INCIDENCE OF HELMINTH PARASITES IN COLD WATER FISHES OF RIVER JEHLUM, SRINAGAR, J&K

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INTRODUCTION

Studies on helminth parasites of fishes have been a point of consideration for many researchers in Kashmir on account of increased eutrophication in natural water bodies of the valley. Persistent anthropogenic activity in both, the lacustrine and riverine water bodies of the valley has not only shown a marked deviation in the physicochemical parameters of water but increase in the incidence of parasitofauna. The total number of described fish helminth alone at present exceeds far more than 30,000 (William and Jones, 1994). The pathogenicity of parasitism has been reported to cause extensive damage to the host leading to the lower production of the fish (Rai, 1986). A positive relationship has been reported between eutrophication and fish parasitism (Zarger et al., 2011). It is observed that helminth parasites which inhabit the intestine mostly cause several pathogenic, biochemical and physiological changes (Vinatha, 2012). The physical connection between parasite and host constitutes the micro-environment and the host’s environment can be seen as the macro-environment of the parasite (Gupta et al., 2014). The characteristics of any water body could influence and determine its parasitic fauna when the environmental conditions like food, water, and temperature become favourable for mass reproduction of parasites; the disease may spread very quickly (Srivastava 1975). Voluminous work has been carried out on the aspects of increased eutrophication and its relationship with the incidence of helminth parasitism. Studies on distribution patterns of helminth parasitism have been made by a number of authors’ like Cole (1954); Schad (1963); Tenora (1973); Gupta et al., (1984); Jha et al (1992); Devi (1995); Rajaiah (1997); Chishti and Peerzada (1998); Richardson and Nickol (2000); Khan et al (2001); Rukhsana et al (2008) Sofi and Fayaz (2012); Khurshid et al (2013) and related with the enhanced instances of pollution in water bodies (Zarger et al 2012; Khurshid and Fayaz 2012 & 2014). Levels of parasitic infestations need to be assessed continuously in order to keep an eye on the scenario of parasitic prevalence and density in fishes. In this context, a study on incidence of helminth parasites in Schizothoracine fishes was carried out in River Jhelum falling in the catchment area of Srinagar city of Kashmir. Some Physicochemical parameters of water were analysed and a correlation between parasitic prevalence was also drawn.

MATERIALS AND METHODS

Study site
Kashmir valley is situated at an altitude of 1583 m above the sea level in Himalayas between 34°20’- 34°36’N latitude and 74°82’ - 74°85’E longitudes (Ahanger et al., 2012). The river falls within the area of geographical coordinates of 31.2000°N, 72.1333°E. However, only few sites that fall within the limits of Srinagar
Table 1: Prevalence, mean intensity, relative density and index in Schizothoracines infested by Pomphorhyncus spp., Neoechinorhyncus spp. and Adenoscolex spp.

<table>
<thead>
<tr>
<th>Fish species</th>
<th>Sex</th>
<th>Pr. %</th>
<th>M.I. %</th>
<th>R.D. %</th>
<th>Ind. %</th>
</tr>
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<tbody>
<tr>
<td>Pomp.</td>
<td>Males</td>
<td>25.9</td>
<td>4.28</td>
<td>1.11</td>
<td>7.77</td>
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<td></td>
<td>Females</td>
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<td>3.5</td>
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<tr>
<td>Schizothor. Plagiostomous</td>
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<td>21.4</td>
<td>3</td>
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<tr>
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<td>10.5</td>
<td>11.5</td>
<td>1.21</td>
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<td>1.17</td>
<td>9.52</td>
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<td>1.26</td>
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Table 2: Parasite-wise prevalence in Schizothoracines

