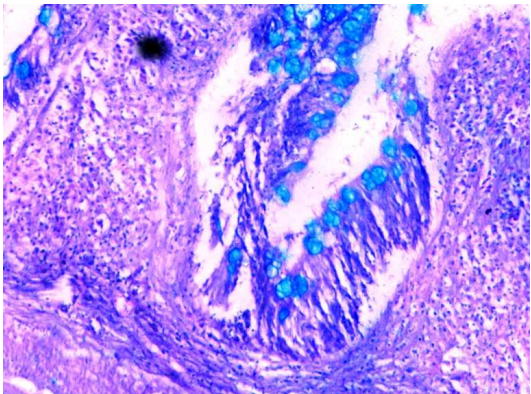


Fig 52. Goblet cells revealed hyperplasia with presence of acid mucopolysaccharide. **Alcian blue PAS staining X 35.**

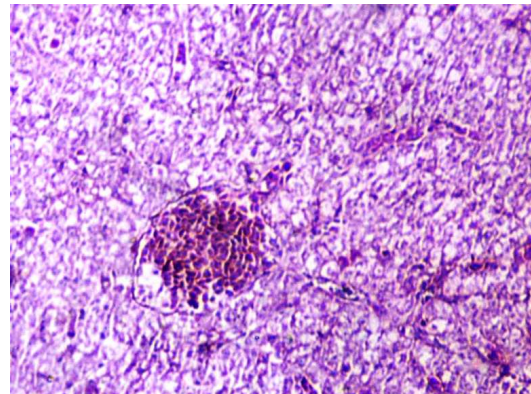


Fig 53. Liver: It continued to reveal cellular swelling vacuolar degeneration and congestion. **H & E X 75.**

Winter season

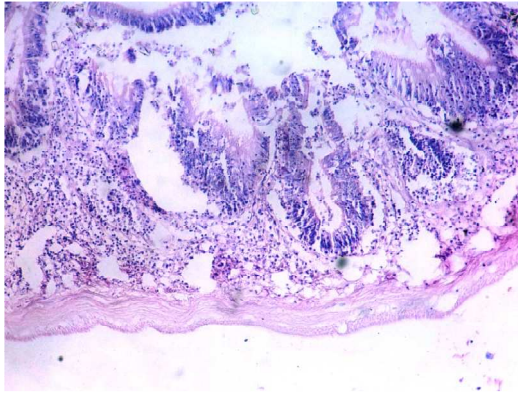


Fig 54. Enteritis characterized by heavy infiltration of inflammatory cells in lamina propria and severely desquamated villar epithelium. **H & E X 33**

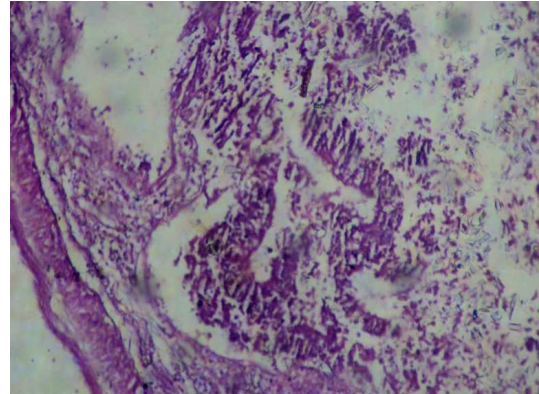


Fig 55. Villi were sometimes completely denuded of epithelial cells. **H & E X 30.**

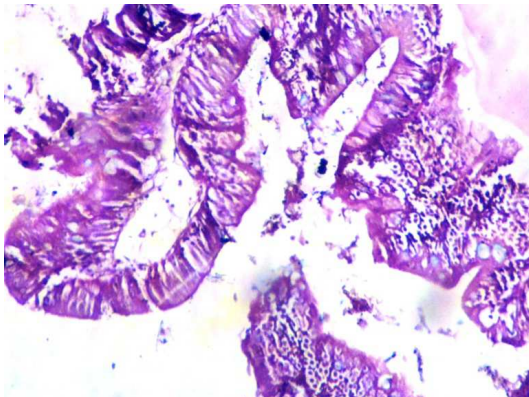


Fig 56. Goblet cells hyperplasia with evidence of acid mucopolysaccharide was in surviving epithelial cells. **Alcian blue PAS staining X 30**

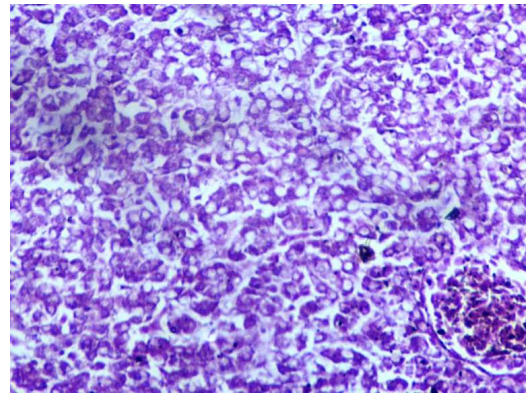


Fig 57. Liver: It revealed vacuolar changes and disruption of hepatocytes. **H & E X 76**

completely denuded of epithelial cells (Fig. 55). Goblet cells hyperplasia with evidence of acid mucopolysaccharide was in surviving epithelial cells (Fig. 56).

Liver: It revealed vacuolar changes (Fig. 57).

4.7.2 *Bothriocephalus acheilognathi* infection

The affected fish were anemic and emaciated (Fig 58). The viscera were dark red on opening the abdomen (Fig 59). The intestinal contents showed lot of mucous and contained dark contents (Fig 60). The mucosal wall was red in colour and revealed necrotic surface (Fig 61). The histopathological changes were varied in severity with the season and parasitic burden.

4.7.2.1 Spring

Intestine: Infection of *Bothriocephalus* with *Adenoscolex* was usually seen together. Severe enteritis with heavy infiltration of inflammatory cells and fibroblasts seen in lamina propria (Fig 62). Lamina epithelialis showed severe desquamation (Fig 63). Necrotic intestinal villi were evident. Goblet hyperplasia was seen alongwith elucidation of acid mucopolysaccharide (Fig 64).

Liver: cells very much swollen, rarefied and vacuolated (Fig 65).

4.7.2.2 Summer

Intestine: compared to spring infection, the enteritis changes with less severe. The lamina propria was infiltrated with inflammatory cells like granulocytes and lymphocytes (Fig. 66). Lamina epithelia were desquamated. Villi were very much thickened towards bases due to cellular infiltration. Epithelial cells revealed acid mucopolysaccharide (Fig. 67). Mast cell infiltration was evident with scattered metachromatic granules evident about the lesion (Fig. 68).

Liver: cells were swollen, vacuolated and rarefied (Fig 69).

4.7.2.3 Autumn

Intestine: Severe enteritis was seen with infiltration of mononuclear cells and

Bothriocephalus acheilognathi infection in *Schizothorax niger*



Fig. 58. The affected fish were anemic and emaciated.



Fig. 59. The viscera were dark red on opening the abdomen.



Fig. 60. The intestinal contents showed lot of mucous and contained dark contents. *Bothriocephalus* seen in contents (arrow)



Fig. 61. The mucosal wall was red in colour and revealed necrotic surface. *Bothriocephalus* seen in contents (arrow)

Spring season

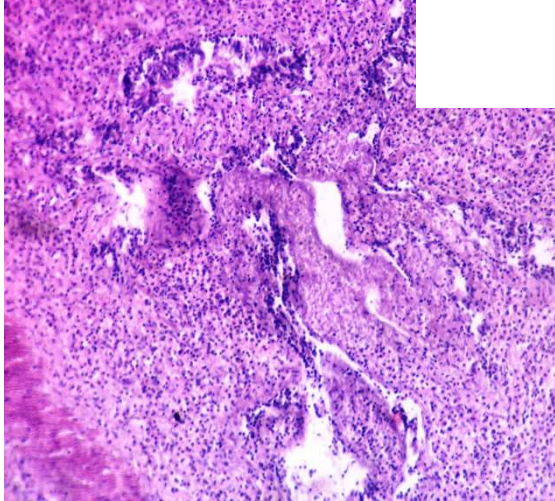


Fig. 62. Section of intestine infected with *Bothriocephalus* showing severe enteritis with heavy infiltration of inflammatory cells and fibroblasts seen in lamina propria. **H & E X 35**

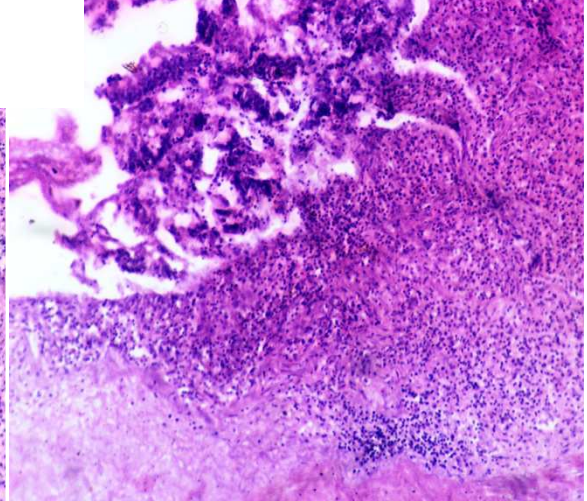


Fig. 63. Lamina epithelialis showed severe desquamation. **H & E X 35**

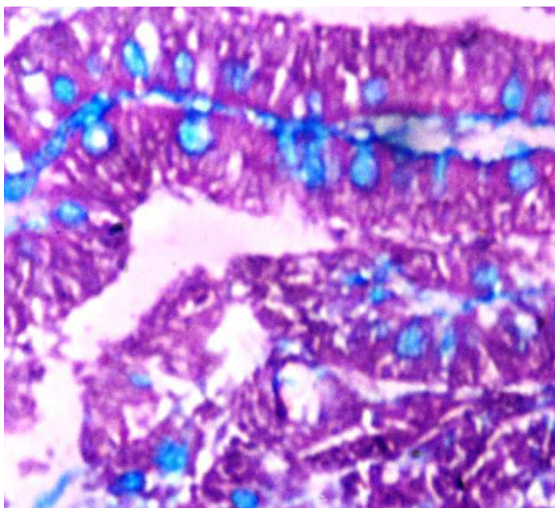


Fig. 64. Goblet hyperplasia was seen along with elucidation of acid mucopolysaccharide. **Alcian Blue PAS staining X 80.**

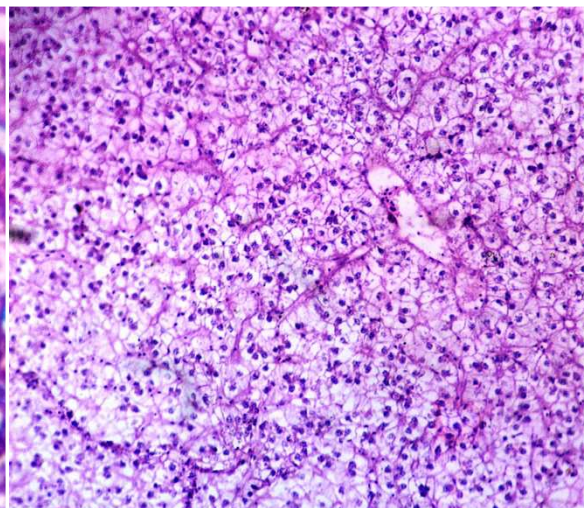


Fig. 65. Liver: cells very much swollen, rarefied and vacuolated. Nuclear pyknosis was evident. **H & E X 11.2.**

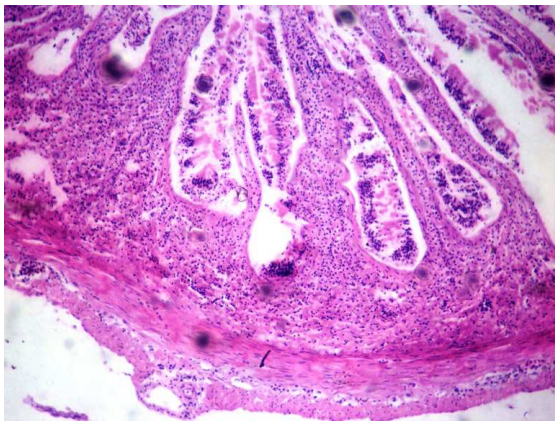


Fig. 66.Intestine: compared to spring infection, the enteritis changes with less severe. The lamina propria was infiltrated with inflammatory cells like granulocytes and lymphocytes. **H & E X28.**

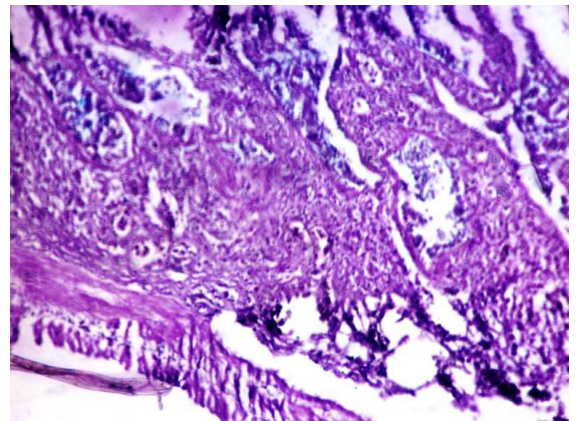


Fig. 67.Necrotic epithelial cells revealed acid mucopolysaccharide. **Alcian blue PAS stain X 41.**

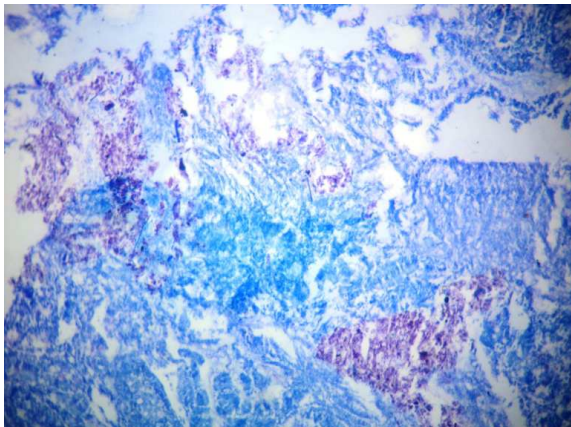


Fig. 68. Mast cell infiltration was evident with scattered metachromatic granules. **Toluidine blue stain X 25.**

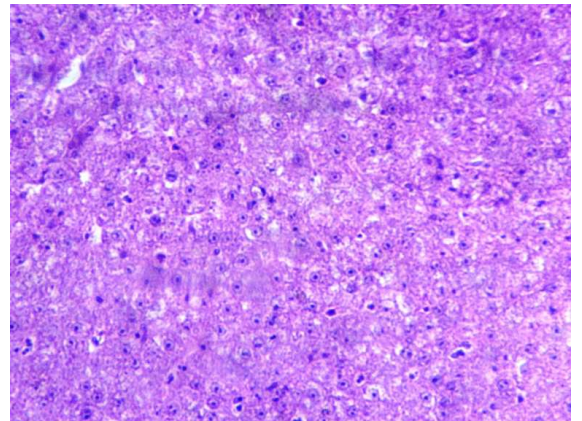


Fig. 69.Liver: cells were swollen, vacuolated and rarefied. **H & E X 76.**

on

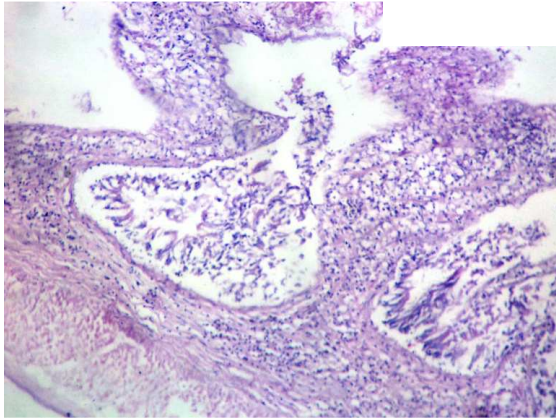


Fig. 70. Severe enteritis was seen with infiltration of mononuclear cells and desquamated necrotic cells. **H & E X 35.**

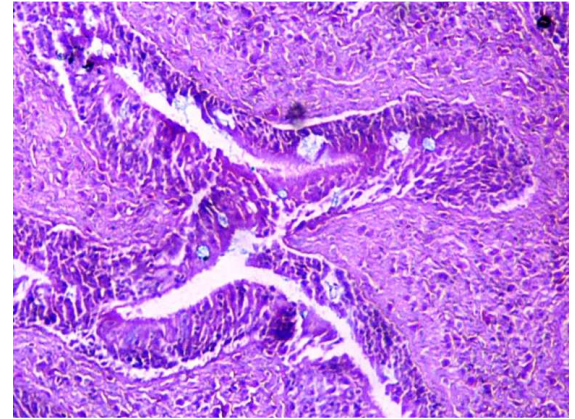


Fig. 71. Lamina epithelialis showed cellular as well as goblet cell hyperplasia with evidence of acid mucopolysaccharide. **Alcian Blue PAS staining X 70.**

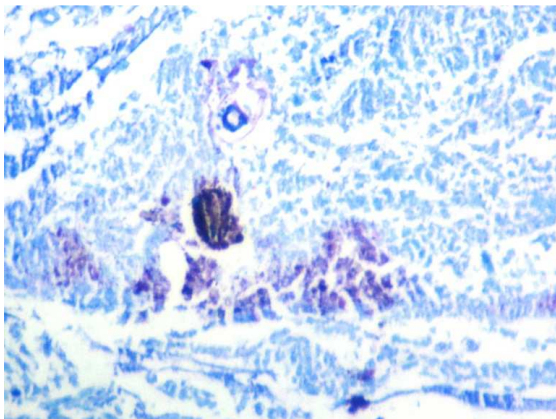


Fig. 72. Mast cells are seen in the lamina propria of intestinal wall. **Toluidine blue stain X 76.**

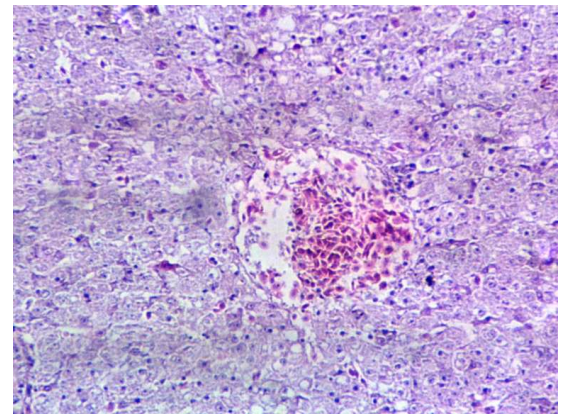


Fig. 73. Liver: Cells showed severe degenerative changes. **H & E X 69.**

Winter season

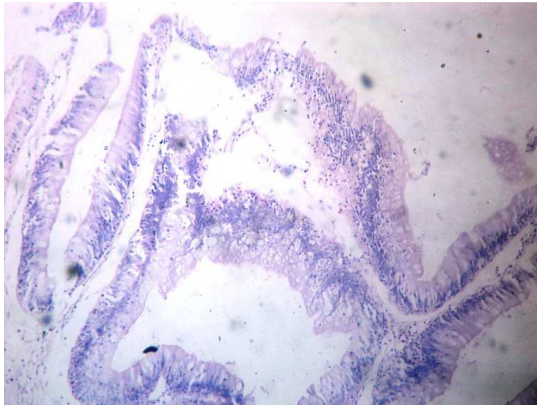


Fig. 74. Intestine: Enteritis was less severe with infiltration of cells in lamina propria and lamina epithelium **H &E X 28.**

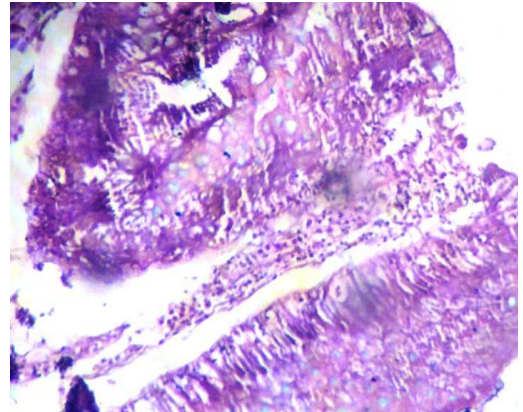


Fig. 75. Goblet cells hyperplasia with presence of acid mucopolysaccharide was seen **Alcian blue PAS stain X 61.**

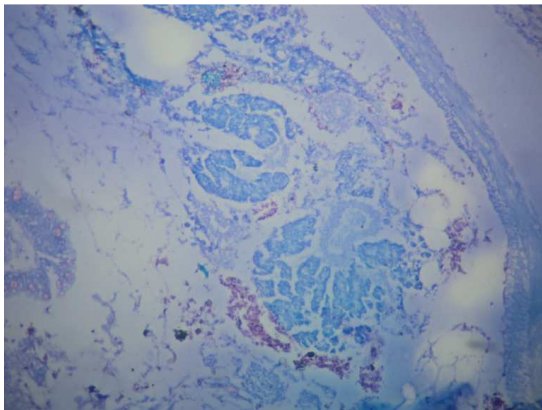


Fig. 76. Mast cells were seen in lamina propria. **Toluidine blue stain X 26.**

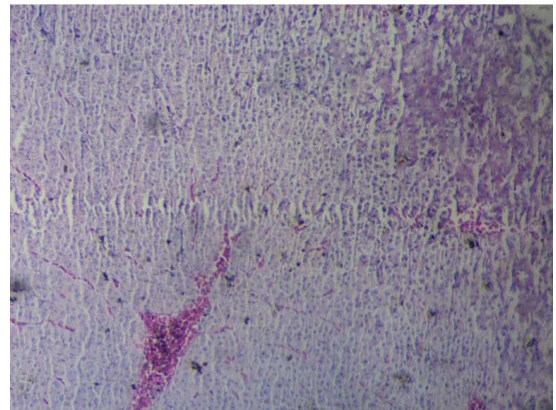


Fig. 77. Liver: Cells revealed degenerative changes and congestion. **H &E X 57.**

desquamated necrotic cells (Fig. 70). The villi were very much thickened and necrotic. Lamina epithelialis showed cellular as well as goblet cell hyperplasia with evidence of acid mucopolysaccharide (Fig. 71). Mast cells were evident in the intestinal wall (Fig. 72).

