ORIGINAL ARTICLE / ARTÍCULO ORIGINAL

SEASONAL DYNAMICS OF TWO SPIRURID NEMATODES (NEMATODA: RHABDOCHONIDAE) IN SCHIZOTHORAX RICHARDSONII (GRAY, 1832) FROM FIVE COLLECTION SITES OF DISTRICT CHAMBA, HIMACHAL PRADESH, INDIA

DINÁMICA ESTACIONAL DE DOS NEMATODOS ESPIRÚRIDOS (NEMATODA: RHABDOCHONIDAE) EN SCHIZOTHORAX RICHARDSONII (GRAY, 1832) DE CINCO SITIOS DE RECOLECCIÓN DEL DISTRITO DE CHAMBA, HIMACHAL PRADESH, INDIA

Suman Kumari¹*; Yanchen Dolma¹ & Deepak C. Kalia²

¹ Department of Life-Sciences, MCM DAV College Kangra (H.P.), India 176001.
² Department of Bio-Sciences, Himachal Pradesh University, Summer Hill Shimla (H.P.), India 171005.
* Corresponding author: sumanhpu11@gmail.com

Suman Kumari: https://orcid.org/0000-0002-3255-8442
Yanchen Dolma: https://orcid.org/0000-0001-8862-4181
Deepak C. Kalia: https://orcid.org/0000-0002-9478-3899

ABSTRACT

The present study deals with a seasonal survey of nematode parasites from district Chamba (H.P.) India, and summarizes the data of prevalence, intensity, density and index of infections of Rhabdochona (Rhabdochona) denudata (Dujardin, 1845) Railliet, 1916 and Rhabdochona (Filocha) hellichis turkestanica (Skrjabin, 1917) Moravec, Scholz, Ash and Kar, 2010 in the freshwater fish, Schizothorax richardsonii (Gray, 1832). Fish samples were collected from five collection sites of district Chamba i.e. (Chamba, Sukrala, Rari, Saho, and Kheri). During the present study, a total of 293 (92 ♂ + 201 ♀) randomly selected fish hosts were examined, in which 163 fish were infected with two species of nematodes. Seasonal studies for both the species from random-sized hosts exhibited higher prevalence, intensity, density, and index of infection in the summer season and least during the rainy season. Of the two species, R. (Filocha) hellichis turkestanica dominated in four localities, with higher values of intensity of infection, density, and index of infection during all three seasons.

Keywords: Nematode – Rhabdochona – Schizothorax – Seasonal dynamics
RESUMEN

El presente estudio trata de la evaluación estacional de nematodos parásitos del distrito de Chamba (H.P.) India, y resume los datos de prevalencia, intensidad, densidad e índice de infecciones de *Rhabdochona (Rhabdochona) denudata* (Dujardin, 1845) Railliet, 1916 y *Rhabdochona (Filochona) hellichi turkestanica* (Skrjabin, 1917) Moravec, Scholz, Ash & Kar, 2010, en peces de agua dulce, *Schizothorax richardsonii* (Gray, 1832). Se recolectaron muestras de peces de cinco sitios del distrito de Chamba (Chamba, Sukrala, Rari, Saho y Kheri). Durante el presente estudio se examinaron un total de 293 (92 ♂ + 201 ♀) peces huéspedes seleccionados al azar, en los cuales 163 peces estaban infectados con dos especies de nematodos. Los estudios estacionales para ambas especies de hospederos de tamaño aleatorio, exhibieron mayor prevalencia, intensidad, densidad e índice de infección en la temporada de verano y menos durante la temporada de lluvias. De las dos especies, *R. (Filochona) hellichi turkestanica* dominó en cuatro localidades, con mayor valor de intensidad de infección, densidad e índice de infección durante las tres temporadas.