<table>
<thead>
<tr>
<th>s.no</th>
<th>Parasite species</th>
<th>No. of samples examined</th>
<th>No. of samples infected</th>
<th>Pr. %</th>
<th>p-value</th>
<th>Int. %</th>
<th>Den. %</th>
<th>Ind. %</th>
</tr>
</thead>
<tbody>
<tr>
<td>1</td>
<td>Pomphorhyncus kashmerensis</td>
<td>200</td>
<td>54</td>
<td>27</td>
<td>&lt; 0.01</td>
<td>3.91</td>
<td>0.27</td>
<td>56.97</td>
</tr>
<tr>
<td>2</td>
<td>Neoechinorhyncus manasbalensis</td>
<td>48</td>
<td>24</td>
<td>24</td>
<td>&lt; 0.01</td>
<td>2.77</td>
<td>0.66</td>
<td>31.92</td>
</tr>
<tr>
<td>3</td>
<td>Adenoscolex orineii</td>
<td>31</td>
<td>15.5</td>
<td>15.5</td>
<td>&lt; 0.01</td>
<td>3.00</td>
<td>0.465</td>
<td>14.41</td>
</tr>
</tbody>
</table>

city were selected for fish and water sample collection on account of intense anthropogenic activities in the catchment area. Randomly selected sites for water and fish sampling included Zero bridge Raj bagh (site 1), Fateh Kadal (site 2) and Qamarwari (site 3). The fishes collected from these sites with the help of local fishermen using Cast net or “Khuri zaal” were brought to the laboratory for identification and subsequent collection of helminth parasites.

Physico-chemical characteristics of water

Water samples from five sites of the River Jhelum were collected on monthly basis and analysed for detection of
concentration of inorganic substances following standard methodology (A.P.H.A, 1998; Golterman and Clyno, 1969). Temperature was recorded by using a mercury filled thermometer and results were expressed as °C. pH of the water was determined with the help of conductivity meter. Depth was measured by Secchi disc. Free carbon dioxide was measured by using phenolphthalein indicator and sodium hydroxide titrant. Conductivity was determined by electrometric method using a laboratory conductivity meter. The alkalinity was examined using phenolphthalein indicator. Dissolved oxygen was determined by modified Wrinkler’s method. Chloride was estimated by Argentometric titration method (APHA, 1998). Nitrate was estimated calorimetrically.

**Examination of fish for Helminth parasitic infestation**

Five Fish species of Schizothoracine ((Schizothorax niger, S. plagiostomous, S. labiatus, S. esocinus and S. curvifrons) were collected on monthly basis and carried to laboratory in plastic bags. Every effort was made to keep them alive. After giving them serial number, morphometric characters including total length, fork length, total weight and sex were determined. The external examination was made by the observation of external features of fish. The colour and skin of the fish was observed by magnifying glass. Presence of cloudiness in eyes, reddening of skin, ragged or torn fins, black spots and ocular reflex were examined. Fishes were killed by giving strong blow on their head. Gills were taken out and kept in normal saline (65% NaCl, Cable, 1977) in a petridish. The parasitological examination of fishes was carried out as per the methodology of Scharperclaus (1986). The parasites were processed and identified with the help of keys provided by Manwell (1968) and Yamaguti (1959). The prevalence, mean intensity and relative density of helminth parasites were calculated in accordance with that of Margolis et al. (1982). The data collected was statistically analysed using SPSS version 20 software. Data were expressed as mean ± S.D. and significant correlation and chi square tests were implied wherever necessary.

**RESULTS**

**Levels of infections in fishes**

During present study sample size of 200 Schizothoracine fishes were examined for helminth examination out of which 102 were found infected with over all prevalence of 51.0%.

**Fish species-wise prevalence**

*Pomphorhyncus kashmerensis* (Acanthocephalan) showed highest incidence in Males of *Schizothorax labiatus* (pr. = 52.9 %,) followed by females of *Schizothorax esocinus* (pr. = 41.6 %,) and by females of *Schizothorax labiatus*. Incidence of *Pomphorhyncus* in males and females *S. esocinus* was found statistically significant (<0.01, $\chi^2 = 2.526$).

Prevalence of *Neoechinorhyncus manasbalensis* (an Acanthocephalan) was found in highest percentage in males of *Schizothorax plagiostomous* (pr. = 42.8%) followed by males of *Schizothorax niger* (pr. = 29.6%) and females of *Schizothorax plagiostomous* (pr. = 13.6%).

Prevalence of *Adenoscolex* (cestode) was found in highest percentage in males of *Schizothorax curvifrons* (pr.0=52.3%) followed by males of *Schizothorax labiatus* (pr. =17.6%) and *Schizothorax esocinus* (pr. =15.7 %). (Table 1).