Liver: Cells showed severe degenerative changes (Fig. 73).

4.7.2.4 Winter

Intestine: Enteritis was less severe with infiltration of cells in lamina propria and lamina epithelium (Fig. 74). Enterocysts showed degenerative changes. Goblet cells hyperplasia with presence of acid mucopolysaccharide was seen (Fig 75). Mast cells were seen in lamina propria (Fig. 76).

Liver: Cells revealed degenerative changes and congestion (Fig. 77).

4.7.3 *Adenoscolex oreini* infection

The fishes infected with *Adenoscolex oreini* were anemic and the abdomen appeared slightly pot bellied (Fig. 78). Viscera appeared red on opening the abdomen and the abdominal fluid was tinged red (Fig. 79). On opening the intestine necrotic debris was present on the surface and numerous parasites were present (Figs. 80 & 81).

4.7.3.1 Spring

Intestine: Enteritis characterized by inflammatory cells in the lamina propria and lamina epithelialis was seen (Fig. 82). Epithelial desquamation was evident and hyperplasia of lymphoid nodules seen. Villi towards the luminal side were completely devoid of epithelial cells and were only survived by connective core. Eosinophils granule cells are seen in lamina propria. Goblet cells hyperplasia was seen with positivity for acid mucopolysaccharide (Fig. 83).

Liver: Cells were swollen showing vacuolar degeneration and Kupffer cell hyperplasia (Figs. 84 & 85).

4.7.3.2 Summer

Intestine: Severe chronic enteritis was seen with infiltration of lymphocytes and fibroblasts in the lamina propria (Fig. 86). The necrotic villi were completely denuded of epithelial mucosa (Fig. 87). Only the goblet cells in surviving epithelia revealed hyperplasia changes with evidence of acid mucopolysaccharide. Mast cells were occasionally seen.

Liver: Cells showed moderate degenerative changes with cellular swelling and distortion (Figs. 88 & 89).

4.7.3.3 Autumn

Intestine: Enteritis was comparatively less severe. *Lamina propria* was infiltrated with mononuclear and eosinophilic granule cells (Fig 90). Necrosis of some villi was seen represented by fibrillar networks (Fig 91). Alcian blue PAS staining revealed goblet cells hyperplasia with presence of acid mucopolysaccharide (Fig 92). Mast cells were occasionally seen in lamina propria.

Liver: cells revealed degenerative changes; cellular swelling and pyknotic nuclei were occasionally seen (Fig. 93).

4.7.3.4 Winter

Intestine: Enteritis was characterized by infiltration of inflammatory cells in lamina propria and lamina epithelial along with necrosis of mucosal epithelial cells (Fig 94). Intestinal villi had become thickened and crypts were obliterated. Eosinophiles granules were seen in lamina propria (Fig. 95). Goblet cell hyperplasia was clearly seen having acid mucopolysaccharide (Fig. 96). Mast cells were evident.

Liver: cells revealed vascular degeneration and cellular disorganization (Fig. 97).

Adenoscolex oreini infection in *Schizothorax niger*



Fig. 78. The fishes infected with *Adenoscolex oreini* were anemic and the abdomen appeared slightly pot bellied.



Fig. 79. Viscera appeared red on opening the abdomen and the abdominal fluid was tinged red.



Fig. 80. Intestinal mucosa showed necrotic debris and numerous parasites were present.



Fig. 81. Numerous *Adenoscolex oreini* were recovered from the intestines.

Spring Season

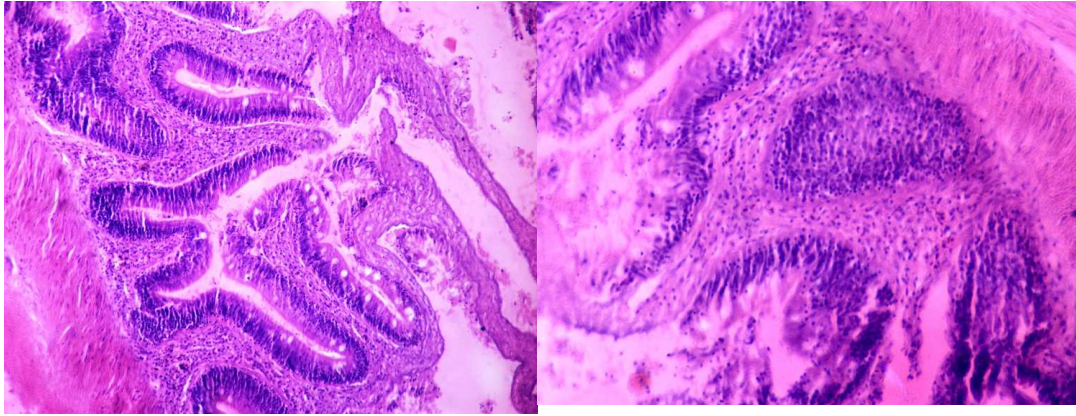


Fig. 82. Intestine of fish infected with *Adenoscolex oreini* revealing enteritis. Infiltration of inflammatory cells in lamina propria and lamina epithelia were seen. **H & E X 25.**

Fig. 83. Epithelial desquamation was evident and hyperplasia of lymphoid nodules seen. **H & E X 65.**

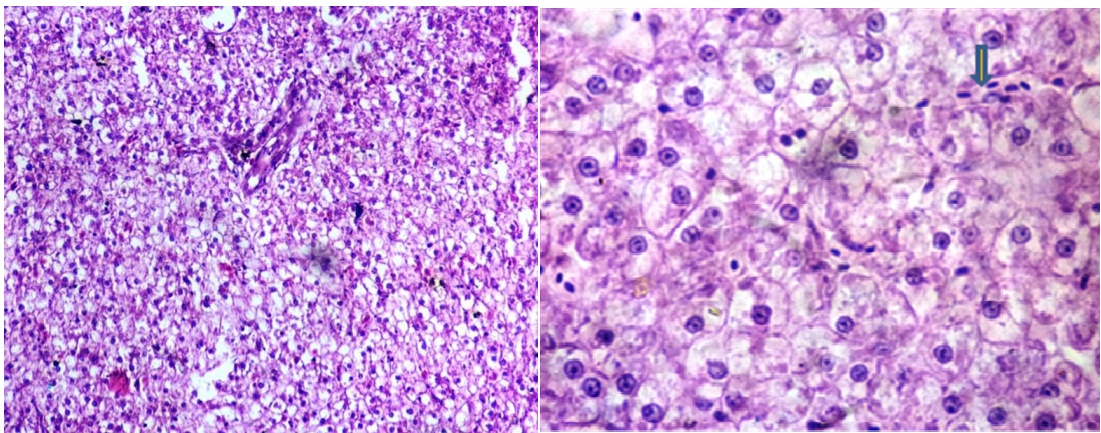


Fig. 84. Cells were swollen showing vacuolar degeneration. **H & E X 57.**

Fig. 85. Higher magnification of the same showing Kupffer cell hyperplasia (arrow). **H & E X 160.**

Summer Season

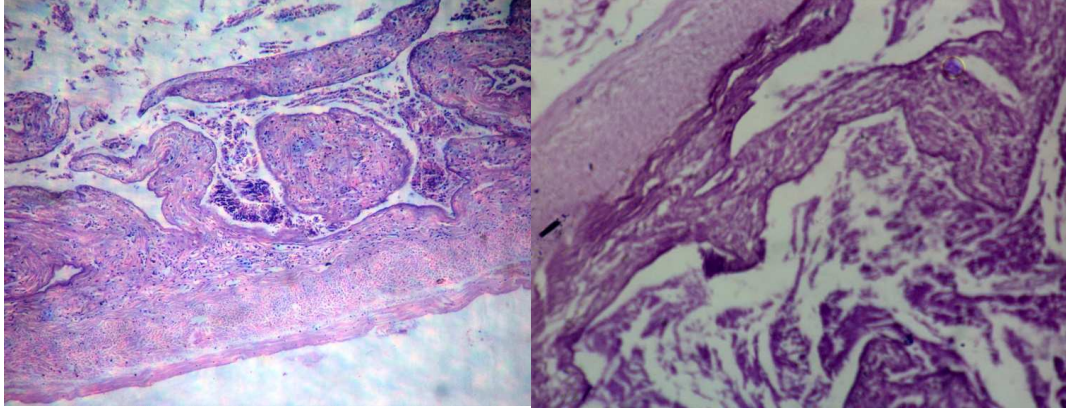


Fig. 86. Severe chronic enteritis was seen with infiltration of lymphocytes and fibroblasts in the lamina propria. **H & E X 28.**

Fig. 87. The necrotic villi were completely denuded of epithelial cells. **Alcian blue PAS X 40.**

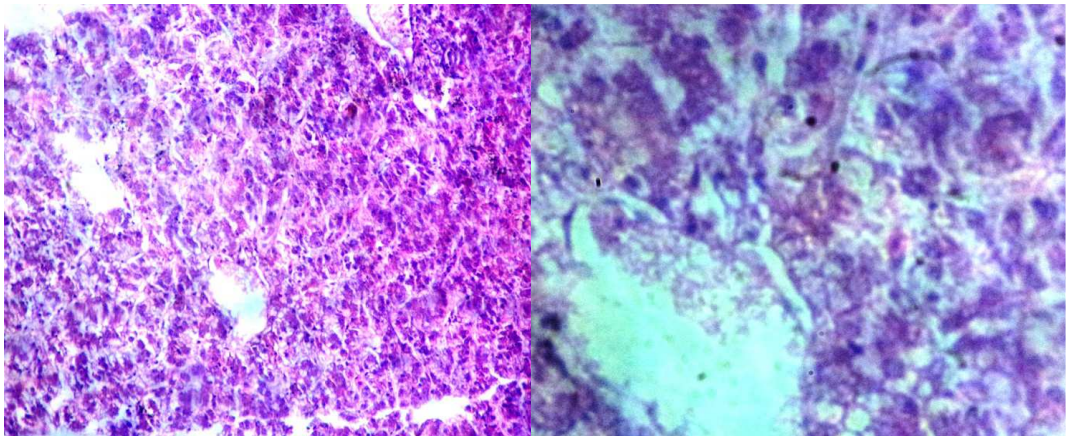


Fig. 88. Cells showed moderate degenerative changes with cellular swelling and distortion. **H & E X 35.**

Fig. 89. Higher magnification of the same. **H & E X 140.**

Autumn Season

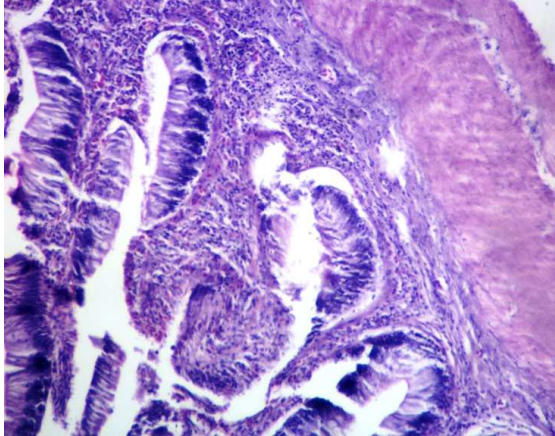


Fig. 90. Enteritis was comparatively less severe. *Lamina propria* was infiltrated with mononuclear and eosinophilic granule cells. **H & E X 41.**

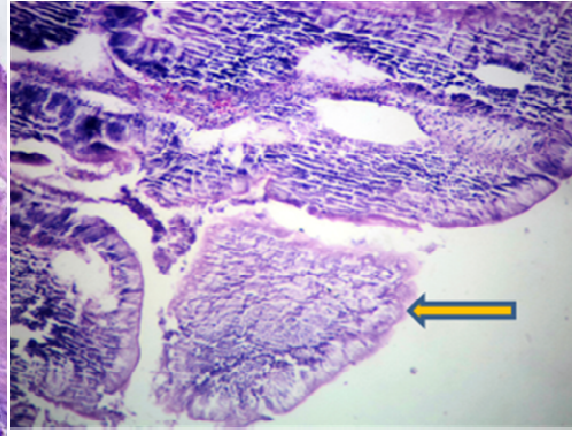


Fig. 91. Necrosis of some villi was seen represented by fibrillar networks (arrow). **H & E X 30.**

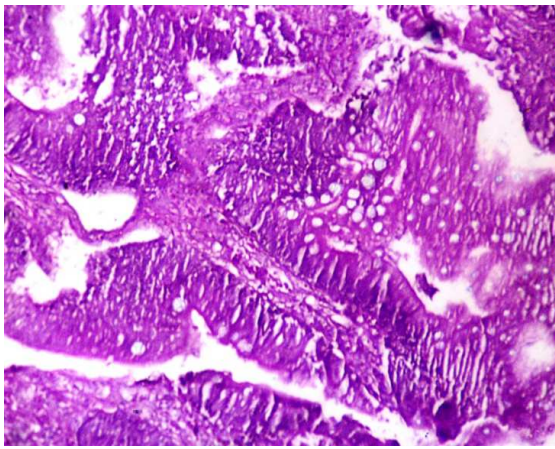


Fig. 92. Goblet cell hyperplasia with presence of acid mucopolysaccharide. **Alcian blue PAS staining 35.**

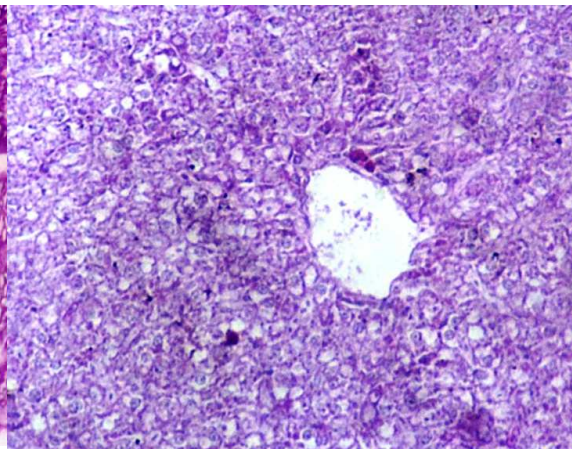


Fig. 93. Hepatic cells revealed degenerative changes with vacuolar degeneration. **H & E X 80.**

Winter season

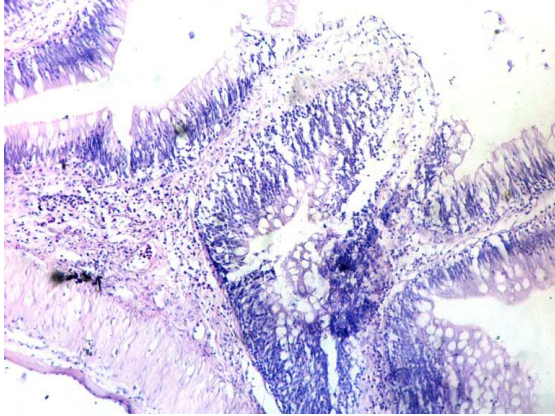


Fig. 94.Enteritis characterized by infiltration of inflammatory cells in lamina propria and lamina epithelial along with necrosis of mucosal epithelial cells. **H & E X 33.**

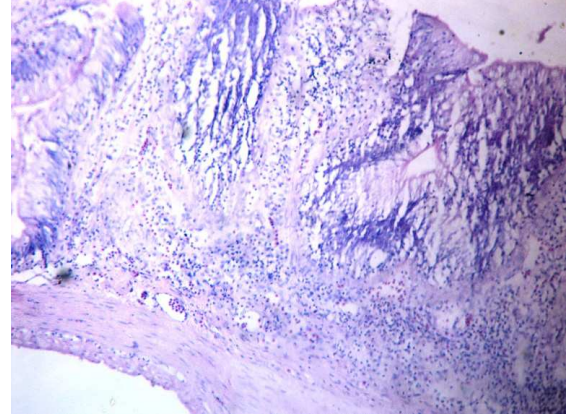


Fig. 95.Intestinal villi had become thickened and crypts were obliterated. **H & E X 33.**

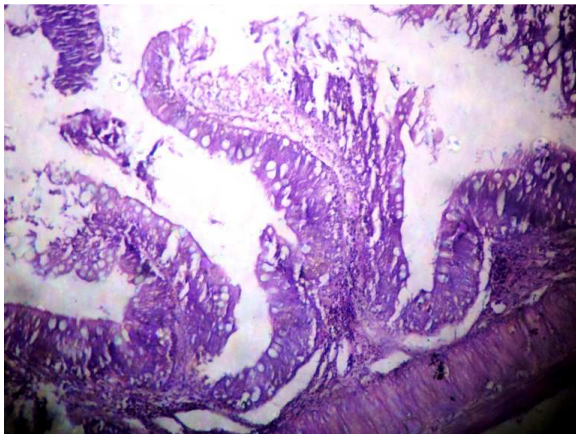


Fig. 96. Goblet cell hyperplasia was clearly seen having acid mucopolysaccharide. **Alcian blue PAS staining X 28**

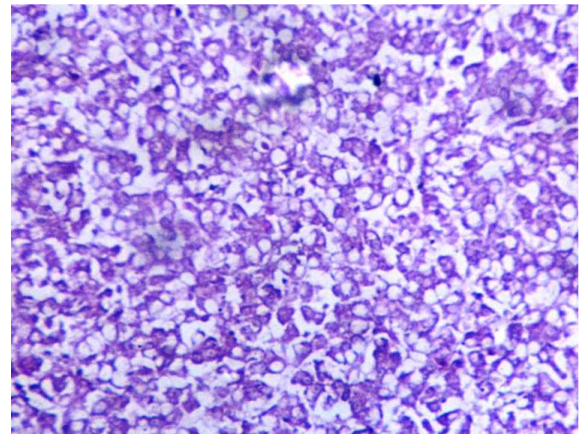


Fig. 97.Liver: cells revealed vascular degeneration and cellular disorganization. **H & E X 80.**

Chapter 5

DISCUSSION

5.1 Physicochemical parameters

All the four physicochemical parameters temperature, dissolved oxygen, pH and free carbon dioxide showed variation vis-a vis the season and location of the stations in water bodies. Generally, the weather in study area was quite cool; however the water temperature plays an important role which influences the chemical, bio-chemical characteristics of water body. Highest temperature in Dal Lake was seen at Dalgate during summer ($27.22^{\circ}\text{C}\pm 1.71$) while the lowest temperature was also recorded at the same site during winter ($6.55\pm 1.48^{\circ}\text{C}$). Similarly, the highest temperature (19.44 ± 1.50) in Jhelum River was recorded at Chattabal weir while lowest (4.38 ± 1.32) was recorded at Khannabal indicating that the water near the source of origin, i.e. Verinag, is at lower degree than down the river flow. Besides, the temperature of water in Jhelum River is at lower degree than that of Dal Lake which might be attributed to flowing nature of water in Jhelum. Water temperature in summer was high due to low water level, high atmospheric temperature and clear atmosphere (Salve and Hiware, 2008). At all the study sites minimum air temperature was recorded in winter, with the lowest recorded temperature as 4°C at Chattabal site, while as the maximum in the summer season, with the highest 27°C at Dalgate site. Overall the temperature was high in summer and low in winter. The results showed that the geographical location and season have a direct impact on the various physico-chemical properties of water. Air temperature at different sites followed the general climatic regime of the valley with minimum in January and maximum in July. Though the winter air temperature at times goes below freezing point, yet the water did not freeze for the reason the insulating function of snow and ice might be protecting the water below from freezing (Sheridan, 1961), also the temperature required to freeze running water is very low (Needham and Jones, 1959). Further, it has been suggested that the low temperature of river Jhelum could be attributed to the

presence of plantations at banks of river which shades the river surface thereby makes it less visible to direct sun light. The variation in water temperature depends mainly on the climatic conditions, sampling times, the number of sunshine hours and also is affected by specific characteristics of water environment such as turbidity, wind force, plant cover and humidity (Mahmoud, 2002 and Tayel, 2002).

In Dal Lake DO was highest at Telbal (6.78 ± 1.25 mg/l) during spring while lowest was at Hazratbal (3.55 ± 1.19 mg/l) during summer. In River Jhelum DO was highest at Khannabal (8.16 ± 1.43 mg/l) during spring while lowest was at Zero bridge (4.97 ± 1.50 mg/l) during autumn. The higher concentration of DO during spring season could be due to the fact that the melting snow and glacier ice flowing at a higher speed might be incorporating more oxygen. Cold water has been reported to contain more oxygen as compared to warm water as the DO is inversely proportional to the water temperature (Hynes, 1960). In addition photosynthesis could have some kind of effect on DO (Berg, 1943). DO of River Jhelum was noted at higher level in all the seasons compared to that in Dal Lake. The fluctuation in the DO value might be due to differences in water temperature (Kumar *et al.*, 1996).

The pH was found within the range of 7.03-8.4 with the highest recorded at Hazratbal site and lowest recorded in Chattabal weir. pH of the waters of Dal Lake and River Jhelum varied within the alkaline range. The pH of Jhelum river water was comparatively less alkaline than that of Dal Lake. In Jhelum the highest pH recorded at Khannabal was during spring (8.35 ± 0.51) and the lowest was at Chattabal weir during autumn (7.03 ± 0.28). In Dal Lake the highest pH recorded at Hazratbal was during summer (8.47 ± 0.41) and the lowest was at Dalgate during autumn (7.45 ± 0.35). The desirable limit for pH is 6.0 to 8.0; however some sites crossed the desirable limit. The fluctuations might be due to low rates of decomposition and good amount of calcium carbonates and magnesium in the area. Moreover, due to greater photosynthetic activity greater utilization of CO₂ is

responsible for increased pH (alkaline). In Dal Lake the highest pH was noted at Hazratbal during summer (8.47 ± 0.41) and lowest was at Dalgate during autumn (7.45 ± 0.35). Most of bio-chemical and chemical reactions are influenced by the pH. The reduced rate of photosynthetic activities reduces the assimilation of carbon dioxide and bicarbonates which are ultimately responsible for increase in pH, the low oxygen values coincided with high temperature during the summer month. The factors like temperature bring about changes the pH of water. The higher pH values observed suggested that carbon dioxide, carbonate-bicarbonate equilibrium is affected more due to change in physico-chemical condition (Karanth, 1987; Trivedi *et al.*, 2009).

Free carbon dioxide in Dal Lake was highest (12.88 ± 2.21 mg/l) at Telbal during spring while the lowest was found at Saidakadal (3.16 ± 0.93 mg/l) during autumn. In River Jhelum highest free CO₂ (8.11 ± 2.26 mg/l) was found at Chattabal weir during summer and lowest was also recorded (3.08 ± 1.83 mg/l) at the same site during autumn. Free carbon dioxide was found higher in Dal Lake than in River Jhelum in all the seasons of the year. This depends upon alkalinity and hardness of water body. The value of CO₂ was high in spring and summer. This could be related to the high rate of decomposition in the warmer months. The increasing trend of free carbon dioxide down the river could be due to the addition of some carbon rich substances as majority of carbon comes from organic matter such as ground water, rock leaching, dead terrestrial plant material (Wetzel, 1992). The reason for increasing trend of calcium down the river could be increased concentration of waste material especially the calcium rich substances like bones and milk products of slaughtered and killed animals, which are usually irresponsibly disposed off in the water bodies by the public.

5.2 Prevalence of helminth parasite

Three different species of *Schizothorax* were recovered from the two water bodies viz *S. niger*, *S. esocinus* and *S. curvifrons*. The recovery of *S. esocinus* was more from Dal Lake followed by *S. niger* and *S. curvifrons* while as

from Jhelum river recovery of *S. niger* was higher followed by *S. esocinus* and *S. curvifrons*. From all the three species of *Schizothorax* the helminth parasites recovered were Acanthocephalan parasite (*Pomphorhynchus kashmirensis*)(27.47%) and two intestinal cestodes (*Bothriocephalus acheilognathi*)(30.63%) and (*Adenoscolex oreini*)(32.43%). Earlier workers have also reported the presence of these helminths in the fishes (Amin, 1975a & b ; El-Naffar *et al.* 1983; Abu El-Ezz, 1988; Ebraheem 1992; El-Ganiny, 1995; Bayoumy, 1996; Abd El-Monem, 1998 and Thabit, 2004).

The present work showed that the infection patterns of *Pomphorhynchus* were greatly influenced by season, fish species and type of water body. It was seen that overall prevalence of *Pomphorhynchus* was low, compared to other two helminths, which was in accordance to the studies done by Spall and Summer (1969) and Chishti and Peerzada (1998) who showed 0.7% and 9.3% infection of the acanthocephalan parasite, respectively. Further, the low prevalence might be due to low availability or consumption of intermediate hosts. The present finding also paralleled with Sinderman (1990); Kuperman (1992) and Dusek *et al.* (1998) who attributed the decrease in the infection rate of fishes lived in highly polluted areas to the fact that, effluents including heavy metals could alter the availability or reduced the number of invertebrate intermediate hosts necessary for life cycle of these parasites. Seasonal variation in incidence of helminth parasitism in fishes was probably influenced by the annual life cycle of the parasites. However, the overall prevalence of 27.47%, observed in the present study, could be attributed to various factors like temperature and availability of food. The host species generally shows a minimum preference for animal food (Chishti and Peerzada, 1998) as they are mostly dependent on plankton (65-70%) which are the intermediate hosts for *P. kashmirensis*, the rest comprises of aquatic invertebrates. It is generally the amount of intake of intermediate host (which is an invertebrate) that determines the intensity of infection, so the present observation with lower prevalence of infection in *Pomphorhynchus kashmirensis*, compared to other two

helminths, in its host is a consequence of the comparatively low quantity of animal food in their diet. In spite of the low prevalence of the parasite still the prevalence rate of 27.47%, is higher than the reported values of Spall and Summer (1969) and Chishti and Peerzada (1998) thereby indicated that the intermediate hosts have increased over a period of time in the water bodies. In the present study the parasite revealed a wide host range. Amin (1987) also found a wide host range for *Pomphorhynchus bulbocoli* in Wisconsin fishes, which he attributed to similar feeding habits of the fish and also to the availability of intermediate host in the habitat.

In Dal Lake and River Jhelum *Pomphorhynchus kashmirensis* was more prevalent in *S. niger* and *S. curvifrons* during summer, respectively. There was a gradual increase in the prevalence rate from spring to summer and fell down with onset of autumn and was least observed during winter. Lowest prevalence of the parasite was during winter in both the water bodies. Overall the mean intensity of *Pomphorhynchus kashmirensis* was highest in Dal Lake whereas its abundance was higher in River Jhelum. Seasonal variation in incidence of helminth parasitism in fishes was probably influenced by the annual life cycle of the parasite. Further, the increased prevalence of the acanthocephalans in water bodies have been attributed to the contamination of water with municipal and industrial effluents (Billiard and Khan, 2003)

For both the cestodes mean intensity and abundance were higher in River Jhelum. *Bothriocephalus acheilognathi* prevalence was highest in *S. esocinus* and *S. niger* in Dal Lake and River Jhelum, respectively, during summer. *Adenoscolex oreini* was seen more in *S. niger* and *S. curvifrons* in Dal lake and River Jhelum during Summer and lowest during winter, respectively. The seasonal prevalence showed significant differences in prevalence. Clear seasonal trend was observed in Dal Lake and River Jhelum with maximum infection level during summer months and least in winter months. Significant differences ($p < 0.01$) in prevalence were recorded vis-à-vis the season in both the water bodies which were in

conformity with the results of Yufa and Tingbao (2011) who concluded that the helminth species like monogeneans showed seasonal alterations associated with environmental changes. The abrupt increase in helminth infection from summer in both water bodies could be due to increased duration of life of the infective larva, and has been reported to assist in the transfer of helminth infection like Diplozoon infection from fish to fish (Chubb, 1979). In the findings of Genc *et al.* (2005) the parasite infection showed seasonal variations with the highest prevalence in summer season, as suggested that decrease in water volume during the dry season caused nutritional imbalance resulting in less production of fish food organisms and moreover thereby reducing the immune response in fish and making them more vulnerable to disease agents. Boping and Wenbin (2007) studied the seasonal population of *Pallisentis (Neosentis) celatus* in the intestine of rice -field eel *Monopterus albus* and inferred that the prevalence of parasite undergoes a distinct seasonal trend being highest in spring and decreased corresponding to a fall in temperature. Further, no significant seasonal differences were found in mean intensities. Results of Singh and Sahay (2007) indicated that all parameters i.e., prevalence, intensity, density and index of infection showed highest values in June. These results clearly suggested that they were related to the rise of water temperature and the availability of secondary host. Wani and Magray (2008) reported highest prevalence of *Adenoscolex* during spring and winter seasons in river Jhelum and in winter in Anchar Lake. Further, the authors noted highest prevalence of *Bothriocephalus* during autumn season, followed by spring in both the water bodies. Interestingly, *Pomphorhynchus* was found throughout the year with peaks during spring and summer season in river Jhelum.

The mean intensity and abundance of both the cestodes were highest in summer and lowest in winter. Kanth and Srivastava (1987) also showed that *Pallisentis ophiocephali* had two peak periods during May and August and then the infestation rate declined gradually through September to February and rose through March to have peaks in May and August. Khan *et al.* (1991) studied

seasonal variation in the occurrence of *P. ophiocephali* and *Acanthosentis betwai* in relation to their fish hosts. *C. striatus* showed greater prevalence of *P. ophiocephali* infection between March and May and males were more heavily infected than females. *P. ophiocephali* collected from *C. punctatus* showed highest prevalence in the month of September, the intensity of infection being highest in August (Jha *et al.*, 1992). Malhotra and Banerjee (1997) observed peak infectivity in October of *P. allahabadi*. The above authors reported the initiation of infection in August when the higher peak of mean worm burden and relatively higher degree of infection were encountered. Higher prevalence of infection was retained until winter period and the peak infection incidence occurred during October. Thereafter, infection declined consistently during spring and early summer period before finally disappearing by the end of summer period. According to Kim *et al.* (2001) abundance of *P. gotoi* sharply increased to a peak in the late fall (November) then declined gradually to May. Vincent and Font (2003) observed that the prevalence, mean abundance and mean intensity of *Camallanus cotti* were higher in summer than in winter whereas the prevalence and mean abundance of *Bothriocephalus acheilognathi* showed no significant difference between the seasons. Khan *et al.* (2004) inferred that the prevalence of infection of *Diplozoon kashmirensis* and *Pomphorhynchus* in *Schizothorax* was highest during spring, while the minimum value was recorded in autumn whereas *Adenoscolex* showed a marked seasonal occurrence as the infection was observed only during spring and summer seasons. Significantly low mean intensity during winter in Dal Lake could be due to negative impact of contaminants on helminth itself and thus decreased intensity. It also seems that helminth show antagonistic response to combined effect of pollutant and eutrophication at this site.

Both the cestodes and the acanthocephalan infection were prevalent more in male fishes compared to females. This study is in full agreement with Machado *et al.* (2000). Reports on the prevalence of parasites with respect to host sex are diversified. Records on higher prevalence in male hosts are supported by the work

of Zelmer and Arai (1998) who observed that the slope of relationship between number and abundance of *Bothriocephalus* species and *Proteocephalus* species was greater for male perch than for females. Takemoto and Pavanelli (2000) reported that male hosts had significantly higher parasite intensity than females. The influence of sex on the susceptibility of animals to infections could be attributed to genetic predisposition and differential susceptibility owing to hormonal control.

The presence of the parasites had reduced the condition coefficient of the infected fishes in both the water bodies. However; the overall condition factor of fish in Jhelum river was higher. Condition coefficient was found to be lower in infected fish than in uninfected fish in both Dal Lake and River Jhelum. It might be due to the fact that parasites decrease the immune system of the hosts, which may lead to decreased growth of fish. Decreased growth may lead to decrease in condition coefficient (Khan and Thulin, 1991; Poulin, 1992). Parasites might also alter the physiological as well as reproductive functions of hosts. This might also lead to decreased growth of fish (Le Cren, 1951). It has been reported earlier that condition factor tend to be higher due to higher food quality and availability (Polacik *et al.*, 2009). In summary, season, condition factor and microhabitat seem to have a significant impact on the helminth infection. According to Le Cren (1951) and George *et al.* (1985) the relative condition factor K_n is an indicator of general well-being of the fish. K_n greater than one (1) is indicative of the general well being of fish, whereas its value less than one (1) indicates that fish is not in a good condition. However, Salam *et al.* (2005) pointed out that 'K' remained constant with increase in length and weight of fish. Yousuf and Pandit (1989) suggested that condition factor of *S. niger* varied seasonally in close association with gonadal development and feeding intensity. Mahapatra and Datta (2004) attributed low mean K_n values in *Aristichthys nobilis* to spawning strain, spent condition and low feeding rate. Likewise, Hatikakoty and Biswas (2004) suggested increase in the weight of body due to weight of mature gonads.

Chatterji (1979), had also related the changes in condition factor with age, feeding intensity and gonadal development.

The present study showed that some of the physico-chemical features showed a significant positive correlation with the prevalence. A positive correlation existed between the water temperature and pH with the prevalence of all the three parasites while as it was negatively correlated with the DO. With regard to free Carbon dioxide, except *Pomphorhynchus kashmirensis*, both the cestodes showed positive correlation. Modu *et al.* (2011) showed that there existed a significant correlation between Helminth infection and water quality parameters in a pond. A number of workers (Beer and German, 1993; Kennedy and Watt, 1994; Marcogliese, 2001; Lafferty and Kuris, 2005) have suggested that natural abiotic factors such as temperature, oxygen, salinity, hydrogen ion concentration and eutrophication have a positive influence on the occurrence of parasite populations. Evidence from the present study suggested that the water temperature played an important role in the progression of all the three parasitic infection. Dissolved O₂ had been found to be the predictor of habitat selection in monogeneans (Ernst *et al.*, 2005). Our results, however, showed insignificant relationship between O₂ level and all three intestinal parasites.

5.3 Metal concentration in water

The concentration of the twelve analyzed heavy metals in the water samples from Dal Lake and River Jhelum displayed significant spatial distribution, suggesting similar patterns of heavy metal enrichment of water within the water bodies. In Dal Lake the ranking of mean concentration of 12 metals at Dalgate, Saidakadal, Hazratbal and Telbal were Fe > Ca > Mn > Al > Zn > Cd > Cu > Ni > Co > Cr; Fe > Mn > Ca > Al > Zn > Cd > Ni > Co > Cr > Cu; Al > Fe > Ca > Mn > Zn > Cd > Ni > Co > Cr > Cu and Fe > Al > Ca > Mn > Cu > Ni > Co > Zn > Cd > Cr respectively. In Jhelum river the ranking of the metals at Khanabal, Zero bridge, and Chattabal weir were Fe > Ca > Al > Mn > Zn > Co > Cd > Cu > Cr > Ni ; Ca > Fe > Al > Mn > Zn > Ni > Cu > Co > Cd

>Cr; and Fe > Mn > Ca > Al > Co > Zn > Cd > Ni > Cu > Cr respectively. Studies with regard to various metal concentrations in water have been made by earlier workers and inference has been deduced that the heavy metal concentrations along the lake shores are influenced by anthropogenic activities (Mwamburi 2003). Although there has been a progressive increase in the heavy metal concentrations in the lake over the past 20 years (Wandiga 1981; Wandiga *et al.* 1983; Ochieng 1987; Onyari and Wandiga 1989; Mwamburi and Oloo 1996/1997; Kishe and Machiwa, 2003), the concentration of the twelve heavy metals examined in this study are still low, compared with heavy metal concentrations observed in water systems in industrialized countries of Europe and America (Schuldermann *et al.* 2003; Thielen *et al.*, 2004; Eira *et al.*, 2009).

Heavy metals in water can be partitioned into dissolved and suspended fraction. It is well known that most dissolved heavy metals are present as organic complexes in natural water (Prego and Cobelo-Garcia, 2003). A fraction of metals is bound to inorganic matters and particulate in water, which reduced the amount of metals for uptake by organism and the ability of metals to affect organism. It is known that bioavailability or toxicity of metals is directly corrected to concentration of free metals ions, which are not bounded to any matter, rather than to total concentrations (Cambell, 1995; ATSDR, 2006).

In the present study, the concentrations of lead and mercury in all the water samples from almost all the sampling sites were found to be below the instrumental detection limit. However, in some samples Cu concentrations of 1.83 mg/l was recorded at Telbal site of Dal Lake during summer and Zn concentrations of 5.31 mg/l was recorded at Khannabal site of River Jhelum during summer while no such contamination was noted in all the other water samples from different sites. The possible explanation for these two heavy metals could be the contribution of the non-point sources of pollutions, especially from agricultural fields that might have used copper containing fertilizers and pesticides. Copper is commonly a natural element in sediment and water. This

metal is non soluble in aqueous media, while lots of its salts are really soluble. It is known that metals accumulate on sediment surface, planktonic organisms, in benthic living things and other living matter which is enhanced through food chain (Javed and Usmani, 2011).

The undetectable concentrations of lead, mercury, copper in other sites in River Jhelum, and zinc in various sites of Dal Lake in water samples might be the result of adsorption and accumulation of metals by suspended solid. Chapman (1992) stated more than 50% of the total metals present (and up to 99.9%) in water are usually adsorbed onto suspended particles. When the dissolved metal concentrations in the Dal lake and River Jhelum waters in the seven sampling sites were compared with international standards, the obtained results obviously showed that the concentration of the heavy metals (Cd, Pb, Hg, Al, Ca and Zn) in Dal Lake and (Cu, Co, Hg, Al, Ca and Pb) in Dal Lake and River Jhelum did not exceed WHO/FEPA. This might be due to most metals being lower in concentration or being adsorbed onto suspended particulate matter. Several mechanisms indicated that heavy metals in water are removed due to (1) adsorption onto particulate matter; (2) chemical transformation in to insoluble form; and (3) precipitation and sedimentation (Balasubramania *et al.*, 1997).

5.4 Metal concentration in fish tissue and parasites

Fish are important aquatic organisms that are used as bio-indicators of aquatic ecosystems for estimation of heavy metal pollution and risk potential for human consumption (Agarwalet *al.*, 2007). Bioaccumulation of metals in fishes takes place directly, from the water by gills and indirectly from food (Barron, 1990). Bioaccumulation of metal by an organism is the consequences of the interactions between physiological factor (growth, weight loss, absorption and accumulation), chemical factors (metal concentration, speciation and bioavailability) and environmental factors (temperature, pH, water hardness, conductivity, salinity and food concentration (Casas and Bacher, 2006).

The difference in the pattern of heavy metals distribution in the fish might be a result of their difference in many factors such as; feeding habits, habitats, ecological needs, metabolism, biology and physiology (Arellano *et al.*, 1999). Generally, heavy metal uptake occurs mainly from water, food and sediment (Canli *et al.*, 1998). However, the efficiency of metal uptake from contaminated water and food may differ in relation to ecological needs, metabolism, and the contamination gradient of water, food and sediment, as well as other environmental factors such as salinity, temperature and interacting agents (Heath, 1987 and Pagenkopt, 1983). *Schizothorax niger* in nature revealed that they are omnivorous fishes. Jan and Das (1970) observed Kashmir fishes prefer phytoplanktons (diatoms, green and blue green algae) followed by detritus and sand which can indicate the bioavailability of heavy metals from sediments (Luoma, 1983). These fishes are benthopelagic, feeds on plants and zoobenthos which might accumulate heavy metals from their food. This is agreed with the literature which reported that some plant and animal taxa such as crustacean and mollusca have high potential for accumulation of metals and other pollutants even from much diluted solutions without obvious noxious effects (Ali and Fishar, 2005).

In the present study, in Dal lake *Pomphorhynchus kashmirensis*; *S.niger* liver and muscle showed the ranking of the 12 metals as Ca > Fe > Mn > Cr > Cu > Ni > Zn > Al > Cd > Co; Ca > Fe > Zn > Al > Cu > Mn > Cr > Ni > Co > Cd and Cd > Ca > Cr > Zn > Cu > Mn > Al > Ni > Co > Cd, respectively. In river Jhelum these ranked as Ca > Fe > Mn > Cr > Al > Cu > Ni > Zn > Co > Cd; Ca > Fe > Zn > Al > Mn > Ni > Cu > Co > Cr > Cd and Zn > Fe > Ca > Cu > Cr > Al > Mn > Co > Ni > Cd respectively.

In Dal lake *Bothriocephalus acheilognathi*; *S. niger* liver and muscle revealed the ranking of the 12 metals as Fe > Ca > Al > Zn > Mn > Cr > Cu > Ni > Cd > Co; Fe > Ca > Cu > Zn > Al > Mn > Ni > Cd > Cr > Co and Fe > Ca > Zn > Al > Cr > Ni > Cu > Mn > Cd > Co respectively. In river Jhelum these ranked

as Fe > Ca > Zn > Al > Cu > Mn > Cr > Ni > Co > Cd; Fe > Ca > Zn > Cu > Al > Mn > Ni > Cr > Cd > Co and Ca > Fe > Zn > Al > Cr > Mn > Ni > Cu > Co > Cd respectively.

In Dal lake the *Adenoscolex oreini*; *S. niger* liver and muscle, the ranking of the 12 metals were Ca > Zn > Fe > Mn > Al > Cu > Co > Ni > Cd > Cr; Fe > Ca > Mn > Al > Cu > Zn > Cd > Ni > Co > Cr and Fe > Ca > Al > Mn > Zn > Cu > Ni > Cd > Cr > Co respectively. In river Jhelum the ranking were Ca > Fe > Cr > Mn > Al > Zn > Cu > Cd > Ni > Co; Ca > Fe > Al > Zn > Cu > Mn > Ni > Co > Cd > Cr and Ca > Fe > Al > Zn > Cu > Cr > Ni > Mn > Co > Cd, respectively.

The patterns of metal concentrations in both water bodies were variable which could be due to bioavailability, intrinsic fish processes, and trophic structure variation. The specificity of concentrations of heavy metals irrespective of the locality of fish capture and the route of uptake of the metals has been reported (Obboh, 2007; Eneji, 2010). The variability observed in the fish species is a reflection of different thresholds of metals which are a function of homeostasis. The thresholds of metals in fish can be considered as the concentration level where the metal starts to interfere with the variable physiology of the fish species in such manner that once a particular level of the metal has been sequestered in the body, equilibrium is established between the fish burden and the ambience. Also, Olaifa *et al.* (2004) reported that fish species can accumulate heavy metals above the abiotic environment to incur bioaccumulation. Species difference in heavy metals bioaccumulation could be linked to difference in feeding habits and behavior of the species (Altindag and Yigit, 2005).