**Parasite wise prevalence**

Among all parasites recovered, *Pomphorhyncus* showed highest prevalence among all parasites (27%) with Mean Intensity, Relative density and Index of 3.91%, 0.27% and

**Table 3:** Fish gender-wise prevalence

<table>
<thead>
<tr>
<th></th>
<th>Males</th>
<th>Neoechinorhyncus</th>
<th>Adenoscolex</th>
<th>Females</th>
<th>Neoechinorhyncus</th>
<th>Adenoscolex</th>
</tr>
</thead>
<tbody>
<tr>
<td>Total no. Of fishes</td>
<td>98</td>
<td>98</td>
<td>98</td>
<td>102</td>
<td>102</td>
<td>102</td>
</tr>
<tr>
<td>Infected hosts</td>
<td>29</td>
<td>22</td>
<td>35</td>
<td>25</td>
<td>9</td>
<td>13</td>
</tr>
<tr>
<td>Pr. %</td>
<td>29.591</td>
<td>22.448</td>
<td>35.714</td>
<td>24.509</td>
<td>8.823</td>
<td>12.745</td>
</tr>
<tr>
<td>Int. %</td>
<td>337.931</td>
<td>413.636</td>
<td>188.571</td>
<td>452.000</td>
<td>466.666</td>
<td>207.692</td>
</tr>
<tr>
<td>Den. %</td>
<td>1.00</td>
<td>0.928</td>
<td>0.673</td>
<td>1.107</td>
<td>0.411</td>
<td>0.264</td>
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<tr>
<td>Ind. %</td>
<td>29</td>
<td>20.428</td>
<td>23.571</td>
<td>27.69</td>
<td>3.705</td>
<td>3.441</td>
</tr>
</tbody>
</table>

**Table 4:** Means of physico-chemical parameters of water of River Jhelum at different sites of fish collection from June to November

<table>
<thead>
<tr>
<th>Physicochemical parameters of water</th>
<th>Site 1</th>
<th>Site 2</th>
<th>Site 3</th>
</tr>
</thead>
<tbody>
<tr>
<td>Atmospheric temperature °C</td>
<td>27.58 ± 11.36</td>
<td>27.76 ± 5.04</td>
<td>27.6 ± 4.82</td>
</tr>
<tr>
<td>Water temperature °C</td>
<td>22.86 ± 4.23</td>
<td>23.63 ± 3.75</td>
<td>23.20 ± 3.44</td>
</tr>
<tr>
<td>Depth (meters)</td>
<td>1.86 ± 0.314</td>
<td>1.23 ± 0.18</td>
<td>1.15 ± 0.15</td>
</tr>
<tr>
<td>Transparency (meters)</td>
<td>0.55 ± 0.2</td>
<td>0.48 ± 0.12</td>
<td>0.55 ± 0.24</td>
</tr>
<tr>
<td>pH</td>
<td>7.91 ± 0.13</td>
<td>7.78 ± 0.12</td>
<td>7.93 ± 0.10</td>
</tr>
<tr>
<td>Dissolved oxygen (mg/l)</td>
<td>6.97 ± 0.32</td>
<td>6.98 ± 0.27</td>
<td>7.0 ± 0.34</td>
</tr>
<tr>
<td>Carbon dioxide (mg/l)</td>
<td>4.15 ± 0.90</td>
<td>4.87 ± 0.97</td>
<td>4.53 ± 0.83</td>
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<tr>
<td>Total Alkalinity (mg/l)</td>
<td>128.66 ± 3.61</td>
<td>97.66 ± 4.08</td>
<td>105.17 ± 12.64</td>
</tr>
<tr>
<td>Nitrate (NO$_3$-N) (μg/l)</td>
<td>1285.67 ± 97.65</td>
<td>1122.33 ± 39.33</td>
<td>1134.33 ± 38.32</td>
</tr>
<tr>
<td>Ammonia (NH$_4$-N) (μg/l)</td>
<td>388.50 ± 45.30</td>
<td>337.83 ± 27.72</td>
<td>326.16 ± 25.40</td>
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<tr>
<td>Orthophosphate po$_4$-P (μg/l)</td>
<td>402.0 ± 7.29</td>
<td>410.33 ± 11.75</td>
<td>425.0 ± 22.19</td>
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<tr>
<td>Total Phosphorous (μg/l)</td>
<td>1467.66 ± 59.70</td>
<td>1385.5 ± 59.98</td>
<td>1490.17 ± 55.96</td>
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Site 1: Zero bridge, site 2: Fatehkadal, site 3: Quramarwar
56.97% respectively followed by Neoechinorhyncus (Pr. = 24%, M.I. = 2.77% R.D. = 0.66% and Ind. = 31.92%) and Adenoscolex (Pr. = 15.5%, M.I. = 3.00%, R.D. = 0.465% and Ind. = 14.41%) (Table: 2). The parasites were found infected with Adenoscolex in bulk attached to fish intestines and liver. Neoechinorhyncus also showed the same pattern of distribution in fishes (pr = 24%, M.I = 2.77%, R.D. = 0.66% and Ind. = 31.92%). Adenoscolex was found in bunches entangled together depicting high index in fishes (pr = 15.5 %, M.I. = 20.428% and R.D. = 0.465% and Ind. = 14.415%). The presence of all the parasite species in all the fishes was found statistically significant (<0.01) in all fishes.