The general pattern of heavy metal accumulation in this study exhibited the following progression: fish parasite > water > fish liver > fish muscle. As a result of the low heavy metal concentrations in the lake water and fish, it appears that the acanthocephalan parasite *Pomphorhynchus kashmirensis* and two intestinal cestodes, *Adenoscolex oreini* and *Bothriocephalus acheilognathi* were

better at accumulating heavy metals to a higher concentration than observed in their environmental media or in their respective intermediate host fish, *Schizothorax niger*. The concentration of Mn, Ca, Cd and Ni; Ni, Mn, Cr, Al, Cd and Co and Zn, Co, Mn, Cu, Cr, Al, Fe and Cd were found higher in *Pomphorhynchus kashmirensis*, *Bothriocephalus acheilognathi* and *Adenoscolex Oreini*, respectively. Malek *et al.* (2007) hypothesized that, because cestodes cannot synthesize their own cholesterol and fatty acids, they efficiently obtain them from their host's intestinal lumen, thereby reducing the absorptive capacity of the heavy metals in the fish body cavity with the presence of parasites. Considering that *L.intestinalis* has a very large surface area-to-volume ratio (Cowx *et al.*, 2008) in the host, they can easily take up organo-metallic complexes, which have been released from the host's bile duct to the small intestines. Consequently, these metals can accumulate in the body of the parasites, thereby rendering the parasites as sinks for these metals. It is also possible that fish have developed effective mechanism by which many heavy metals are transported, stored and excreted (Bijvelds *et al.*, 1998). Such cellular mechanism possibilities, however, have not yet been reported for cestodes. Thus, the higher bioaccumulation of metals in the cestode could be reflected by this physiological mechanism.

The cestode parasites exhibited a consistent site-specific bioaccumulation pattern in this study. The highest bioaccumulation factor of the parasite was exhibited for River Jhelum. This finding might indicate a coupling of geological metals from the environment that could increase the metal burden in fish. It is probable, therefore, that competition for elements between hosts and parasites for essential elements could lead to increased absorption of other essential heavy metals, including Cu. The Cu concentration might be regulated by the fish, as well as by the parasite, albeit the physiologically required concentrations are actually higher for the parasite. This higher Cu accumulation in the parasite, therefore, cannot be considered as the bioaccumulation of environmental pollutants. It was

also observed that fish with higher abundances of endoparasites exhibited higher Cd and Cr concentrations. This finding, however, cannot be interpreted as meaning that fish heavily infested with parasites necessarily contain higher metal concentrations from the parasites. Rather, it could be associated with disruption of fish physiological functions by the parasites that impair the ability of the fish to regulate metal contents in their tissues. Moreover, infested fish already weakened by the parasite infection might be more vulnerable to other suboptimal or adverse environmental conditions, such as food shortage or water pollution. Fish endoparasites, mainly *L. intestinalis*, have been reported to affect normal physiological functions (Cowx *et al.*, 2008), some of which would render their host prone to metal bioaccumulation from the aquatic environment. The parasitized fish samples in this study were observed to be heavier than the unparasitized fish samples, contrary to the observations of Sures (2003) for acanthocephalans heavily infested by parasites. This scenario might not be related to either the metal content or kinetics in the fish, but rather by hormonal disturbances through endocrine disruption (Jobling and Tyler, 2003). Kennedy (1997) has outlined the disadvantages of using fish parasites for this purpose, including the possibility of the parasite being directly or indirectly influenced by the host metabolic factors or immune system, by parasite over dispersion and aggregation, by inadequate knowledge of their physiological responses to metal pollution, and, in this study, to site-specific variations in metal accumulation. In regard to the bioaccumulation factor, it appears that *Adenoscolex oreini* and *Pomphorhynchus kashmirensis* is a sensitive indicator and early warning sign for Al, Cd and Cr pollution in water systems. As this parasite is easy to identify, even in different hosts (Olson *et al.*, 2000), because of its high abundance and high prevalence, these should be considered as a biomonitor model and early warning sign for localized Al, Cd and Cr pollution in water bodies.

Copper and zinc are essential elements and are regulated by physiological mechanisms in most organisms. However, they show toxic effects

when organisms are exposed to levels higher than normally required (Biney *et al.*, 1994). The levels of Cu and Zn in the muscle were significantly different during different seasons. This shows that there is a variation of the accumulations of Cu and Zn in the edible muscle of *Schizothorax niger* in the two water bodies. Copper is a fundamental micronutrient to all forms of life in enzyme activity or random rearrangement of natural protein (Bower, 1979). Copper is available in surface water and ground water due to the extensive use of pesticides sprays containing copper compounds for agricultural purposes. It is an essential element in human metabolism but can cause anemia, disorders of bone and connective tissues and liver damage at excessive levels. The toxicity of copper depends upon the hardness and pH of the water, and therefore, it is more toxic in soft water and in water with low alkalinity (Taha, 2004). The elevation of copper accumulation in this study might have been due to industrial and sewage wastes. These results agree with those obtained by (Ibrahim and Mahmoud, 2005) and (Tayel *et al.*, 2008) who revealed that this increase is anticipated to industrial, drainage and sewage effluents. Also, it may be due to elevated metal-binding protein synthesis as recorded by Yacoub (2007).

Fish can biomagnify toxic substances, particularly those which are least soluble in water because they are in close contact with the source that brings these toxic substances in suspension or solution form. Besides this, fish use water soluble oxygen by filtering the massive quantity of water through their gills. Kargin (1991) reported that biomagnifications of metals in different organs of the fish is mainly concerned with different factors such as season, chemical, and physical properties of aquatic body. Exploitation of fresh water habitation may occur from any activity that changes the chemical and physical characteristics of a river or stream (Charles, 1992). Zinc is one of the essential elements as copper, and cobalt for both animals and humans. A deficiency of zinc is marked by retarded growth, loss of taste and hypogonadism, leading to decreased fertility. Zinc toxicity is rare, but at concentrations in water up to 40 mg/l, may induce

toxicity, characterized by symptoms of irritability, muscular stiffness and pain, loss of appetite, and nausea (NAS-NRC, 1974). Zinc appears to have a protective effect against the toxicities of both cadmium (Calabrese *et al.*, 1985) and lead (Sanstead, 1976). Roesijadi and Robinson(1994) mentioned that the heavy metals such as Zn and Cu accumulation pattern have shown much variation in the fish organs. High level of Zn and Cu in metabolically active organs that is liver represented much accumulation due to strapping of these metals to metallothionine which acts as detoxifying agent. On the other hand metabolically less active tissues such as muscle showed low magnitude of accumulation.

Zinc is an essential element and is a common pollutant as well. Mining smelting and sewage disposal are major source of zinc pollution. Fish take it up directly from water, especially by mucous and gills (Skidmore, 1964). The high accumulation of zinc in studied fish liver agrees with Hamed (1998). Ibrahim and Mahmoud (2005) revealed that this increase is anticipated to industrial effluents from Talkha Electricity station and sewage from El-Rahawy drain. The relatively higher zinc concentration in the liver of the different fish species may be due to the role of zinc as an activator of numerous enzymes present in the liver as recorded by (Yacoub, 2007) and (Cogun *et al.*, 2005).

Iron is an abundant and important element, unsurpassed by any other heavy metals in the earth's crust (Ibrahim and Tayel, 2005). The increase of iron accumulation in fish liver in this study may be related to the increase of total dissolved iron in water and consequently increase free metal iron concentration and thereby lead to an increase in metal uptake by different organs (Tayel *et al.*,2008; Carbonell *et al.*,1998). Haggag *et al.* (1993) and Yacoub (2007) observed accumulation of iron ligand protein (Hemosidrin) scattered in liver section of fish exposed to high iron concentration.

Nickel is also an essential micronutrient required for red blood cells. It is known to be toxic at high intakes. The long term exposure can cause decrease in body weight, heart and liver damage and skin irritation. The toxicity of Ni to

aquatic life has been shown to vary significantly with organism species, pH and water hardness (Birge and Black, 1980). In this study, the mean accumulation of Ni in muscle of *Schizothorax niger* fish was above the recommended level of 0.80 µg/g. The accumulation of Ni was significantly different during different seasons.

Manganese functions as an essential constituent for bone structure, reproduction and normal functioning of enzymes system (Fleck, 1976). It is toxic only when present in higher amount, but at low level is considered as micronutrient (Sarkka *et al.*, 1978). The high accumulation of Manganese in fish liver obtained from Dal Lake might be due to sewage deposits from sewage treatment plant. These results agree with those obtained by Yacoub (2007).

Cadmium is highly toxic non-essential heavy metal and does not seem to have a role in biological processes in living organisms. Thus even in low concentration cadmium could be harmful to living organisms (Burden *et al.*, 1998). In the present study the concentration of cadmium was higher in all the three parasites followed by liver tissue. Muscles showed the lowest levels of Cd. The values of cadmium accumulation in present study range from 0.78 µg/g in Dal Lake and 0.19 µg/g in River Jhelum were higher than those obtained by Yacoub (2007) as 0.04 - 0.28 µg/g dry weight. High accumulation of cadmium in liver may be due to its strong binding with cystine residues of metallothionein (Tayel *et al.*, 2008). Cadmium is rarely found in natural water (Hem, 1989). It is considered to be toxic if its concentration exceeds 0.01 mg/l both in drinking and irrigation water (Taha, 2004). Cadmium with some other heavy metals lead and mercury are of no biological function in human system and they are potentially toxic even at trace concentrations (Robert, 1991). The effects of acute cadmium are high blood pressure, kidney damage, destruction of testicular tissue as well as destruction of red blood cells (Gupta and Mathur, 1983).

Although fish muscle is the most important part to be used for human consumption, fish skin and liver may also be consumed to some extent. Target

organs such as liver, kidney, gonads and gills, have a tendency to accumulate heavy metals in high values, as shown in many species of fish in different areas (Kargin, 1996; Yilmaz, 2003; Yilmaz, 2005 and Abdel-Moniem, *et al.*, 1994). It is generally accepted that muscle is not an organ in which metals accumulate (Legorburu *et al.*, 1988). Similar results were reported from a number of fish species showing that muscle is not an active tissue in accumulating heavy metals (Karadede and Unlo, 2000). This is agreed with the present study. The levels of heavy metal in fish vary in various species and different aquatic environments (Canli and Atli, 2003).

The presence of trace metals, particularly Zinc in River Jhelum might due to the agricultural influx and sewage via surrounding cultivated lands. Consequently, it can be concluded that the concentration of nickel (0.80 µg/g), manganese (1.69 µg/g), cadmium (0.78 µg/g) and chromium (2.86 µg/g), aluminum (2.99 µg/g) were higher than the permissible limits. In River Jhelum the maximum concentration of nickel (0.80 µg/g), manganese (0.41 µg/g), aluminum (2.99 µg/g), cadmium (0.19 µg/g) and chromium (0.82 µg/g) in the muscles of infected fishes were higher than the permissible limits. Accumulation of heavy metals in fish viscera and other organs may be considered as an important warning signal for fish health and human consumption. The present study shows that precautionary measures are needed to be taken in order to prevent future heavy metal pollution.

5.5 Correlation among metal concentrations of water, fish tissues, and parasites, and physicochemical parameters of Dal Lake

Surface water temperature is one of the important factors affecting aquatic environments for two reasons. First, water temperature affects nearly all other water parameters and second aquatic organisms are adapted to certain temperature range. It exerts an important effect on metal speciation because most chemical reaction rates are highly sensitive to temperature change (Prosi, 1989). Due to increased temperature effects are seen both on uptake and elimination rates

of metals, so net bioaccumulation may or may not increase. Average temperature was same throughout the sampling sites during a season. In the present study Cr, Mn, Cu, Zn, Al, Ca concentrations in water showed positive correlation with water temperature. Cu concentration in *Pomphorhynchus kashmirensis* and *Adenoscolex oreini* showed positive correlation with water temperature. Cr concentration in *Pomphorhynchus kashmirensis* showed positive correlation with water temperature.

Fe, Zn, Cd Cu and Co concentrations in water showed positive correlation with free CO₂.

The water chemistry of the system controls the rate of adsorption and desorption of metals to and from sediment. Adsorption removes the metal from the water and stores the metal in the sediment and suspended solid. Desorption returns the metal to the water column, where recirculation and bio-assimilation may take place. Metals may be desorbed from the sediment and suspended solid if the water experiences increases in salinity, decreases in redox potential, or decreases in pH. A lower pH increases the competition between metals and hydrogen ions for binding sites. A decrease in pH may also dissolve metals-carbonate complexes, releasing metals ions into the water column (Osmond *et al.*, 1995). pH has an impact on solubility and bioavailability of metals in the natural water. The lower the pH the higher the solubility of heavy metals, and thus increases in metal bioavailability (Waite *et al.*, 1984). Cu and Cr in *Pomphorhynchus kashmirensis* showed positive correlation with pH of water. Zn and Cr in *Bothriocephalus acheilognathi* showed positive correlation with pH of water. Cd concentration in *Adenoscolex* showed positive correlation with pH of water.

A decreased redox potential, as it is often seen under oxygen deficient conditions, will change the composition of metal complexes and release the metals into the overlying water (Osmond *et al.*, 1995). During the sampling period of this study, the dissolved oxygen was found to be in the range 3.55 to 6.78

mg/land 4.97to 8.16 mg/l in Dal Lake and River Jhelum, respectively. Also, in all the sites pH of the water measured to be alkaline. These oxygen and pH conditions are not favorable for the solubility of the metals; it was not expected to find high amounts of dissolved metals in water samples. Zn concentration of water and Fe concentration in *Bothriocephalus acheilognathi* was positively correlated with DO. However, the two parasites which have relation directly with water are *Adenoscolex oreini* and *Pomphorhynchus kashmirensis*. *Adenoscolex oreini* was positively correlated with Cd concentration in water during autumn and winter whereas *Pomphorhynchus kashmirensis* was positively correlated with Al and Cr concentration in water. Therefore, the two parasites seem to act as bio-indicators for the corresponding metals. The solubility of heavy metals is predominately controlled by the water pH (Osmond *et al.*, 1995), water temperature (Iwashita and Shimamura, 2003; Papafilippaki *et al.*, 2008) and the redox environment of the lake water (Osmond *et al.*, 1995; Iwashita and Shimamura, 2003; Papafilippaki *et al.*, 2008). The behavior of metals in surface water is reported to be a function of the substrate sediment composition, the suspended solid composition and the water chemistry (Osmond *et al.*, 1995).

5.6 Histopathology of parasitic infections of *Schizothorax niger*

Histopathologically, all the three parasites seemed to induce lesions in intestines characterized by various intensities of enteritis coupled with hyperplastic goblet cells with increased acid mucopolysaccharide concentrations. *Pomphorhynchus kashmirensis* was severely pathogenic compared to other two cestodes which could be the result of the spined proboscis of the acanthocephala. The degenerative lesions of liver could be the result of various toxic metal concentrations in water in addition to the additive damaging effect of parasites. Generally, acanthocephalans cause more damage to the intestinal tissues and induce more complex host response, mainly, due to deeper penetration into the gut mucosa and worm burdens (Bullock, 1963). The pathogenicity caused by acanthocephalans also depends on the parasite/host species and site of localization

(Esch and Huffines, 1973; Hine and Kennedy, 1974; McDonough and Gleason, 1981 and Hamers *et al.*, 1992). The fishes showed grayish or yellowish discoloration with various degrees of emaciation and viscera looked reddish pink on opening the abdomen which might be due to deeper penetration of proboscis and bulb and can be used as an indicator for the presence of parasite. This finding is similar to those reported by Dezfuli (1991) and Dezfuli *et al.*(2002) for *P. laevis* infection in *Leuciscus cephalus*. Sometimes, the proboscis of *Pomphorhynchus* perforates the capsule and penetrates the liver (Chaicharn and Bullock, 1967; McDonough and Gleason, 1981 and Taraschewski, 1989). Excessive mucous production at the host-parasite interface may be a consequence of host reaction for defence, as mucous layer on intestinal mucosa acts as a physical barrier for microorganisms, parasites and their toxins (Lamont, 1992 and Bosi *et al.*, 2005) and therefore prevents secondary infection. Excess mucous secretion and significant increase in the number of mucous cell has been reported in the infected fish with acanthocephalans as well as in infected mammals with other parasites, which was correlated with the defence of host (Bosi *et al.*, 2005). Significant increase in the number of goblet cells had been shown in the parasitized pyloric caeca of green sunfish, *Lepomis cyanellus* with *Leptorhynchoidesthecatus*, which secrete excessive mucus and provide defence to the host (de Buron and Nickol, 1994). *P. kashmiriensis* parasites had pierced the intestinal wall and were firmly attached and to be deeply penetrated till lamina propria and thereby causing erosion of villi and epithelial layer. Similarly, complete penetration of proboscis and bulb and thereby fibrosis, hemorrhage, inflammation and cell necrosis have been reported in infected fishes with several acanthocephalan species (Bullock, 1963; Chaicharn and Bullock, 1967; McDonough and Gleason, 1981; Dezfuli, 1991; Taraschewski, 1989 a; Wanstall *et al.*, 1986; Abu El-Ezz, 1988 and Dezfuli *et al.*, 2002, 2008). The deeper penetration of proboscis and bulb may be assisted by proteolytic enzyme as suggested by Polzar and Taraschewski (1994) for deeper penetration of *P. laevis*. It was further shown that those acanthocephalan species which, lack the activity of this enzyme cannot penetrate the collagenous structure

and take several days to weeks to penetrate the intestinal wall (Taraschewski, 1989). The compression/erosion of epithelial cells in the areas of trunk contact with the host tissues is similar to those reported by Bullock (1963) and Chaicharn and Bullock (1967) for *Acanthocephalus jacksoni* and *P. Bulbicoli* infections respectively. Intense host reactions and cellular infiltration at the site of *P. kashmiriensis* attachment was similar to those reported by many workers on other species of *Pomphorhynchus* (Esch and Huffines, 1973; Hine and Kennedy, 1974; McDonough and Gleason, 1981 and Dezfuli, 1991). Chronic inflammation leading to an increased amount of connective tissue, thickening of lamina propria and infiltration of leucocytes has been reported at the site of *A. jacksoni* attachment (Bullock, 1963). Hamers *et al.* (1992) reported inter-specific differences in the response of leucocytes in infected fish with *P. ambiguous* and suggested that the cellular defense might be involved in determining the host specificity as unsuitable host expel the parasite within a few days.

The histopathological changes caused by cestodes were varied in severity with the season and parasitic burden. The fish infected with *Bothriocephalus acheilognathi* were anemic and emaciated. The viscera were dark red on opening the abdomen. The intestinal contents showed lot of mucous and contained dark contents. The mucosal wall was red in colour and also revealed necrotic surface. Fishes infected with *Adenoscolex oreini* were anemic and the abdomen appeared slightly pot bellied. Viscera appeared red on opening the abdomen and the abdominal fluid was tinged red. On opening the intestine necrotic debris was present on the surface and numerous parasites were present. The histopathological alterations observed in the intestine of fish (severe degenerative and necrotic changes in the intestinal mucosa as well as edema between submucosa and mucosa) might be the result of cestodes, and has previously been attributed also to the uptake of toxic metals (Hanna *et al.*, 2005).

The contributing effect of heavy metals cannot be refuted. The present results are in agreement with those observed by many investigators about the

effects of metals on fish intestine (Giari *et al.*, 2007; Hanna *et al.*, 2005). Uptake of metals occurs mainly through gills but may also occur via intestinal epithelium (Mohamed, 2008). The histopathological alterations observed in intestine might have been also aggravated by toxic metals (Hanna *et al.*, 2005). Toxic lesions most common in the intestine of fishes exposed to cadmium chloride include atrophy in the muscularis, degenerative changes in the tips of villi and necrosis of submucosa (Kaoud *et al.*, 2011). In the intestine of *Channa punctatus* exposed to HgCl, the degenerative changes in the tips of mucosal folds, 2 hypertrophy and necrosis were observed (Sastri and Gupta 1978). Similarly, cellular debris, vacuolation in intestine of *Mugil auratus* exposed to inorganic and organic mercury were also observed in another study (Establier *et al.*, 1978). The major damage induced by cestodes consisted of necrosis and sloughing of stomach and intestine epithelium. At the site of infection, numerous RCs and mucous cells were seen in the epithelium. Rodlet cells are exclusive to fish, and their ultrastructure is well known. Data from several recent surveys of wild and farmed fish support the suggestion that RCs are an immune cell type closely related to other piscine inflammatory cells such as mast cells (Dezfuli *et al.*, 2008; Jordanova *et al.*, 2007; Reite, 2005; Reite and Evensen, 2006; Vigliano *et al.*, 2009). The attachment organ of endoparasitic helminths often provokes inflammation of the host gastrointestinal tract. Inflammation is a protective reaction of the host in response to injury, resulting in specific chemical and morphological alterations in cells and tissues (Suzuki and Iida, 1992). The first level of defense consists of the substances secreted into the lumen, including mucus, bicarbonate, acid, immunoglobulins, and other antibacterial and surface-active phospholipid materials (Martin and Wallace, 2006; Wallace and Ma, 2001). Severe enteritis with heavy infiltration of inflammatory cells and fibroblasts were seen in lamina propria. Lamina epithelialis showed severe desquamation. Necrotic intestinal villi were evident. Goblet hyperplasia was seen alongwith elucidation of acid mucopolysaccharide. It appears that several peptides involved in the regulation of intestinal mucus secretion are released during inflammation (Fairweather, 1997;

Lamont, 1992; Plaisancie *et al.*, 1998). Fish mucus is involved in a wide range of functions, including respiration, reproduction, excretion, feeding, ionic and osmotic regulation, and protection against, and resistance to, disease (Schroers *et al.*, 2009; Shephard, 1994; Smirnova *et al.*, 2003; Yan *et al.*, 2007). It has been reported that, in some fish species, mucous cells produce and release defensive materials (Cho *et al.*, 2002; Nakamura *et al.*, 2001).

Although, the present study did not include all these findings, yet we observed severe degenerative changes in villi structure. In brown trout naturally infected with an acanthocephalan, the number of mucous cells increased significantly, and copious mucus secretion appeared as an adherent blanket around the worm body, at the site of infection (Bosi *et al.*, 2005). In a comparison of uninfected to infected brown trout intestines, helminths were associated with increased thickness of the mucus layer (Bosi *et al.*, 2005). In the present study, in parasitized *Schizothorax niger*, hyperplasia of intestinal mucous cells and enhanced mucus secretion were documented. Cestode bodies were often covered with an adherent mucus blanket. Our data are in agreement with the suggestion that the mucus gel layer protects the underlying epithelium as a physical barrier against pathogens and their toxins (Lamont, 1992; Schroers *et al.*, 2009). *C. rudolphii* larvae induced severe damage within the tunica propria and on the external surface of the stomach and intestine, with conspicuous granulomas. Data on fish granulomas provoked by helminths have been reported by Taraschewski (1988, 1989) and Karanis and Taraschewski (1993). Cellular composition and zonation of fish granulomas appear to be similar to that of granulomas observed in other vertebrates (Boros, 1978). In *A. anguilla* granulomatous tissue was formed mainly by fibroblasts which were interspersed with mast cells and a small number of scattered macrophages.

It has been reported that mast cells are major effector cells in the immune response to helminth infection (Dezfuli *et al.*, 2008; Sharp *et al.*, 1989) and suggested that mast cells or their products are pivotal in mediating leukocyte

recruitment to inflammatory sites (Mekori, 2004). Mast cells have been associated with defense against bacteria (Wedemeyer *et al.*, 2000) and metazoan parasites (Dezfuli *et al.*, 2000, 2008; Dezfuli and Giari, 2008; Reite, 2005). Their primary function is considered to be stimulating the activation of cells such as neutrophils to kill pathogens (Reite and Evensen, 2006), but some evidence suggests that they may also participate directly in killing microbes (Murray *et al.*, 2007; Silphaduang *et al.*, 2006; Silphaduang and Noga, 2001). Recently it has been reported that the mast cells of Perciformes, the largest and most evolutionarily advanced order of teleosts, contain histamine, which regulates the fish inflammatory response (Mulero *et al.*, 2007, 2008). A close relationship between mast cells and fibroblasts in various fish species has been reported (Flaño *et al.*, 1996; Kent *et al.*, 1993). In mammals and fish, several lines of evidence indicate that mast cells are involved in the fibrotic process and tissue remodeling (Dezfuli *et al.*, 2008; Hrckova *et al.* 2006; Metcalfe *et al.*, 1997; Rocha and Chiarini-Garcia, 2007).

Liver of fish is sensitive to environmental contaminants because many contaminants tend to accumulate in the liver and exposing it to a much higher levels than in the environment, or in other organs (Oliveira-Ribeiro *et al.*, 2002). The liver of fish infected with cestodes showed marked histopathological changes. Degeneration and necrosis of the hepatocytes might be due to the cumulative effect of metals and the increase in their concentrations in the liver. These results agreed with Authman and Abbas(2007) who stated that the liver has an important detoxical role of endogenous waste products as well as externally derived toxins as heavy metals. The cellular degeneration in the liver might also be due to oxygen deficiency as a result of gill degeneration and/or to the vascular dilation and intravascular haemolysis observed in the blood vessels with subsequent stasis of blood (Mohamed, 2001). Many authors have reported similar histopathological alterations in the liver of fish exposed to metals (Athikesavan *et al.*, 2006; Triebskorn *et al.*, 2007; Van *et al.*, 2007). The liver showed degeneration of the hepatocytes, congestion of central vein and nuclear pyknosis in the majority of

hepatic cells. These findings were apparent as the liver is considered to be the organ of detoxification, excretion and binding proteins such as metallothionein (MTs). The metal-binding proteins in the nuclei of hepatocytes have been suggested to increase the cell damages (De Smet and Blust, 2001). Similar results were observed by Van Dyk (2003) and Mela *et al.* (2007). Liver of fish is sensitive to environmental contaminants because many contaminants tend to accumulate in the liver and exposing it to a much higher levels than in the environment, or in other organs (Heath, 1995). Pacheco & Santos (2002) described increased vacuolisation of the hepatocytes as a signal of degenerative process that suggested metabolic damage, possibly related to exposure to contaminated water. Presence of mainly glycogen in the vacuoles of the hepatocytes shown by PAS method in the present study, however, suggested that, the vacuolization need not be related to a degenerative process. The liver parenchyma of the fish showed signs of degeneration (cytoplasmic and nuclear degeneration, and nuclear vacuolation). These alterations are more severe and have been associated with the exposure of the fishes to contamination by metals, such as copper (Paris-Palacios *et al.*, 2000) and mercury (Oliveira Ribeiro *et al.*, 1996), and by polychlorinated biphenyls (PCBs) (Chang *et al.*, 1998). The histological changes observed in the intestines and liver of the *S. niger* in the present study indicated that the fish were responding to the direct effects of the contaminants as much as to the secondary effects caused by stress. The analysis of the seasonal variation in the histological parameters leads to the conclusion that the changes observed in the three organs were not apparently related to the seasons, and neither were the distribution or the severity of the lesions. Such information confirms that histopathological alterations are good biomarkers for field assessment, in particular in tropical areas that are naturally subject to a multiplicity of environmental variations. It must be emphasized that histopathology is able to evaluate the early effects and the responses to acute exposure to parasitic infections and chemical stressors.

Chapter6

SUMMARY AND CONCLUSION

The present study was undertaken with the objectives to determine the incidence of helminth parasites in fishes with special reference to water quality parameters, and histopathological changes caused by the residues of these metals in the liver and intestines effected by different parasites, also to assess the concentration of various metals in water, fish tissue and helminth parasites of the fishes in Dal Lake and River Jhelum and show correlation among these metal levels and physicochemical parameters. For this purpose water and fish samples were collected seasonally. Water samples from the selected sites were collected 10cm below the surface. Three replicate of surface water samples were collected from three different points of the same location using polyethylene sampling bottle. The water samples were acidified immediately after collection by adding 5 ml nitric acid to minimize adsorption of heavy metals onto the walls of the bottles and stored at 4°C until metal analysis. Fish species (*Schizothorax niger*, *Schizothorax esocinus*, *Schizothorax curvifrons*) along with their parasites were collected randomly from two water bodies with the aid of local fishermen, quickly killed and stored on ice.

All the four physicochemical parameters Temperature, Dissolved oxygen, pH and free carbon dioxide showed variation vis-a-vis the season and location of the stations in water bodies. Highest temperature in Dal Lake was seen at Dalgate during summer ($27.22 \pm 1.71^{\circ}\text{C}$) while the lowest temperature was also recorded at the same site during winter ($6.55 \pm 1.48^{\circ}\text{C}$). Similarly, the highest temperature (19.44 ± 1.50) in Jhelum River was recorded at Chattabal weir while lowest ($4.38 \pm 1.32^{\circ}\text{C}$) was recorded at Khannabal indicating that the water near the source of origin is at lower degree than down the river flow. Besides the temperature of water in Jhelum River is at lower degree than that of Dal Lake. pH of the waters of Dal Lake and River Jhelum was varied within the alkaline range. The pH of Jhelum river water was comparatively less alkaline than that of Dal

Lake. In Jhelum the highest pH recorded at Khannabal was during spring (8.35 ± 0.51) and the lowest was at Chattabal weir during autumn (7.03 ± 0.28). In Dal Lake the highest pH was noted at Hazratbal during summer (8.47 ± 0.41) and lowest was at Dalgate during autumn (7.45 ± 0.35). In Dal Lake DO was highest at Telbal (6.78 ± 1.25 mg/l) during spring while lowest was at Hazratbal (3.55 ± 1.19 mg/l) during summer. In River Jhelum DO was highest at Khannabal (8.16 ± 1.43 mg/l) during spring while lowest was at Zero bridge (4.97 ± 1.50 mg/l) during autumn. DO of River Jhelum was noted at higher level in all the seasons compared to that in Dal Lake. Free carbon dioxide in Dal Lake was highest (12.88 ± 2.21 mg/l) at Telbal during spring while the lowest was found at Saidakadal (3.16 ± 0.93 mg/l) during autumn. In River Jhelum highest free CO₂ (8.11 ± 2.26 mg/l) was found at Chattabal weir during summer and lowest was also recorded (3.08 ± 1.83 mg/l) at the same site during autumn. Free carbon dioxide was found higher in Dal Lake than in River Jhelum in all the seasons of the year.

Three different species of *Schizothorax* viz. *S. niger*, *S. esocinus* and *S. curvifrons* were recovered from both the water bodies. The recovery of *S. esocinus* was more from Dal Lake followed by *S. niger* and *S. curvifrons* while as from Jhelum river recovery of *S. niger* was higher followed by *S. esocinus* and *S. curvifrons*. Acanthocephalan parasite (*Pomphorhynchus kashmirensis*)(27.47%) and two intestinal cestodes (*Bothriocephalus acheilognathi*)(30.63%) and (*Adenoscolex oreini*)(32.43%) were recovered from all the three species of *Schizothorax*. In Dal Lake and River Jhelum *Pomphorhynchus kashmirensis* was more prevalent in *S. niger* and *S. curvifrons* during summer, respectively. Lowest prevalence of the parasite was during winter in both the water bodies. Overall the mean intensity of *Pomphorhynchus kashmirensis* was highest in Dal Lake whereas its abundance was higher in River Jhelum. For both the cestodes mean intensity and abundance were higher in River Jhelum. *Bothriocephalus acheilognathi* prevalence was highest in *S. esocinus* and *S. niger* in Dal Lake and River Jhelum, respectively, during summer. *Adenoscolex oreini* was seen more in

S. niger and *S. curvifrons* in Dal lake and River Jhelum during Summer and lowest during winter, respectively. Both the cestodes and the acanthocephalan infection were prevalent more in male fishes compared to females. The mean intensity and abundance of both the cestodes was highest in summer and lowest in winter. The presence of the parasites had reduced the condition coefficient of the infected fishes in both the water bodies however; the overall condition factor of Jhelum river fishes was higher. In summary, season, condition factor and microhabitat seem to have a significant impact on the helminth infection. A positive correlation existed between the water temperature and pH with the prevalence of all the three parasites while as it was negatively correlated with the DO. With regard to free carbon dioxide, except *Pomphorhynchus kashmirensis*, both the cestodes showed positive correlation.

Histopathologically, all the three parasites seemed to induce lesions in intestines characterized by various intensities of enteritis coupled with hyperplastic goblet cells with increased acid mucopolysaccharide concentrations. *Pomphorhynchus kashmirensis* was severely pathogenic compared to other two cestodes which could be the result of the spined proboscis of the acanthocephalan. The degenerative lesions of liver could be the result of various toxic metal concentrations in water in addition to the additive damaging effect of parasites.

The overall concentrations of different heavy metals in the two water bodies showed significant variations both season and location wise. In Dal Lake the concentrations of Co, Ni, Mn, Cr, Fe and Cd were higher than the permissible limits as recommended by WHO/FEPA/IAEA-407. Similarly, in River Jhelum the concentrations of Ni, Mn, Cr, Fe, Cd in water were higher than the permissible limits. Pb and Hg were below detection levels in both the water bodies while Cu and Zn were recorded only in Dal Lake and River Jhelum, respectively.

In Dal Lake the ranking of mean concentration of 12 metals at Dalgate, Saidakadal, Hazratbal and Telbal were Fe > Ca > Mn > Al > Zn > Cd > Cu > Ni

> Co > Cr; Fe > Mn > Ca > Al > Zn > Cd > Ni > Co > Cr > Cu; Al > Fe > Ca > Mn > Zn > Cd > Ni > Co > Cr > Cu and Fe > Al > Ca > Mn > Cu > Ni > Co > Zn > Cd > Cr respectively. In Jhelum river the ranking of the metals at Khanabal, Zero bridge, and Chattabal weir were Fe > Ca > Al > Mn > Zn > Co > Cd > Cu > Cr > Ni; Ca > Fe > Al > Mn > Zn > Ni > Cu > Co > Cd > Cr; and Fe > Mn > Ca > Al > Co > Zn > Cd > Ni > Cu > Cr respectively. In Dal lake the *Pomphorhynchus kashmirensis*; *S.niger* liver and muscle and the ranking of the 12 metals were Ca > Fe > Mn > Cr > Cu > Ni > Zn > Al > Cd > Co, Ca > Fe > Zn > Al > Cu > Mn > Cr > Ni > Co > Cd and Cd > Ca > Cr > Zn > Cu > Mn > Al > Ni > Co > Cd respectively. In river Jhelum were Ca > Fe > Mn > Cr > Al > Cu > Ni > Zn > Co > Cd; Ca > Fe > Zn > Al > Mn > Ni > Cu > Co > Cr > Cd and Zn > Fe > Ca > Cu > Cr > Al > Mn > Co > Ni > Cd respectively. In Dal lake the *Bothriocephalus acheilognathi*; *S. niger* liver and muscle and the ranking of the 12 metals were Fe > Ca > Al > Zn > Mn > Cr > Cu > Ni > Cd > Co; Fe > Ca > Cu > Zn > Al > Mn > Ni > Cd > Cr > Co and Fe > Ca > Zn > Al > Cr > Ni > Cu > Mn > Cd > Co respectively. In river Jhelum were Fe > Ca > Zn > Al > Cu > Mn > Cr > Ni > Co > Cd; Fe > Ca > Zn > Cu > Al > Mn > Ni > Cr > Cd > Co and Ca > Fe > Zn > Al > Cr > Mn > Ni > Cu > Co > Cd respectively. In Dal lake the *Adenoscolex oreini*; *S. niger* liver and muscle and the ranking of the 12 metals were Ca > Zn > Fe > Mn > Al > Cu > Co > Ni > Cd > Cr; Fe > Ca > Mn > Al > Cu > Zn > Cd > Ni > Co > Cr and Fe > Ca > Al > Mn > Zn > Cu > Ni > Cd > Cr > Co respectively. In river Jhelum were Ca > Fe > Cr > Mn > Al > Zn > Cu > Cd > Ni > Co; Ca > Fe > Al > Zn > Cu > Mn > Ni > Co > Cd > Cr and Ca > Fe > Al > Zn > Cu > Cr > Ni > Mn > Co > Cd, respectively.

In Dal Lake the concentrations of copper (1.25 µg/g), zinc (5.69 µg/g) and iron (29.3 µg/g) were maximum in the muscles of infected fishes which were still lower than the Maximum Limit recommended by WHO/FEPA/IAEA-407 whereas on the other the concentration of nickel (0.80 µg/g), manganese (1.69 µg/g), cadmium (0.78 µg/g) and chromium (2.86 µg/g), aluminum (2.99 µg/g) were higher than the permissible limits. In River Jhelum the maximum

concentration of nickel (0.80 µg/g), manganese (0.41 µg/g), aluminum (2.99µg/g), cadmium (0.19 µg/g) and chromium (0.82 µg/g) in the muscles of infected fishes were higher than the permissible limits. Whereas on the other the concentration of copper (0.86 µg/g), zinc (3.93 µg/g), iron (24.4 µg/g) in the muscles of infected fishes were still lower than the maximum Limit recommended by WHO/FEPA/IAEA-407.

Cr, Mn, Cu, Zn, Al, Ca concentrations in water showed positive correlation with water temperature. Cu concentration in *Pomphorhynchus kashmirensis* and *Adenoscolex oreini* showed positive correlation with water temperature. Cr concentration in *Pomphorhynchus kashmirensis* showed positive correlation with water temperature. Fe, Zn, Cd, Cu and Co concentrations in water showed positive correlation with Free CO₂. Cu and Cr in *Pomphorhynchus kashmirensis* showed positive correlation with pH of water. Zn and Cr in *Bothriocephalus acheilognathi* showed positive correlation with pH of water. Cd concentration in *Adenoscolex* showed positive correlation with pH of water. Zn concentration of water and Fe concentration in *Bothriocephalus acheilognathi* was positively correlated with DO. However, the two parasites which have relation directly with water are *Adenoscolex oreini* and *Pomphorhynchus kashmirensis*. *Adenoscolex oreini* was positively correlated with Cd concentration in water during autumn and winter whereas *Pomphorhynchus kashmirensis* was positively correlated with Al and Cr concentration in water. Therefore, the two parasites seem to act as bio-indicators for the corresponding metals.

Therefore, the following conclusions could be drawn from the present studies.

- All the four physicochemical parameters temperature, dissolved oxygen, pH and free carbon dioxide showed variation vis-a-vis the season and location of the stations in water bodies.
- Both the cestodes and the acanthocephalan parasites were prevalent more

in male fishes compared to females. The mean intensity and abundance of both the cestodes was highest in summer and lowest in winter. The presence of the parasites reduced the condition factor of the infected fishes in both the water bodies. However, the overall condition factor of fish in Jhelum river was higher. A positive correlation existed between the water temperature and pH with the prevalence of all the three parasites while as it was negatively correlated with the DO. With regard to free carbon dioxide, except *Pomphorhynchus kashmirensis*, both the cestodes showed positive correlation.

- Histopathologically, all the three parasites seemed to induce lesions in intestines characterized by various intensities of enteritis coupled with hyperplastic goblet cells with increased acid mucopolysaccharide concentrations. The lesions could have been aggravated by the metal concentration in water under field conditions.
- The overall concentrations of different heavy metals in the two water bodies showed significant variations both season and location wise. In Dal Lake the concentrations of Co, Ni, Mn, Cr, Fe and Cd were higher than the permissible limits as recommended by WHO/FEPA/IAEA-407. Similarly, in River Jhelum the concentrations of Ni, Mn, Cr, Fe, Cd in water were higher than the permissible limits. Pb and Hg were below detection levels in both the water bodies while Cu and Zn were recorded only in Dal Lake and River Jhelum, respectively.
- The two parasites which have direct relation with water are *Adenoscolex oreini* and *Pomphorhynchus kashmirensis*. *Adenoscolex oreini* was positively correlated with Cd concentration in water during autumn and winter whereas *Pomphorhynchus kashmirensis* was positively correlated with Al and Cr concentration in water. Therefore, the two parasites seem to act as bio-indicators for the corresponding metals.

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APPENDICES

Correlation studies:

Table 1: Correlation between the concentrations of heavy metals of Fish tissues and different parasites during summer season of Dal Lake

Water	<i>Adenoscolex</i>	LAP	MAP	<i>Bothriocephalus</i>	LBP	MBP	<i>Pomphorhynchus</i>	LPP	MPP
Copper	-.235	-.930*	.322	-.016	-.822	-.087	-.460	.238	.028
Zinc	-.286	-.027	-.439	-.124	-.679	-.841	-.358	-.721	.129
Cobalt	-.263	.147	.350	-.629	.826	.627	-.550	.009	.088
Nickel	-.599	.142	-.490	-.019	.775	-.252	-.690	.261	-.128
Manganese	-.774	.175	-.070	-.295	-.717	-.280	-.397	.521	-.021
Chromium	-.578	-.229	.402	.369	-.900*	.463	.542	.368	-.001
Aluminum	.174	-.922*	-.534	.408	-.973**	-.751	-.643	-.265	-.736
Iron	-.258	-.378	.104	-.549	-.729	-.082	.321	.633	.137
Calcium	.874	-.193	.551	-.706	-.654	.121	-.403	.572	-.515
Cadmium	-.409	-.657	.027	.319	-.514	-.955*	-.966**	-.080	-.031

Table 2: Correlation between the concentrations of heavy metals of Fish tissues and different parasites during spring season of Dal Lake

water	<i>Adenoscolex</i>	LAP	MAP	<i>Bothriocephalus</i>	LBP	MBP	<i>Pomphorhynchus</i>	LPP	MPP
Copper	-.503	.089	-.279	-.741	.433	-.519	-.897*	.313	-.668
Zinc	.204	-.139	.201	.480	-.047	-.280	.525	-.450	-.493
Cobalt	-.141	.784	.791	-.812	.031	-.387	.381	.453	.220
Nickel	.690	.791	.886*	-.995**	-.496	.292	-.876	.336	-.489
Manganese	-.003	-.890*	-.149	.062	-.753	.320	.848	-.041	-.202
Chromium	.633	-.554	.183	-.172	-.947*	-.848	.758	.672	.415
Aluminum	-.370	-.363	.641	.134	-.804	.640	-.663	-.025	.526
Iron	-.929*	.053	-.396	-.784	-.007	-.209	-.231	-.438	-.024
Calcium	.552	-.123	-.183	-.817	.293	-.823	-.300	.671	-.218
Cadmium	-.381	-.060	-.874	-.525	.298	.762	.276	-.096	.958*

Table 3: Correlation between the concentrations of heavy metals of Fish tissues and different parasites during autumn season of Dal Lake

water	<i>Adenoscolex</i>	LAP	MAP	<i>Bothriocephalus</i>	LBP	MBP	<i>Pomphorhynchus</i>	LPP	MPP
Copper	-.595	-.522	-.798	-.183	.677	-.494	.563	-.034	.141
Zinc	.432	-.947*	-.418	-.387	-.595	-.958*	-.081	-.653	-.351
Cobalt	.313	.835	.670	-.767	-.860	-.171	.864	-.510	.120
Nickel	.675	-.717	.255	.086	-.804	.991**	-.362	-.805	.128
Manganese	-.800	-.009	.634	-.282	-.826	.435	-.128	-.550	-.852
Chromium	.784	.583	.148	.462	.531	.991**	-.366	-.640	.875
Aluminum	-.207	-.679	-.626	.554	-.184	.727	.313	.751	-.536
Iron	-.513	-.557	-.274	.393	.164	.524	.316	-.653	-.524
Calcium	-.253	-.591	-.582	-.211	.976**	.611	-.650	-.949*	-.675
Cadmium	.984**	-.644	-.609	-.385	.145	-.893*	-.535	-.754	.047

Table 4: Correlation between the concentrations of heavy metals of Fish tissues and different parasites during winter season of Dal Lake

water	<i>Adenoscolex</i>	LAP	MAP	<i>Bothriocephalus</i>	LBP	MBP	<i>Pomphorhynchus</i>	LPP	MPP
Copper	-.248	-.253	.864	-.859	-.837	.672	-.125	.274	-.017
Zinc	-.870	.091	.686	.182	-.553	.398	-.693	-.890*	-.661
Cobalt	-.005	.096	.909*	.805	-.090	-.092	.601	-.149	.300
Nickel	.190	.295	-.207	-.278	.156	-.267	.688	-.379	-.853
Manganese	-.377	.373	.648	.648	.837	.071	.119	.977**	-.937*
Chromium	-.462	.283	-.164	.248	.624	-.029	-.404	.021	.042
Aluminum	-.316	.285	.881*	-.127	.211	-.850	-.349	.470	.939*
Iron	.388	-.597	-.882*	.247	.252	.082	.406	-.477	-.321
Calcium	.192	.019	-.554	.253	-.297	.257	-.582	.784	-.496
Cadmium	.929*	-.842	.907*	-.378	.479	.105	-.482	-.138	-.316