**Fish gender-wise prevalence**
Among 98 male Schizothoracine fishes, 29 were found infected with *Pomphorhyncus kashmerensis* with overall pr. = 29.591%, M.I. = 337.931%, R.D. = 1.00% and Ind. = 29.00%. *Neoechinorhyncus manasbalensis* infection was found in 35 fishes (pr = 22.448%, M.I. = 413.636%, R.D. = 0.928% and Ind. = 20.428%) and *Adenoscolex orineii* was found in 22 Schizothoracine fish species (pr = 35.714%, M.I. = 188.571%, R.D. = 0.673% and Ind. = 23.571%).

Among 102 female *Schizothoracine* fishes 25 were found infected with *Pomphorhyncus kashmerensis* (pr. = 24.509%, M.I. = 452.00%, R.D. = 1.107% and Ind. = 27.69%). 9 females were found infected with *Neoechinorhyncus manasbalensis* (pr. = 8.823%, M.I. = 466.66%, R.D. = 0.441% and Ind. = 3.705%) and 13 with *Adenoscolex orineii* (pr. = 12.745%, M.I. = 207.692%, R.D. = 0.264% and Ind. = 3.441%).

**Water Quality Parameters**
Atmospheric temperature at the river showed a wide fluctuation during the study period and ranged from 24.64±9.41 in the month of June to 10.78±1.36°C in December. Water temperature ranged from 15.7±0.483 to 8.525±0.947°C during these months. Depth of the river was 0.855±0.29 m during the month of June and 1.221±0.37 m in November depicting some obvious effects on other physico-chemical parameters. Dissolved oxygen of the river was recorded in the range of 3.838±0.537 and 4.49±1.16 mg/l from summer to winter respectively. Carbon dioxide showed a little variation from 3.2 ± 2.1 mg/l from summer to end of autumn. pH didn't show wide fluctuation and generally remained alkaline in the range of 7.713±0.181 to 7.5±0.382 from June to November. Alkalinity varied from 238.3±38.8 to 334±49.1 mg/l. Calcium levels ranged between 35.45±5.53 and 29.44±2.05 mg/l and Magnesium between 9.95±1.76 and 11.64±1.89 mg/l during the study period. Ammonia was 131.8±17.3 in June and 140.8±24.5 in November. Nitrite change to Nitrate was shown between the range of 440.6±74.3 and 357.6±48.9 from June to November.