Table 5: Correlation between physicochemical parameters and heavy metal concentrations of water during summer season of Dal Lake

	Copper	Zinc	Cobalt	Nickel	Manganese	Chromium	Aluminum	Iron	Calcium	Cadmium
Temperature	-.706	-.665	-.625	.056	.832	-.269	.927*	.716	.957*	.211
Dissolved O ₂	.550	-.402	.615	.286	-.419	.772	-.716	-.953*	-.829	.229
Free CO ₂	-.672	.885*	-.621	-.135	.471	-.726	.721	.914*	.841	-.106
pH	.654	-.570	.405	-.054	-.360	.607	-.569	-.905*	-.723	.153

Table 6: Correlation between physicochemical parameters and heavy metal concentrations of water during spring season of Dal Lake

	Copper	Zinc	Cobalt	Nickel	Manganese	Chromium	Aluminum	Iron	Calcium	Cadmium
Temperature	.340	.619	-.318	.690	.879*	-.479	-.598	.743	.434	.707
Dissolved O ₂	-.558	.218	-.704	.239	-.275	.412	.939*	-.795	.473	-.846
Free CO ₂	.414	.073	-.098	.128	.693	-.046	-.755	.825	.304	.918*
pH	-.933*	.194	-.158	.110	-.625	.806	.367	-.666	.620	-.434

Table 7: Correlation between physicochemical parameters and heavy metal concentrations of water during autumn season of Dal Lake

	Copper	Zinc	Cobalt	Nickel	Manganese	Chromium	Aluminum	Iron	Calcium	Cadmium
Temperature	-.752	.982**	.638	.254	-.578	-.928*	-.815	.952*	-.936*	-.902*
Dissolved O ₂	-.258	.480	.175	-.436	.748	.472	.416	.324	.571	.478
Free CO ₂	.541	.754	-.707	-.477	.135	.856	.869	.856	.878	.911*
pH	-.698	-.680	.441	.275	-.306	-.540	-.368	-.650	-.575	-.658

Table 8: Correlation between physicochemical parameters and heavy metal concentrations of water winter season of Dal Lake

	Copper	Zinc	Cobalt	Nickel	Manganese	Chromium	Aluminum	Iron	Calcium	Cadmium
Temperature	-.136	-.665	-.661	-.630	.972**	-.108	-.363	-.423	.162	.287
Dissolved O ₂	-.403	-.402	-.684	-.404	.852	-.363	-.518	-.398	.004	.037
Free CO ₂	.255	.885*	.345	.435	-.866	.215	.414	.484	.010	-.639
pH	-.618	-.570	-.590	-.366	.832	-.607	-.356	-.701	.094	.286

Table 9: Correlation between the concentrations of heavy metals of Fish tissues and different parasites during summer season of River Jhelum

water	<i>Adenoscolex</i>	LAP	MAP	<i>Bothriocephalus</i>	LBP	MBP	<i>Pomphorhynchus</i>	LPP	MPP
Copper	-.787	.821	-.478	.024	.370	.933*	.797	-.724	-.654
Zinc	-.992**	-.444	-.497	-.141	.247	.008	-.052	-.431	.825
Cobalt	-.509	.385	.749	.709	-.359	-.147	.415	.328	-.214
Nickel	.823	.146	.424	.211	-.452	.102	.806	-.307	-.041
Manganese	.558	-.319	.300	-.504	-.740	-.569	-.357	.939*	-.630
Chromium	-.215	.057	.957*	-.160	-.470	-.871	-.263	-.647	-.714
Aluminum	.220	-.906*	.067	.518	-.872	.719	.159	-.413	.455
Iron	.550	.163	.584	-.114	-.366	.607	.399	-.402	.630
Calcium	.229	.746	.141	-.115	.402	.280	-.397	-.425	-.760
Cadmium	-.486	.126	-.165	.631	-.444	.070	-.660	.068	.216

Table 10: Correlation between the concentrations of heavy metals of Fish tissues and different parasites during spring season of River Jhelum

water	<i>Adenoscolex</i>	LAP	MAP	<i>Bothriocephalus</i>	LBP	MBP	<i>Pomphorhynchus</i>	LPP	MPP
Copper	.167	-.571	-.365	-.606	-.446	.408	.741	-.245	-.005
Zinc	-.344	-.087	-.248	.856	-.033	.924*	-.238	-.957*	-.598
Cobalt	.244	.213	.720	-.183	.968**	.875	-.508	-.243	.258
Nickel	-.009	-.691	.254	-.249	.286	.542	.692	-.384	-.450
Manganese	-.406	-.096	.832	.764	-.265	.219	-.468	-.010	.633
Chromium	.519	.565	.404	-.195	.600	-.599	.598	-.243	.258
Aluminum	.220	-.466	-.983**	.139	-.247	-.935*	.013	.073	.065
Iron	-.166	-.761	.252	-.427	-.742	-.080	.302	.774	-.725
Calcium	-.249	.611	-.780	.319	-.245	.888*	.851	.752	-.504
Cadmium	-.160	.042	.099	-.091	.009	.057	.423	.988**	.010

Table 11: Correlation between the concentrations of heavy metals of Fish tissues and different parasites during autumn season of River Jhelum

water	<i>Adenoscolex</i>	LAP	MAP	<i>Bothriocephalus</i>	LBP	MBP	<i>Pomphorhynchus</i>	LPP	MPP
Copper	-.037	-.280	.163	-.323	-.461	-.274	-.886*	-.435	.365
Zinc	.499	.146	.521	.168	.732	.139	-.287	-.813	.852
Cobalt	.596	-.670	-.054	-.076	.136	-.109	.876	-.392	-.070
Nickel	-.561	-.295	.452	-.817	.547	-.387	-.243	-.495	.695
Manganese	.220	-.683	.178	-.800	-.710	.986**	-.267	.796	.530
Chromium	-.320	.268	-.044	-.427	-.533	-.567	.539	.258	-.100
Aluminum	.225	-.571	-.743	.204	.100	-.756	.933*	-.961**	.550
Iron	-.842	.289	-.155	.258	.878	-.066	-.276	-.374	-.422
Calcium	.282	.283	-.688	-.108	-.974**	-.834	.036	.296	-.508
Cadmium	-.777	-.113	-.853	.444	-.565	-.436	.603	-.262	.059

Table 12: Correlation between the concentrations of heavy metals of Fish tissues and different parasites during winter season of River Jhelum

water	<i>Adenoscolex</i>	LAP	MAP	<i>Bothriocephalus</i>	LBP	MBP	<i>Pomphorhynchus</i>	LPP	MPP
Copper	-.399	-.291	-.504	.284	.345	-.158	-.316	-.467	-.501
Zinc	-.437	-.060	.355	.337	.077	-.210	.339	.049	-.386
Cobalt	.620	.686	.337	.269	-.621	-.673	.485	.240	-.262
Nickel	-.718	.661	-.482	.173	.483	.056	-.427	-.317	.688
Manganese	-.135	.512	.572	.151	.731	.852	-.410	-.552	.832
Chromium	-.582	-.404	-.134	-.312	-.404	.477	.908*	.699	.549
Aluminum	-.240	.752	.646	-.285	.936*	-.251	.227	.462	-.318
Iron	.214	.029	-.258	-.136	.473	.352	-.672	.061	-.735
Calcium	-.124	.237	-.654	-.459	.270	.240	.568	-.337	.669
Cadmium	.533	-.090	-.075	-.783	-.513	-.513	-.746	.047	.898*

Table 13: Correlation between physicochemical parameters and heavy metal concentrations of water during summer season of River Jhelum

	Copper	Zinc	Cobalt	Nickel	Manganese	Chromium	Aluminum	Iron	Calcium	Cadmium
Temperature	.596	-.816	.605	-.496	.940*	.982**	.801	.025	-.605	-.213
Dissolved O ₂	-.797	.924*	-.358	.456	-.842	-.818	-.816	-.473	.160	-.303
Free CO ₂	.939*	-.046	.047	-.159	.919*	.852	.960**	.599	.033	.437
pH	.099	-.298	.409	-.295	.484	.519	.299	-.356	-.547	-.428

Table 14: Correlation between physicochemical parameters and heavy metal concentrations of water during spring season of River Jhelum

	Copper	Zinc	Cobalt	Nickel	Manganese	Chromium	Aluminum	Iron	Calcium	Cadmium
Temperature	.730	-.946*	-.129	.963**	.457	.496	.817	.771	-.865	.559
Dissolved O ₂	-.928*	.846	.048	-.788	-.133	-.692	-.907*	-.849	.826	-.368
Free CO ₂	-.151	-.902*	-.134	.622	.781	-.226	.071	.072	-.354	.569
pH	.403	-.489	.515	-.301	-.216	.451	.217	.149	-.176	.061

Table 15: Correlation between physicochemical parameters and heavy metal concentrations of water during autumn season of River Jhelum

	Copper	Zinc	Cobalt	Nickel	Manganese	Chromium	Aluminum	Iron	Calcium	Cadmium
Temperature	-.719	.972**	-.835	-.791	-.978**	-.625	-.945*	.021	-.942*	-.336
Dissolved O ₂	-.772	-.948*	-.897*	-.801	-.971**	-.580	-.937*	.079	-.885*	-.390
Free CO ₂	.056	-.492	-.054	-.444	-.425	-.637	-.334	-.455	-.469	.362
pH	-.458	-.642	-.257	-.031	-.572	-.009	-.569	.350	-.677	-.333

Table 16: Correlation between physicochemical parameters and heavy metal concentrations of water winter season of River Jhelum

	Copper	Zinc	Cobalt	Nickel	Manganese	Chromium	Aluminum	Iron	Calcium	Cadmium
Temperature	.400	-.521	-.319	-.608	.918*	.384	-.783	.005	-.921*	.234
Dissolved O ₂	.482	.096	-.833	-.739	.824	-.252	-.857	-.296	-.854	-.164
Free CO ₂	-.306	-.207	.886*	.570	-.749	.237	.747	.494	.763	.365
pH	-.078	.155	-.664	-.474	.461	.481	-.768	-.015	-.642	-.067

Table 17: Correlation between the physicochemical parameters and Iron concentrations of Fish tissues and different parasites during autumn season of Dal Lake

Water	<i>Adenoscolex</i>	LAP	MAP	<i>Bothriocephalus</i>	LBP	MBP	<i>Pomphorhynchus</i>	LPP	MPP
Temperature	.293	.687	.083	-.638	-.359	-.574	-.122	.757	.430
Dissolved O ₂	.009	-.195	.617	-.049	-.161	-.538	.333	.257	-.815
Free CO ₂	-.352	-.062	-.125	.111	-.359	.170	.757	-.457	-.460
pH	-.094	.586	.069	-.923*	-.533	-.800	.174	.976**	-.175

Table 18: Correlation between the physicochemical parameters and Iron concentrations of Fish tissues and different parasites during winter season of Dal Lake

Water	<i>Adenoscolex</i>	LAP	MAP	<i>Bothriocephalus</i>	LBP	MBP	<i>Pomphorhynchus</i>	LPP	MPP
Temperature	.559	.573	-.043	-.059	-.487	.611	-.436	.980**	.035
Dissolved O ₂	.482	.817	.003	-.442	-.245	.702	-.703	.967**	.408
Free CO ₂	-.611	-.439	-.083	-.014	.785	-.245	.156	-.847	.126
pH	.280	.854	.372	-.468	-.397	.396	-.628	.904*	.423

Table 19: Correlation between the physicochemical parameters and Iron concentrations of Fish tissues and different parasites during spring season of Dal Lake

Water	<i>Adenoscolex</i>	LAP	MAP	<i>Bothriocephalus</i>	LBP	MBP	<i>Pomphorhynchus</i>	LPP	MPP
Temperature	-.686	.662	-.099	-.723	-.077	.395	.233	-.179	.577
Dissolved O ₂	.584	.355	.053	.306	.078	.369	.509	.826	.203
Free CO ₂	-.805	.038	-.340	-.802	.461	.243	-.469	-.443	.058
pH	.694	-.028	.538	.517	.426	.717	-.192	-.079	.260

Table 20: Correlation between the physicochemical parameters and Iron concentrations of Fish tissues and different parasites during summer season of Dal Lake

Water	<i>Adenoscolex</i>	LAP	MAP	<i>Bothriocephalus</i>	LBP	MBP	<i>Pomphorhynchus</i>	LPP	MPP
Temperature	-.240	-.844	-.075	-.626	-.700	-.727	.144	.988**	-.182
Dissolved O ₂	.505	.589	.002	.767	.871	.272	-.227	-.720	.129
Free CO ₂	-.506	-.632	-.181	-.765	-.826	-.297	.342	.716	-.114
pH	.303	.486	.324	.574	.641	.140	-.580	-.613	-.141

Table 21: Correlation between the physicochemical parameters and Iron concentrations of Fish tissues and different parasites during autumn season of River Jhelum

Water	<i>Adenoscolex</i>	LAP	MAP	<i>Bothriocephalus</i>	LBP	MBP	<i>Pomphorhynchus</i>	LPP	MPP
Temperature	.296	-.398	.977**	.854	.141	.586	-.200	-.460	-.388
Dissolved O ₂	.136	-.455	.912*	.950*	.292	.428	-.119	-.296	-.237
Free CO ₂	.862	.077	.662	.052	-.646	.544	.022	-.401	-.305
pH	.208	.289	.533	.204	.029	.587	-.308	-.802	-.749

Table 22: Correlation between the physicochemical parameters and Iron concentrations of Fish tissues and different parasites during winter season of River Jhelum

Water	<i>Adenoscolex</i>	LAP	MAP	<i>Bothriocephalus</i>	LBP	MBP	<i>Pomphorhynchus</i>	LPP	MPP
Temperature	.228	-.921*	.703	-.682	.518	-.441	-.155	-.233	-.188
Dissolved O ₂	.612	-.624	.951*	-.854	.110	.119	-.251	-.432	.504
Free CO ₂	-.519	.578	-.939*	.728	-.046	.012	.041	.446	-.593
pH	.733	-.295	.827	-.367	.780	-.180	.136	-.898*	-.055

Table 23: Correlation between the physicochemical parameters and Iron concentrations of Fish tissues and different parasites during spring season of River Jhelum

Water	<i>Adenoscolex</i>	LAP	MAP	<i>Bothriocephalus</i>	LBP	MBP	<i>Pomphorhynchus</i>	LPP	MPP
Temperature	.185	-.572	.775	-.247	-.876	.449	.002	.877	-.508
Dissolved O ₂	-.181	.392	-.543	.042	.863	-.005	-.379	-.920*	.332
Free CO ₂	.116	-.489	.652	-.475	-.258	.959*	-.673	.157	-.457
pH	.352	.242	-.701	-.100	.363	-.856	.951*	-.231	.278

Table 24: Correlation between the physicochemical parameters and Iron concentrations of Fish tissues and different parasites during summer season of River Jhelum

Water	<i>Adenoscolex</i>	LAP	MAP	<i>Bothriocephalus</i>	LBP	MBP	<i>Pomphorhynchus</i>	LPP	MPP
Temperature	-.399	.223	.762	-.922*	-.463	-.746	-.175	-.912*	-.439
Dissolved O ₂	.272	.238	-.917*	.907*	.908*	.154	-.154	.923*	.046
Free CO ₂	.249	.391	.929*	-.693	-.419	-.245	.287	-.872	-.112
pH	.053	.597	.209	-.298	.342	-.817	-.047	-.288	-.715

Table 25: Correlation between the physicochemical parameters and Calcium concentrations of Fish tissues and different parasites during autumn season of Dal Lake

Water	<i>Adenoscolex</i>	LAP	MAP	<i>Bothriocephalus</i>	LBP	MBP	<i>Pomphorhynchus</i>	LPP	MPP
Temperature	-.054	.710	.362	-.034	-.976**	-.677	.356	.831	.891*
Dissolved O ₂	-.435	.135	-.234	-.704	.471	.138	-.678	-.573	.029
Free CO ₂	-.454	-.630	-.842	-.300	.791	.598	-.828	-.873	-.417
pH	-.594	.882*	-.061	-.214	-.654	-.811	-.084	.352	.931*

Table 26: Correlation between the physicochemical parameters and Calcium concentrations of Fish tissues and different parasites during winter season of Dal Lake

Water	<i>Adenoscolex</i>	LAP	MAP	<i>Bothriocephalus</i>	LBP	MBP	<i>Pomphorhynchus</i>	LPP	MPP
Temperature	.792	.158	.214	.480	.661	-.787	.029	-.163	-.016
Dissolved O ₂	.746	.109	.122	.093	.852	-.854	-.166	-.387	.248
Free CO ₂	-.881*	-.471	-.185	-.606	-.679	.603	-.374	.074	-.173
pH	.939*	.490	-.189	.130	.901*	-.587	-.084	-.105	.456

Table 27: Correlation between the physicochemical parameters and Calcium concentrations of Fish tissues and different parasites during spring season of Dal Lake

Water	<i>Adenoscolex</i>	LAP	MAP	<i>Bothriocephalus</i>	LBP	MBP	<i>Pomphorhynchus</i>	LPP	MPP
Temperature	.058	.743	-.503	-.448	.933*	-.059	-.746	-.183	-.803
Dissolved O ₂	.664	-.872	.572	-.547	-.607	-.866	.689	.457	.736
Free CO ₂	-.474	.799	-.430	-.325	.881*	.271	-.859	-.204	-.888*
pH	.485	-.500	-.157	-.212	-.292	-.618	.009	.961**	.171

Table 28: Correlation between the physicochemical parameters and Calcium concentrations of Fish tissues and different parasites during summer season of Dal Lake

Water	<i>Adenoscolex</i>	LAP	MAP	<i>Bothriocephalus</i>	LBP	MBP	<i>Pomphorhynchus</i>	LPP	MPP
Temperature	.863	-.386	.396	-.529	-.673	-.043	-.556	.336	-.740
Dissolved O ₂	-.808	.113	-.645	.759	.463	-.305	.061	-.596	.363
Free CO ₂	.731	-.217	.541	-.697	-.598	.175	.020	.558	-.349
pH	-.571	.424	-.315	.483	.670	.038	-.114	-.332	.411

Table 29: Correlation between the physicochemical parameters and Calcium concentrations of Fish tissues and different parasites during autumn season of River Jhelum

Water	<i>Adenoscolex</i>	LAP	MAP	<i>Bothriocephalus</i>	LBP	MBP	<i>Pomphorhynchus</i>	LPP	MPP
Temperature	-.457	-.325	.836	.201	.863	.870	-.083	-.247	.374
Dissolved O ₂	-.528	-.547	.730	.139	.819	.946*	.065	-.033	.518
Free CO ₂	-.180	.564	.863	.381	.329	.114	-.566	-.759	-.517
pH	-.276	.292	.633	-.247	.711	.174	.021	-.877	-.052

Table 30: Correlation between the physicochemical parameters and Calcium concentrations of Fish tissues and different parasites during winter season of River Jhelum

Water	<i>Adenoscolex</i>	LAP	MAP	<i>Bothriocephalus</i>	LBP	MBP	<i>Pomphorhynchus</i>	LPP	MPP
Temperature	-.032	.074	.700	.450	-.603	-.156	-.377	.391	-.525
Dissolved O ₂	-.231	-.157	.654	.423	.123	-.265	-.531	.343	-.426
Free CO ₂	.443	-.035	-.682	-.408	-.095	.233	.421	-.374	.264
pH	-.155	.309	.978**	.965**	-.134	-.840	-.832	-.360	-.716

Table 31: Correlation between the physicochemical parameters and Calcium concentrations of Fish tissues and different parasites during spring season of River Jhelum

Water	<i>Adenoscolex</i>	LAP	MAP	<i>Bothriocephalus</i>	LBP	MBP	<i>Pomphorhynchus</i>	LPP	MPP
Temperature	.195	-.436	.734	-.181	.130	-.937*	-.633	-.745	.387
Dissolved O ₂	-.411	.398	-.860	.565	-.448	.711	.674	.902*	-.006
Free CO ₂	-.366	-.203	.003	.650	-.570	-.711	-.127	.081	.854
pH	.395	-.251	.227	-.639	.588	.288	-.398	-.267	-.288

Table 32: Correlation between the physicochemical parameters and Calcium concentrations of Fish tissues and different parasites during summer season of River Jhelum

Water	<i>Adenoscolex</i>	LAP	MAP	<i>Bothriocephalus</i>	LBP	MBP	<i>Pomphorhynchus</i>	LPP	MPP
Temperature	-.268	-.976**	-.784	-.539	-.527	.197	.369	.638	.822
Dissolved O ₂	.087	.621	.770	.892*	.811	-.140	.317	-.027	-.316
Free CO ₂	.374	-.601	-.740	-.785	-.320	.310	.246	.472	.364
pH	-.622	-.624	-.143	.033	.124	-.070	.924*	.826	.638

Table 33: Correlation between the physicochemical parameters and Zinc concentrations of Fish tissues and different parasites during autumn season of Dal Lake

Water	<i>Adenoscolex</i>	LAP	MAP	<i>Bothriocephalus</i>	LBP	MBP	<i>Pomphorhynchus</i>	LPP	MPP
Temperature	-.316	.898*	.568	.471	.668	.929*	.249	.759	.270
Dissolved O ₂	.978**	-.587	.236	-.221	-.358	-.570	.665	.080	-.496
Free CO ₂	.255	-.899*	-.462	-.598	-.318	-.561	-.260	-.741	.267
pH	.073	.485	.861	.572	.779	.644	.653	.842	.068

Table 34: Correlation between the physicochemical parameters and Zinc concentrations of Fish tissues and different parasites during winter season of Dal Lake

Water	<i>Adenoscolex</i>	LAP	MAP	<i>Bothriocephalus</i>	LBP	MBP	<i>Pomphorhynchus</i>	LPP	MPP
Temperature	.721	.278	-.910*	-.067	.737	-.266	.367	.642	.681
Dissolved O ₂	.548	.518	-.728	.059	.773	-.112	.212	.409	.354
Free CO ₂	-.842	-.249	.809	.153	-.736	.268	-.627	-.842	-.627
pH	.777	.318	-.736	.289	.952*	-.375	.208	.426	.248

Table 35: Correlation between the physicochemical parameters and Zinc concentrations of Fish tissues and different parasites during spring season of Dal Lake

Water	<i>Adenoscolex</i>	LAP	MAP	<i>Bothriocephalus</i>	LBP	MBP	<i>Pomphorhynchus</i>	LPP	MPP
Temperature	-.114	-.476	.653	-.019	.416	.297	-.027	-.830	-.389
Dissolved O ₂	.102	.051	-.823	.144	-.919*	-.587	-.027	.390	-.322
Free CO ₂	.148	-.076	.537	.019	.751	.242	-.513	-.444	-.419
pH	.926*	.877	-.696	.885*	-.116	-.984**	-.094	.752	-.694

Table 36: Correlation between the physicochemical parameters and Zinc concentrations of Fish tissues and different parasites during summer season of Dal Lake

Water	<i>Adenoscolex</i>	LAP	MAP	<i>Bothriocephalus</i>	LBP	MBP	<i>Pomphorhynchus</i>	LPP	MPP
Temperature	.091	-.545	-.443	.345	-.281	-.843	-.116	-.608	.557
Dissolved O ₂	-.601	.258	.855	-.423	-.200	.813	.569	.749	-.721
Free CO ₂	.595	-.290	-.796	-.882*	.184	.505	-.612	-.836	.604
pH	-.714	.455	.656	-.720	-.362	.817	.523	.774	-.570

Table 37: Correlation between the physicochemical parameters and Zinc concentrations of Fish tissues and different parasites during autumn season of River Jhelum

Water	<i>Adenoscolex</i>	LAP	MAP	<i>Bothriocephalus</i>	LBP	MBP	<i>Pomphorhynchus</i>	LPP	MPP
Temperature	-.639	-.007	-.657	.023	-.796	-.290	.364	.683	-.798
Dissolved O ₂	-.743	.089	-.730	.004	-.618	-.308	.521	.760	-.720
Free CO ₂	-.116	-.449	-.306	-.062	-.908*	.115	-.437	-.043	-.372
pH	.274	-.523	.198	-.314	-.760	.177	-.368	.453	-.798

Table 38: Correlation between the physicochemical parameters and Zinc concentrations of Fish tissues and different parasites during winter season of River Jhelum

Water	<i>Adenoscolex</i>	LAP	MAP	<i>Bothriocephalus</i>	LBP	MBP	<i>Pomphorhynchus</i>	LPP	MPP
Temperature	.173	.834	.511	.311	.155	-.121	-.970**	-.065	.019
Dissolved O ₂	-.528	.961**	.897*	.372	.362	.193	-.757	.055	.041
Free CO ₂	.462	-.918*	-.866	-.350	-.484	-.038	.722	.118	.181
pH	.067	.761	.806	.921*	-.296	-.438	-.801	.367	-.056

Table 39: Correlation between the physicochemical parameters and Zinc concentrations of Fish tissues and different parasites during spring season of River Jhelum

Water	<i>Adenoscolex</i>	LAP	MAP	<i>Bothriocephalus</i>	LBP	MBP	<i>Pomphorhynchus</i>	LPP	MPP
Temperature	.306	.169	.302	-.654	.306	-.967**	.094	.984**	.536
Dissolved O ₂	-.044	.442	.151	.805	.221	.833	-.571	-.835	-.182
Free CO ₂	.247	.224	.122	-.886*	-.317	-.737	.079	.776	.791
pH	.665	.828	.803	-.282	.550	-.375	-.386	.439	.722

Table 40: Correlation between the physicochemical parameters and Zinc concentrations of Fish tissues and different parasites during summer season of River Jhelum

Water	<i>Adenoscolex</i>	LAP	MAP	<i>Bothriocephalus</i>	LBP	MBP	<i>Pomphorhynchus</i>	LPP	MPP
Temperature	.740	-.039	.772	.345	-.510	.455	-.187	.810	-.827
Dissolved O ₂	-.897*	-.418	-.479	.042	.590	-.109	-.044	-.552	.968**
Free CO ₂	-.066	-.858	.802	.822	.028	.808	-.437	.742	.033
pH	.415	.838	-.281	-.495	.381	-.503	.728	-.407	-.005

Table 41: Correlation between the physicochemical parameters and Cadmium concentrations of Fish tissues and different parasites during autumn season of Dal Lake

Water	<i>Adenoscolex</i>	LAP	MAP	<i>Bothriocephalus</i>	LBP	MBP	<i>Pomphorhynchus</i>	LPP	MPP
Temperature	.324	.626	.608	-.103	.308	-.817	-.510	-.572	.570
Dissolved O ₂	.631	-.195	-.358	.002	.500	.482	-.530	.081	.226
Free CO ₂	-.410	-.944*	-.184	.190	-.489	.918*	.060	.938*	-.142
pH	.614	.463	.267	.294	.580	-.596	-.690	-.358	.492

Table 42: Correlation between the physicochemical parameters and Cadmium concentrations of Fish tissues and different parasites during winter season of Dal Lake

Water	<i>Adenoscolex</i>	LAP	MAP	<i>Bothriocephalus</i>	LBP	MBP	<i>Pomphorhynchus</i>	LPP	MPP
Temperature	-.309	.779	-.301	.853	-.202	-.052	.086	-.191	-.418
Dissolved O ₂	-.247	.543	-.480	.595	-.323	.014	-.069	-.369	-.573
Free CO ₂	-.069	-.670	.170	-.930*	.439	-.122	-.480	-.133	.554
pH	.013	.567	-.162	.657	-.638	-.113	.140	.021	-.545

Table 43: Correlation between the physicochemical parameters and Cadmium concentrations of Fish tissues and different parasites during spring season of Dal Lake

Water	<i>Adenoscolex</i>	LAP	MAP	<i>Bothriocephalus</i>	LBP	MBP	<i>Pomphorhynchus</i>	LPP	MPP
Temperature	-.486	-.175	-.603	.363	.782	.100	-.169	.768	-.094
Dissolved O ₂	-.566	-.353	.452	.411	-.679	.832	-.876	.270	.571
Free CO ₂	-.003	.069	-.893*	-.152	.973**	-.070	.229	.544	-.729
pH	.249	.613	-.273	.434	-.392	.536	-.372	.044	-.153

Table 44: Correlation between the physicochemical parameters and Cadmium concentrations of Fish tissues and different parasites during summer season of Dal Lake

Water	<i>Adenoscolex</i>	LAP	MAP	<i>Bothriocephalus</i>	LBP	MBP	<i>Pomphorhynchus</i>	LPP	MPP
Temperature	.306	-.808	-.729	.566	-.900*	-.842	-.582	-.544	-.852
Dissolved O ₂	-.760	.655	.426	-.711	.559	.561	-.021	.752	.357
Free CO ₂	.802	-.722	-.345	.813	-.556	-.479	.020	-.700	-.367
pH	-.881*	.603	.321	-.920*	.400	.377	-.130	.776	.344

Table 45: Correlation between the physicochemical parameters and Cadmium concentrations of Fish tissues and different parasites during autumn season of River Jhelum

Water	<i>Adenoscolex</i>	LAP	MAP	<i>Bothriocephalus</i>	LBP	MBP	<i>Pomphorhynchus</i>	LPP	MPP
Temperature	-.082	.642	.492	-.181	.177	.779	-.879*	.952*	-.748
Dissolved O ₂	-.177	.440	.561	-.421	-.012	.646	-.855	.882*	-.751
Free CO ₂	.308	.810	-.269	.525	.891*	.696	-.377	.536	-.583
pH	-.272	.732	.395	.607	.437	.941*	-.729	.660	-.328

Table 46: Correlation between the physicochemical parameters and Cadmium concentrations of Fish tissues and different parasites during winter season of River Jhelum

Water	<i>Adenoscolex</i>	LAP	MAP	<i>Bothriocephalus</i>	LBP	MBP	<i>Pomphorhynchus</i>	LPP	MPP
Temperature	-.373	-.261	-.368	.704	-.816	.540	.103	-.330	.293
Dissolved O ₂	-.004	-.472	-.336	.145	-.928*	.245	-.461	-.793	.311
Free CO ₂	.156	.346	.118	-.204	.895*	-.364	.387	.784	-.168
pH	.093	-.738	-.131	.602	-.850	.725	.313	-.024	-.355

Table 47: Correlation between the physicochemical parameters and Cadmium concentrations of Fish tissues and different parasites during spring season of River Jehlum

Water	<i>Adenoscolex</i>	LAP	MAP	<i>Bothriocephalus</i>	LBP	MBP	<i>Pomphorhynchus</i>	LPP	MPP
Temperature	.609	-.694	-.811	.048	.345	-.929*	.193	-.401	.475
Dissolved O ₂	-.270	.327	.939*	.213	-.025	.941*	-.127	.257	-.238
Free CO ₂	.782	-.889*	.007	.523	.690	-.264	.237	-.416	.625
pH	-.903*	.541	-.174	.011	-.866	-.113	.307	.248	-.177

Table 48: Correlation between the physicochemical parameters and Cadmium concentrations of Fish tissues and different parasites during summer season of River Jehlum

Water	<i>Adenoscolex</i>	LAP	MAP	<i>Bothriocephalus</i>	LBP	MBP	<i>Pomphorhynchus</i>	LPP	MPP
Temperature	-.392	-.919*	.055	.681	-.991**	.660	-.345	-.592	-.163
Dissolved O ₂	.066	.806	-.607	-.434	.771	-.925*	-.350	.767	-.210
Free CO ₂	.224	-.786	.015	.302	-.825	.535	.135	-.446	.435
pH	-.395	-.331	-.569	.199	-.554	-.218	-.776	-.116	-.374

Table 49: Correlation between the physicochemical parameters and Aluminum concentrations of Fish tissues and different parasites during autumn season of Dal Lake

Water	<i>Adenoscolex</i>	LAP	MAP	<i>Bothriocephalus</i>	LBP	MBP	<i>Pomphorhynchus</i>	LPP	MPP
Temperature	-.948*	.740	.877	.174	.286	.943*	.421	.681	-.191
Dissolved O ₂	.445	-.255	-.325	.347	.098	-.150	.425	-.861	-.003
Free CO ₂	.831	-.277	-.415	-.647	.389	-.705	-.557	-.737	-.355
pH	-.759	.870	.641	.169	.348	.923*	.659	.137	-.565

Table 50: Correlation between the physicochemical parameters and Aluminum concentrations of Fish tissues and different parasites during winter season of Dal Lake

Water	<i>Adenoscolex</i>	LAP	MAP	<i>Bothriocephalus</i>	LBP	MBP	<i>Pomphorhynchus</i>	LPP	MPP
Temperature	.183	-.460	.197	-.728	.795	-.346	-.748	-.674	.721
Dissolved O ₂	.044	-.088	.071	-.502	.562	-.507	-.523	-.807	.902*
Free CO ₂	-.553	.660	-.556	.677	-.782	.300	.746	.664	-.474
pH	.381	-.203	.419	-.615	.672	-.250	-.668	-.671	.814

Table 51: Correlation between the physicochemical parameters and Aluminum concentrations of Fish tissues and different parasites during spring season of Dal Lake

Water	<i>Adenoscolex</i>	LAP	MAP	<i>Bothriocephalus</i>	LBP	MBP	<i>Pomphorhynchus</i>	LPP	MPP
Temperature	-.676	.291	-.933*	-.047	.826	.159	-.186	-.542	.797
Dissolved O ₂	-.026	.446	.506	.417	.183	-.907*	-.444	-.225	-.727
Free CO ₂	-.435	.314	-.936*	-.533	.531	.523	.315	-.097	.881*
pH	.646	.730	.261	.446	.166	-.639	.583	.657	-.604

Table 52: Correlation between the physicochemical parameters and Aluminum concentrations of Fish tissues and different parasites during summer season of Dal Lake

Water	<i>Adenoscolex</i>	LAP	MAP	<i>Bothriocephalus</i>	LBP	MBP	<i>Pomphorhynchus</i>	LPP	MPP
Temperature	-.589	.296	-.924*	-.366	-.107	-.423	-.197	-.647	-.504
Dissolved O ₂	.577	-.795	.665	.531	-.221	-.021	-.254	.880*	.787
Free CO ₂	-.712	.767	-.621	-.400	.078	-.071	.110	-.902*	-.783
pH	.791	-.811	.577	.166	.126	-.039	-.085	.779	.612

Table 53: Correlation between the physicochemical parameters and Aluminum concentrations of Fish tissues and different parasites during autumn season of River Jhelum

Water	<i>Adenoscolex</i>	LAP	MAP	<i>Bothriocephalus</i>	LBP	MBP	<i>Pomphorhynchus</i>	LPP	MPP
Temperature	-.298	.058	.399	-.883*	-.487	.528	-.586	.046	-.074
Dissolved O ₂	-.177	-.022	.350	-.971**	-.450	.623	-.429	-.133	-.052
Free CO ₂	-.813	.256	-.043	-.158	-.612	-.149	-.478	.385	-.156
pH	-.118	.673	.450	-.246	.102	-.224	-.891*	.852	-.514

Table 54: Correlation between the physicochemical parameters and Aluminum concentrations of Fish tissues and different parasites during winter season of River Jhelum

Water	<i>Adenoscolex</i>	LAP	MAP	<i>Bothriocephalus</i>	LBP	MBP	<i>Pomphorhynchus</i>	LPP	MPP
Temperature	.532	.693	-.802	.391	.386	.386	-.768	.339	-.007
Dissolved O ₂	-.076	.855	-.285	.486	.137	.137	-.188	.583	-.107
Free CO ₂	.140	-.841	.330	-.660	-.292	-.292	.068	-.534	.325
pH	.568	.213	-.219	.523	.602	.602	-.231	-.251	-.020

Table 55: Correlation between the physicochemical parameters and Aluminum concentrations of Fish tissues and different parasites during spring season of River Jhelum

Water	<i>Adenoscolex</i>	LAP	MAP	<i>Bothriocephalus</i>	LBP	MBP	<i>Pomphorhynchus</i>	LPP	MPP
Temperature	.634	.338	-.550	.621	-.559	-.412	.743	.632	-.779
Dissolved O ₂	-.859	-.073	.498	-.840	.361	.066	-.922*	-.403	.634
Free CO ₂	-.261	.570	-.230	-.246	-.505	-.733	-.103	.657	-.462
pH	.084	-.892*	.159	.238	.421	.819	.436	-.082	.628

Table 56: Correlation between the physicochemical parameters and Aluminum concentrations of Fish tissues and different parasites during summer season of River Jhelum

Water	<i>Adenoscolex</i>	LAP	MAP	<i>Bothriocephalus</i>	LBP	MBP	<i>Pomphorhynchus</i>	LPP	MPP
Temperature	.951*	-.412	-.767	.526	-.526	-.302	-.104	.906*	-.566
Dissolved O ₂	-.576	.177	.864	-.923*	.754	-.234	.698	-.950*	.034
Free CO ₂	.540	-.288	-.687	.780	-.732	-.437	-.340	.766	-.507
pH	.665	-.135	-.005	-.144	-.180	-.771	.681	.210	-.708

Table 57: Correlation between the physicochemical parameters and Chromium concentrations of Fish tissues and different parasites during autumn season of Dal Lake

Water	<i>Adenoscolex</i>	LAP	MAP	<i>Bothriocephalus</i>	LBP	MBP	<i>Pomphorhynchus</i>	LPP	MPP
Temperature	-.630	-.324	.154	-.626	-.598	-.919*	.036	.531	-.772
Dissolved O ₂	.721	-.004	.017	.553	.858	.528	-.178	.157	.039
Free CO ₂	.475	.485	.532	.505	.392	.899*	-.650	-.381	.802
pH	-.092	.073	.429	-.603	-.347	-.530	-.366	.292	-.492

Table 58: Correlation between the physicochemical parameters and Chromium concentrations of Fish tissues and different parasites during winter season of Dal Lake

Water	<i>Adenoscolex</i>	LAP	MAP	<i>Bothriocephalus</i>	LBP	MBP	<i>Pomphorhynchus</i>	LPP	MPP
Temperature	-.454	-.424	.330	-.320	.615	.938*	.495	.503	-.021
Dissolved O ₂	-.333	-.635	.125	-.288	.274	.838	.737	.235	.003
Free CO ₂	.118	.398	-.684	.143	-.616	-.937*	-.181	-.787	-.202
pH	-.145	-.450	.341	-.422	.169	.751	.610	.385	-.078

Table 59: Correlation between the physicochemical parameters and Chromium concentrations of Fish tissues and different parasites during spring season of Dal Lake

Water	<i>Adenoscolex</i>	LAP	MAP	<i>Bothriocephalus</i>	LBP	MBP	<i>Pomphorhynchus</i>	LPP	MPP
Temperature	-.547	.728	-.760	-.736	.240	.543	-.226	-.692	-.372
Dissolved O ₂	-.211	-.872	.717	-.044	-.536	.078	.220	.585	-.587
Free CO ₂	.131	.816	-.916*	-.641	-.066	-.112	.005	-.410	.239
pH	.156	-.570	.084	-.387	-.777	-.551	.970**	.242	.295

Table 60: Correlation between the physicochemical parameters and Chromium concentrations of Fish tissues and different parasites during summer season of Dal Lake

Water	<i>Adenoscolex</i>	LAP	MAP	<i>Bothriocephalus</i>	LBP	MBP	<i>Pomphorhynchus</i>	LPP	MPP
Temperature	-.589	.282	.567	.642	.106	-.977**	-.675	-.977**	-.632
Dissolved O ₂	.072	-.524	.024	-.057	-.560	.883*	.745	.827	.476
Free CO ₂	-.130	.566	-.006	.057	.565	-.870	-.836	-.795	-.613
pH	.234	-.761	.125	.058	-.426	.776	.781	.707	.712

Table 61: Correlation between the physicochemical parameters and Chromium concentrations of Fish tissues and different parasites during autumn season of River Jhelum

Water	<i>Adenoscolex</i>	LAP	MAP	<i>Bothriocephalus</i>	LBP	MBP	<i>Pomphorhynchus</i>	LPP	MPP
Temperature	-.142	-.164	-.325	.853	.545	.878	-.312	.111	-.482
Dissolved O ₂	-.245	-.367	-.506	.714	.450	.894*	-.158	.070	-.560
Free CO ₂	.131	.272	.265	.800	.357	.507	-.484	-.219	-.064
pH	-.161	.529	.109	.880*	.281	.412	-.243	.259	-.368

Table 62: Correlation between the physicochemical parameters and Chromium concentrations of Fish tissues and different parasites during winter season of River Jhelum

Water	<i>Adenoscolex</i>	LAP	MAP	<i>Bothriocephalus</i>	LBP	MBP	<i>Pomphorhynchus</i>	LPP	MPP
Temperature	-.898*	-.802	.218	-.457	-.134	.672	.731	.166	.555
Dissolved O ₂	-.327	-.549	.612	.162	.454	.523	.155	.048	.376
Free CO ₂	.258	.401	-.498	-.204	-.363	-.644	-.156	-.093	-.517
pH	-.409	-.682	.670	.309	.408	.726	.683	.792	.687

Table 63: Correlation between the physicochemical parameters and Chromium concentrations of Fish tissues and different parasites during spring season of River Jhelum

Water	<i>Adenoscolex</i>	LAP	MAP	<i>Bothriocephalus</i>	LBP	MBP	<i>Pomphorhynchus</i>	LPP	MPP
Temperature	.010	.132	.932*	.170	.840	-.383	.634	-.580	-.317
Dissolved O ₂	-.052	-.502	-.688	-.165	-.840	.577	-.905*	.688	-.150
Free CO ₂	-.038	-.686	.724	.115	.275	.208	-.311	.067	-.966**
pH	.662	.288	-.484	.370	.245	-.755	.398	.328	.735

Table 64: Correlation between the physicochemical parameters and Chromium concentrations of Fish tissues and different parasites during summer season of River Jhelum

Water	<i>Adenoscolex</i>	LAP	MAP	<i>Bothriocephalus</i>	LBP	MBP	<i>Pomphorhynchus</i>	LPP	MPP
Temperature	-.367	-.132	.918*	-.252	-.468	-.840	-.298	-.624	-.665
Dissolved O ₂	-.236	-.394	-.880*	.161	.199	.536	-.289	.259	.627
Free CO ₂	.147	.493	.855	.263	-.544	-.911*	-.386	-.781	-.761
pH	-.749	-.380	.306	-.147	-.330	-.690	-.866	-.661	-.166

Table 65: Correlation between the physicochemical parameters and Cobalt concentrations of Fish tissues and different parasites during autumn season of Dal Lake

Water	<i>Adenoscolex</i>	LAP	MAP	<i>Bothriocephalus</i>	LBP	MBP	<i>Pomphorhynchus</i>	LPP	MPP
Temperature	.373	.895*	.890*	-.575	-.324	.412	.603	-.451	.128
Dissolved O ₂	.414	-.112	-.572	.376	-.002	-.139	-.275	.591	.026
Free CO ₂	-.375	-.883*	-.805	.672	.618	.302	-.791	.893*	.374
pH	.737	.738	.446	-.052	.066	.524	.133	.045	.009

Table 66: Correlation between the physicochemical parameters and Cobalt concentrations of Fish tissues and different parasites during winter season of Dal Lake