**DISCUSSION**
Water bodies have undergone a progressive deterioration due to persistent anthropogenic pressure as an outcome of urbanisation and industrialisation. Parasitic infestations regarded as secondary disease occur mostly in the fishes already stressed or infected with any other pathogen. Environmentally stressed fish fall easy prey to the larval forms of parasites and larval recruitment become more intensive during these conditions. However, one obvious prediction is that pollutants may reduce the immunological capabilities of hosts, rendering them more susceptible to some parasites (McDowell et al., 1999). It is now established that eutrophication plays an important role in rise, maturation and abundance of fish parasites. Valtonen et al. (1997) described how eutrophication among lakes and over time was associated with greater overall parasite species richness in *Rutilus rutilus* and *Perca fluviatilis*.

Water quality of River Jhelum was found altered in the area falling in the catchment of Srinagar city. From site 1 (Zero bridge Rajbagh) up to site 3 (Qamarwari) encompasses the whole urbanised area all the sewage from houses, sullage, animal droppings and human excretions directly find their way into the river. Moreover, the entire domestic wastes from dongas, home to numerous people residing in the river is also dumped in it. The fishermen community living along the banks of the river depict low socio-economic status, less education to maintain the cleanliness of the water body and of course no or insignificant drainage system that has led to disturbance in its water quality. Our findings are in agreement with Khan et al. (2012) who reported the river Jhelum was polluted with altered water quality. Parasitic prevalence was found high at the sites where alterations in the physicochemical parameters of water were noticed. Moreover, the density of *Pomphorhyncus kashmerensis* was found high at site 2 & 3 (Fateh Kadal and Qamarwari) in the month of July. This can be attributed to increase in temperatures (27.76±27.6±4.82°C respectively) and reported by Ali et al., (2010) who reported that the prevalence of Acanthocephala is dependent on various factors like temperature, food availability. Eure and Esch (1974) found that acanthocephalans were more common near thermal effluent because intermediate hosts were able to retain high densities in winter. We found the *Pomphorhyncus kashmerensis* attached with its proboscis into intestinal bulb, crypts of Lieberkuhn and to liver. Moreover, at the site of attachment of proboscis in the intestines, fishes had developed a tuft of granulomatic cells depicting the reaction of host defence towards parasitic attachment (Feroz et al. 2005).

Among Acanthocephalans, the highest number of *Pomphorhynchus* (pr. = 27%) observed in the study may be due to its wide host range that is inconsonance with Chishti et al., 1999). It is now established that river Jhelum was polluted with anthropogenic pressure as an outcome of urbanisation and industrialisation. Parasitic infestations regarded as secondary disease occur mostly in the fishes already stressed or infected with any other pathogen. Environmentally stressed fish fall easy prey to the larval forms of parasites and larval recruitment become more intensive during these conditions. However, one obvious prediction is that pollutants may reduce the immunological capabilities of hosts, rendering them more susceptible to some parasites (McDowell et al., 1999). It is now established that eutrophication plays an important role in rise, maturation and abundance of fish parasites. Valtonen et al. (1997) described how eutrophication among lakes and over time was associated with greater overall parasite species richness in *Rutilus rutilus* and *Perca fluviatilis*.
the addition of various carbon rich substances as majority of carbon comes from organic matter such as dead terrestrial plant material (Wetzel, 1992) and direct inf low of untreated sewage. The change in CO₂ content in water has resulted in the changing trend of alkalinity also, albeit later depicted a decreasing trend from June to November at all the three sites.

Depth of the river showed a gradual decreasing trend on account of changing season from summer to autumn as during summer melting of ice is more that results in increase of volume of water in the river. It has a direct effect of the transparency of the river also which in the present study showed a similar decreasing trend from June to November. pH of the river remained more or less alkaline during the period of study. Ammonia (NH₃-N), Nitrate (NO₃-N), orthophosphates PO₄-P and total phosphorous showed an overall increase with slight fluctuation at certain points. On these sites Adenoscolex orinei was found in bunches and at certain instances entangled with other each depicting their heavy density in fishes. It may be attributed to direct inf low of untreated sewage at various point source sites of the river. Moreover, increase in the population around the catchment of the lake has definitely resulted in the cultural eutrophication in the river.

The incidence of helminth parasites presents a review in monitoring water pollution of water bodies (Sures, 2004). Various workers 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 (Beer and German, 1993; Kennedy and Watt, 1994; Marcogliese, 2001; Lafferty and Kuris, 2005; Zarger et al., 2012).

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