Water	<i>Adenoscolex</i>	LAP	MAP	<i>Bothriocephalus</i>	LBP	MBP	<i>Pomphorhynchus</i>	LPP	MPP
Temperature	-.149	.586	-.828	-.891*	.207	-.151	.198	-.293	.224
Dissolved O ₂	-.426	.288	-.818	-.726	.145	-.432	.113	-.539	-.155
Free CO ₂	.013	-.753	.526	.767	-.427	.076	-.519	.418	-.443
pH	-.149	.269	-.617	-.734	.530	-.205	.181	-.578	-.099

Table 67: Correlation between the physicochemical parameters and Cobalt concentrations of Fish tissues and different parasites during spring season of Dal Lake

Water	<i>Adenoscolex</i>	LAP	MAP	<i>Bothriocephalus</i>	LBP	MBP	<i>Pomphorhynchus</i>	LPP	MPP
Temperature	.577	-.586	.028	.417	-.140	.577	-.580	-.705	.784
Dissolved O ₂	-.317	-.136	-.946*	.449	.099	.325	.206	.243	-.569
Free CO ₂	.803	-.625	.417	.410	.134	-.061	-.481	-.674	.431
pH	.420	.182	-.466	.527	.928*	.049	.802	.650	-.571

Table 68: Correlation between the physicochemical parameters and Cobalt concentrations of Fish tissues and different parasites during summer season of Dal Lake

Water	<i>Adenoscolex</i>	LAP	MAP	<i>Bothriocephalus</i>	LBP	MBP	<i>Pomphorhynchus</i>	LPP	MPP
Temperature	.040	.382	.213	.283	-.489	-.309	.368	.420	.181
Dissolved O ₂	.020	-.560	-.077	.063	.755	.504	.115	.153	.282
Free CO ₂	-.179	.635	.208	-.116	-.807	-.371	-.031	-.209	-.136
pH	.392	-.823	-.440	.357	.698	.138	.141	.262	.021

Table 69: Correlation between the physicochemical parameters and Cobalt concentrations of Fish tissues and different parasites during autumn season of River Jhelum

Water	<i>Adenoscolex</i>	LAP	MAP	<i>Bothriocephalus</i>	LBP	MBP	<i>Pomphorhynchus</i>	LPP	MPP
Temperature	-.939*	.554	.212	.420	.197	.530	-.926*	.418	-.076
Dissolved O ₂	-.850	.476	.163	.186	-.013	.528	-.862	.348	-.140
Free CO ₂	-.783	.288	.451	.876	.612	.551	-.473	.148	.076
pH	-.667	.265	-.289	.649	.919*	.275	-.636	.828	-.281

Table 70: Correlation between the physicochemical parameters and Cobalt concentrations of Fish tissues and different parasites during winter season of River Jhelum

Water	<i>Adenoscolex</i>	LAP	MAP	<i>Bothriocephalus</i>	LBP	MBP	<i>Pomphorhynchus</i>	LPP	MPP
Temperature	-.655	-.712	-.215	-.148	-.134	.468	.563	.399	-.430
Dissolved O ₂	-.572	-.836	-.288	-.006	.454	.469	-.193	-.228	-.277
Free CO ₂	.584	.911*	.469	.025	-.363	-.545	.235	.206	.105
pH	-.991**	-.432	.069	-.777	.408	.924*	.254	.502	.200

Table 71: Correlation between the physicochemical parameters and Cobalt concentrations of Fish tissues and different parasites during spring season of River Jhelum

Water	<i>Adenoscolex</i>	LAP	MAP	<i>Bothriocephalus</i>	LBP	MBP	<i>Pomphorhynchus</i>	LPP	MPP
Temperature	.674	.779	-.024	.789	-.332	-.514	.831	.741	-.039
Dissolved O ₂	-.321	-.468	.131	-.484	.173	.288	-.732	-.916*	-.418
Free CO ₂	.883*	.835	.223	.796	-.345	-.535	.415	-.116	-.858
pH	-.462	-.372	-.174	-.750	.605	.656	-.188	.273	.838

Table 72: Correlation between the physicochemical parameters and Cobalt concentrations of Fish tissues and different parasites during summer season of River Jhelum

Water	<i>Adenoscolex</i>	LAP	MAP	<i>Bothriocephalus</i>	LBP	MBP	<i>Pomphorhynchus</i>	LPP	MPP
Temperature	.243	-.290	.968**	.222	-.244	.208	-.337	.127	-.639
Dissolved O ₂	-.322	-.038	-.602	-.499	-.271	.249	.123	-.480	.966**
Free CO ₂	.430	-.335	.583	-.016	.319	-.020	-.437	-.215	-.637
pH	-.168	-.303	.659	-.176	-.334	.313	-.199	-.625	.221

Table 73: Correlation between the physicochemical parameters and Copper concentrations of Fish tissues and different parasites during autumn season of Dal Lake

Water	<i>Adenoscolex</i>	LAP	MAP	<i>Bothriocephalus</i>	LBP	MBP	<i>Pomphorhynchus</i>	LPP	MPP
Temperature	.701	-.167	.995**	-.253	-.792	.591	-.521	-.050	.000
Dissolved O ₂	-.038	.812	-.257	.552	.062	-.155	.387	.270	-.662
Free CO ₂	-.033	.154	-.711	.697	.868	.070	.529	.713	-.295
pH	.767	-.016	.844	-.046	-.747	.534	.004	.232	-.596

Table 74: Correlation between the physicochemical parameters and Copper concentrations of Fish tissues and different parasites during winter season of Dal Lake

Water	<i>Adenoscolex</i>	LAP	MAP	<i>Bothriocephalus</i>	LBP	MBP	<i>Pomphorhynchus</i>	LPP	MPP
Temperature	-.815	-.635	-.253	.174	.282	.016	.374	.285	-.540
Dissolved O ₂	-.657	-.301	-.383	.434	.633	-.101	.267	.088	-.516
Free CO ₂	.589	.693	.518	-.127	-.199	.301	-.638	-.490	.260
pH	-.561	-.421	-.608	.625	.626	-.265	.275	.000	-.500

Table 75: Correlation between the physicochemical parameters and Copper concentrations of Fish tissues and different parasites during spring season of Dal Lake

Water	<i>Adenoscolex</i>	LAP	MAP	<i>Bothriocephalus</i>	LBP	MBP	<i>Pomphorhynchus</i>	LPP	MPP
Temperature	.612	.916*	-.712	-.536	.741	.581	-.543	-.403	.470
Dissolved O ₂	.009	-.444	.819	.258	-.432	.312	.240	.539	.283
Free CO ₂	.464	.780	-.814	-.715	.288	.374	-.391	-.500	.162
pH	.522	-.048	.184	.524	-.619	.543	.883*	-.362	.592

Table 76: Correlation between the physicochemical parameters and Copper concentrations of Fish tissues and different parasites during summer season of Dal Lake

Water	<i>Adenoscolex</i>	LAP	MAP	<i>Bothriocephalus</i>	LBP	MBP	<i>Pomphorhynchus</i>	LPP	MPP
Temperature	-.122	.713	-.516	.188	.283	-.466	.949*	.342	-.045
Dissolved O ₂	-.162	-.490	.430	.440	-.096	.559	-.675	.015	-.384
Free CO ₂	.270	.577	-.372	-.425	.270	-.420	.633	-.085	.406
pH	-.500	-.464	.107	.375	-.378	.204	-.558	.010	-.598

Table 77: Correlation between the physicochemical parameters and Copper concentrations of Fish tissues and different parasites during autumn season of River Jhelum

Water	<i>Adenoscolex</i>	LAP	MAP	<i>Bothriocephalus</i>	LBP	MBP	<i>Pomphorhynchus</i>	LPP	MPP
Temperature	.330	.615	-.074	-.143	.387	.627	.437	.107	.343
Dissolved O ₂	.215	.432	-.052	-.228	.218	.427	.505	.212	.294
Free CO ₂	.401	.852	-.156	-.207	.486	.788	-.332	-.267	.675
pH	.098	.929*	-.514	.358	.969**	.706	.214	.223	-.010

Table 78: Correlation between the physicochemical parameters and Copper concentrations of Fish tissues and different parasites during winter season of River Jhelum

Water	<i>Adenoscolex</i>	LAP	MAP	<i>Bothriocephalus</i>	LBP	MBP	<i>Pomphorhynchus</i>	LPP	MPP
Temperature	-.956*	.245	-.869	-.368	.674	.091	.286	.496	-.466
Dissolved O ₂	-.571	-.433	-.876	.393	.982**	.308	.366	.423	.158
Free CO ₂	.490	.469	.793	-.358	-.999**	-.475	-.370	-.512	-.290
pH	-.692	.284	-.798	-.034	.605	-.196	.919*	.894*	.120

Table 79: Correlation between the physicochemical parameters and Copper concentrations of Fish tissues and different parasites during spring season of River Jhelum

Water	<i>Adenoscolex</i>	LAP	MAP	<i>Bothriocephalus</i>	LBP	MBP	<i>Pomphorhynchus</i>	LPP	MPP
Temperature	-.504	-.042	-.873	-.093	-.752	.054	.333	.121	.497
Dissolved O ₂	.108	.242	.639	.485	.740	-.276	-.652	-.082	-.255
Free CO ₂	-.865	.362	-.705	.734	-.234	-.444	-.468	.080	.631
pH	.948*	-.624	.473	-.421	.370	.093	.745	-.627	-.327

Table 80: Correlation between the physicochemical parameters and Copper concentrations of Fish tissues and different parasites during summer season of River Jhelum

Water	<i>Adenoscolex</i>	LAP	MAP	<i>Bothriocephalus</i>	LBP	MBP	<i>Pomphorhynchus</i>	LPP	MPP
Temperature	-.621	.119	-.920*	-.499	.184	.638	.499	.000	.013
Dissolved O ₂	.970**	-.570	.650	.564	-.640	-.765	-.908*	.329	.510
Free CO ₂	-.708	.682	-.578	.044	.109	.853	.608	-.516	-.362
pH	.095	-.200	-.424	.146	-.670	.057	-.352	.328	.630

Table 81: Correlation between the physicochemical parameters and Manganese concentrations of Fish tissues and different parasites during autumn season of Dal Lake

Water	<i>Adenoscolex</i>	LAP	MAP	<i>Bothriocephalus</i>	LBP	MBP	<i>Pomphorhynchus</i>	LPP	MPP
Temperature	.872	-.596	-.431	-.548	.168	.401	-.588	.417	.644
Dissolved O ₂	-.565	-.194	.881*	-.193	-.621	.137	.175	-.493	-.713
Free CO ₂	-.544	.459	.491	.738	.247	-.821	.938*	-.298	-.372
pH	.494	-.881*	-.155	-.650	.142	.315	-.422	-.107	.165

Table 82: Correlation between the physicochemical parameters and Manganese concentrations of Fish tissues and different parasites during winter season of Dal Lake

Water	<i>Adenoscolex</i>	LAP	MAP	<i>Bothriocephalus</i>	LBP	MBP	<i>Pomphorhynchus</i>	LPP	MPP
Temperature	-.238	.222	.598	.641	.844	.227	-.008	.968**	-.917*
Dissolved O ₂	.151	.207	.459	.367	.883*	.386	-.156	.924*	-.910*
Free CO ₂	.250	-.300	-.800	-.866	-.843	.021	-.265	-.802	.837
pH	.118	.543	.747	.537	.997**	-.001	.245	.858	-.964**

Table 83: Correlation between the physicochemical parameters and Manganese concentrations of Fish tissues and different parasites during spring season of Dal Lake

Water	<i>Adenoscolex</i>	LAP	MAP	<i>Bothriocephalus</i>	LBP	MBP	<i>Pomphorhynchus</i>	LPP	MPP
Temperature	.088	-.674	-.113	-.043	-.475	.479	.747	-.039	-.192
Dissolved O ₂	.736	.539	-.883*	-.891*	-.322	-.367	-.042	-.800	-.012
Free CO ₂	-.557	-.587	.180	.256	-.111	-.013	.772	.027	-.642
pH	.113	.885*	-.347	-.585	.571	-.505	-.207	-.555	-.408

Table 84: Correlation between the physicochemical parameters and Manganese concentrations of Fish tissues and different parasites during summer season of Dal Lake

Water	<i>Adenoscolex</i>	LAP	MAP	<i>Bothriocephalus</i>	LBP	MBP	<i>Pomphorhynchus</i>	LPP	MPP
Temperature	-.920*	.356	.006	-.308	-.901*	.268	.018	.861	.006
Dissolved O ₂	.529	-.666	-.435	.568	.588	-.446	-.248	-.718	.154
Free CO ₂	-.521	.573	.364	-.518	-.528	.338	.276	.637	-.313
pH	.496	-.357	-.163	.285	.431	-.409	-.511	-.563	.483

Table 85: Correlation between the physicochemical parameters and Manganese concentrations of Fish tissues and different parasites during autumn season of River Jhelum

Water	<i>Adenoscolex</i>	LAP	MAP	<i>Bothriocephalus</i>	LBP	MBP	<i>Pomphorhynchus</i>	LPP	MPP
Temperature	-.380	.678	-.028	.749	.643	-.944*	.229	-.771	-.633
Dissolved O ₂	-.242	.802	-.252	.782	.606	-.954*	.047	-.856	-.691
Free CO ₂	-.558	.144	.618	.016	.450	-.301	.547	-.261	-.237
pH	-.158	-.047	.328	.324	.724	-.535	.923*	-.208	.223

Table 86: Correlation between the physicochemical parameters and Manganese concentrations of Fish tissues and different parasites during winter season of River Jhelum

Water	<i>Adenoscolex</i>	LAP	MAP	<i>Bothriocephalus</i>	LBP	MBP	<i>Pomphorhynchus</i>	LPP	MPP
Temperature	.088	.207	.778	.196	.791	.892*	-.021	-.264	.960**
Dissolved O ₂	.253	.782	.206	-.141	.410	.899*	-.435	-.501	.731
Free CO ₂	-.400	-.687	-.109	.335	-.256	-.897*	.358	.503	-.706
pH	.665	.271	.580	.211	.549	.805	.397	.341	.824

Table 87: Correlation between the physicochemical parameters and Manganese concentrations of Fish tissues and different parasites during spring season of River Jhelum

Water	<i>Adenoscolex</i>	LAP	MAP	<i>Bothriocephalus</i>	LBP	MBP	<i>Pomphorhynchus</i>	LPP	MPP
Temperature	-.349	-.041	.758	.216	-.542	.371	.287	.140	.034
Dissolved O ₂	.091	-.067	-.417	-.168	.758	-.220	-.666	.038	-.051
Free CO ₂	-.579	-.215	.873	.209	.211	.378	-.633	.354	.036
pH	.774	-.155	-.576	.322	-.518	-.677	.398	-.195	.424

Table 88: Correlation between the physicochemical parameters and Manganese concentrations of Fish tissues and different parasites during summer season of River Jhelum

Water	<i>Adenoscolex</i>	LAP	MAP	<i>Bothriocephalus</i>	LBP	MBP	<i>Pomphorhynchus</i>	LPP	MPP
Temperature	.630	-.317	-.024	-.483	-.902*	-.617	-.244	.902*	-.450
Dissolved O ₂	-.708	-.213	-.274	.030	.424	.510	.109	-.943*	.548
Free CO ₂	.241	-.477	.639	-.649	-.547	-.516	-.402	.801	-.814
pH	.085	-.818	-.074	-.735	-.697	-.099	-.580	.211	-.074

Table 89: Correlation between the physicochemical parameters and Nickel concentrations of Fish tissues and different parasites during autumn season of Dal Lake

Water	<i>Adenoscolex</i>	LAP	MAP	<i>Bothriocephalus</i>	LBP	MBP	<i>Pomphorhynchus</i>	LPP	MPP
Temperature	.301	-.726	.677	.562	-.376	.365	-.076	.099	.233
Dissolved O ₂	.060	.445	.359	-.129	-.097	-.433	.587	.676	.331
Free CO ₂	-.040	.446	-.566	.108	.498	-.584	-.297	.086	-.677
pH	.693	-.759	.870	.739	-.624	.356	-.092	.185	.177

Table 90: Correlation between the physicochemical parameters and Nickel concentrations of Fish tissues and different parasites during winter season of Dal Lake

Water	<i>Adenoscolex</i>	LAP	MAP	<i>Bothriocephalus</i>	LBP	MBP	<i>Pomphorhynchus</i>	LPP	MPP
Temperature	.213	-.127	.778	.068	-.420	-.223	-.807	.843	.143
Dissolved O ₂	.082	-.083	.868	-.251	-.342	-.259	-.573	.872	-.100
Free CO ₂	-.313	-.170	-.608	-.091	.666	.531	.889*	-.650	.013
pH	-.129	.312	.616	-.430	-.683	-.581	-.721	.617	-.060

Table 91: Correlation between the physicochemical parameters and Nickel concentrations of Fish tissues and different parasites during spring season of Dal Lake

Water	<i>Adenoscolex</i>	LAP	MAP	<i>Bothriocephalus</i>	LBP	MBP	<i>Pomphorhynchus</i>	LPP	MPP
Temperature	.113	.186	.804	-.750	-.877	-.038	-.372	.768	-.286
Dissolved O ₂	.343	.385	.188	-.186	.250	.030	-.350	-.779	.399
Free CO ₂	-.545	-.227	.228	-.191	-.894*	.070	.311	.630	-.155
pH	-.047	.596	-.288	-.031	.003	.860	.015	-.630	-.310

Table 92: Correlation between the physicochemical parameters and Nickel concentrations of Fish tissues and different parasites during summer season of Dal Lake

Water	<i>Adenoscolex</i>	LAP	MAP	<i>Bothriocephalus</i>	LBP	MBP	<i>Pomphorhynchus</i>	LPP	MPP
Temperature	-.469	.237	.213	.638	.282	.010	-.355	.813	-.604
Dissolved O ₂	.272	.366	-.438	-.275	.031	-.364	-.221	-.407	.281
Free CO ₂	-.318	-.408	.309	.294	.132	.288	.179	.390	-.222
pH	.513	.487	-.341	-.097	-.171	-.418	-.072	-.412	.277

Table 93: Correlation between the physicochemical parameters and Nickel concentrations of Fish tissues and different parasites during autumn season of River Jhelum

Water	<i>Adenoscolex</i>	LAP	MAP	<i>Bothriocephalus</i>	LBP	MBP	<i>Pomphorhynchus</i>	LPP	MPP
Temperature	.231	.323	-.048	.726	-.914*	-.149	-.366	.237	-.161
Dissolved O ₂	.167	.081	-.043	.591	-.896*	-.203	-.261	.185	-.279
Free CO ₂	.164	.878	-.425	.757	-.324	.118	-.225	.065	.041
pH	-.079	.606	.356	.414	-.645	-.407	-.876	-.034	.672

Table 94: Correlation between the physicochemical parameters and Nickel concentrations of Fish tissues and different parasites during winter season of River Jhelum

Water	<i>Adenoscolex</i>	LAP	MAP	<i>Bothriocephalus</i>	LBP	MBP	<i>Pomphorhynchus</i>	LPP	MPP
Temperature	.779	-.313	.902*	.080	-.641	-.164	.512	-.007	-.727
Dissolved O ₂	.639	-.524	.814	.510	-.651	.091	-.047	-.267	-.989**
Free CO ₂	-.553	.426	-.842	-.678	.629	-.137	.186	.423	.978**
pH	.917*	-.826	.797	.345	.038	.655	.427	.302	-.550

Table 95: Correlation between the physicochemical parameters and Nickel concentrations of Fish tissues and different parasites during spring season of River Jhelum

Water	<i>Adenoscolex</i>	LAP	MAP	<i>Bothriocephalus</i>	LBP	MBP	<i>Pomphorhynchus</i>	LPP	MPP
Temperature	.166	-.716	.416	-.484	.078	.486	.489	-.518	-.603
Dissolved O ₂	-.377	.405	-.703	.676	-.077	-.144	-.327	.396	.538
Free CO ₂	-.394	-.765	-.444	.236	.081	.802	.494	-.374	-.292
pH	-.211	.840	.077	-.311	.396	-.242	-.015	.028	-.038

Table 96: Correlation between the physicochemical parameters and Nickel concentrations of Fish tissues and different parasites during summer season of River Jhelum

Water	<i>Adenoscolex</i>	LAP	MAP	<i>Bothriocephalus</i>	LBP	MBP	<i>Pomphorhynchus</i>	LPP	MPP
Temperature	.046	-.323	-.296	-.936*	-.510	-.798	.082	-.357	.419
Dissolved O ₂	.116	.084	-.233	.716	.228	.490	.177	.621	-.388
Free CO ₂	.144	-.667	-.210	-.708	-.416	-.397	.238	-.814	-.057
pH	-.003	-.705	-.897*	-.420	-.264	-.349	.187	.078	-.196

LAP=liver of fishes infected with *Adenoscolex*; MAP= muscle of fishes infected with *Adenoscolex*; LBP= liver of fishes infected with *Bothriocephalus*; MBP= muscle of fishes infected with *Bothriocephalus*; Lpp= liver of fishes infected with *Pomphorhynchus*; Mpp= muscle of fishes infected with *Pomphorhynchus*; *. Correlation is significant at the 0.05 level (2-tailed); **. Correlation is significant at the 0.01 level (2-tailed).

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CERTIFICATE

Certified that all the corrections/amendments as suggested by External Examiners Prof. M.Z. Chisti, Prof. Emeritus Centre of Research for Development (CORD), University of Kashmir and Prof. R.S. Chauhan, Head Department of Aquaculture, College of Fisheries G.B. Pant, University of Agri. & Tech. Pantnagar, during thesis evaluation and Viva-Voce examination held on December 28, 2015 have been incorporated in the manuscript entitled **“Studies on Gastrointestinal Helminths in the Snow Trouts (Schizothoracids) as Indicators of Aquatic Pollution in Kashmir”** submitted by **Ms. Asifa Wali (Regd. No. 2012-441-D)**.

Prof. M.H. Balkhi
Chairman
Advisory Committee