Palabras clave: Nematodo – *Rhabdochona* – *Schizothorax* – Dinámicas estacionales

INTRODUCTION

Fish is a cheap and important source of protein which also contains calcium, lipids, minerals, vitamins and oils with desirably low cholesterol level in the diets of fish lovers (Ugbede et al., 2019). India is third largest producer of fish in the world and second in inland fish production. Fisheries are important for the Indian economy as it provides employment opportunities; is a source of nutritional food and foreign exchanges. The fish is said to be ‘gold’ from water (Kalse & Bhosle, 2015). These edible fishes are known to harbor a number of helminth parasite which cause deterioration in their health, hence their market and nutritive value is affected. The damage caused by helminthes to their hosts is generally related to intensity of infection and depth of parasite penetration with respect to host tissue. Seasonal fluctuation, locality, age, size and sex of the host also determine the parasitic community diversity and burden (Fartade & Chati, 2016). Humans may contract fish- borne zoonotic nematodes by eating fish that has been undercooked or eaten uncooked. Serious outcomes from human infections are possible, including the unexpected death of infected people (Eiras et al., 2018). *Rhabdochona* spp. in the intestine of snow trout has a long term relationship and is considered as natural infection in cyprinids and zoonotic risk to human (Dujardin, 1845; Railliet & Henry, 1916; Khan et al., 2022).

Nematode cause an economic threat to the market value of fish and infected fillets get rejected and can increase production cost (Abiyu et al., 2020). Nematodes can infect fish as adults but larval stages of nematodes infecting piscivorous birds, mammals or reptiles, or less frequently predatory fish, can also infect fish species. Ingestion of uncooked infected fish meat poses a zoonotic threat to humans (Leela, 2016).

*Schizothorax richardsonii* (Gray,1832), commonly known as snow trout is a hillstream and a high valued fish which is famous and preferred for its excellent taste (Yadav et al., 2014).

*Rhabdochona (Rhabdochona) denudata* (Dujardin, 1845) Railliet, 1916 and *Rhabdochona (Filochona) hellichi turkestanica* (Skrjabin, 1917) Moravec, Scholz, Ash and Kar, 2010 are parasitic species of the genus *Rhabdochona*, a widely diverse group of nematode parasitic in freshwater fish (Skrjabin, 1917; Moravec et al., 2010; Moravec & Adlard, 2016).

The present study examines the prevalence, density/abundance, intensity and index of infection of two species *Rhabdochona (Rhabdochona) denudata* (Dujardin, 1845) Railliet, 1916 and *Rhabdochona (Filochona) hellichi turkestanica* (Skrjabin, 1917) Moravec, Scholz, Ash and Kar, 2010 recovered from *S. richardsonii* during three different seasons during 2014.

MATERIALS AND METHODS

During a survey on nematode parasites of *S. richardsonii*, a total of 293 (92 ♂ + 201 ♀) fish hosts were examined from five collection sites (Fig. 1) of district Chamba from Indian hill state of Himachal Pradesh. Chamba district is situated between North latitude 32° 10’ and 33° 13’ and East longitude 75° 45’ and 77° 33’. The principal
river that drains the entire Chamba valley between the Dhauladhar and Pangi hills is the Ravi. It ascends in the mid-Himalaya and Bara Bhangal ranges. It departs the neighborhood at Kheri and travels to Shahpur’s plains (Himachal Pradesh Development Report, 2014; Balokhra, 2015).

Figure 1. Collection sites.

The fish was purchased from the local fisherman, almost immediate to its capture to avoid the deterioration of health of parasitic fauna. The hosts were dissected along the ventral line. The visceral organs were placed in different containers containing saline solution. The mucosal lining of the intestine was scrapped gently with the help of scalpel whereas, the other visceral organs were thoroughly teased into smaller bits to facilitate the release of the embedded parasites. The specimens were washed thoroughly in saline to remove the debris, if any.

The nematode parasites recovered in saline from the hosts were killed and fixed by pouring boiling hot 3-4% formalin or boiling hot 70% ethyl alcohol on the bulk of specimens retained in the thin film of saline by draining off excess of the saline. This step also caused the almost straightening of the nematodes. The fixed nematodes were preserved in fresh fixative in glass vials and labelled for date, name, locality of host and location of parasites. Voucher specimens (668) are deposited in the parasitology laboratory, department of Biosciences, Himachal Pradesh University, Shimla.

The nematodes were bulk cleared in lactophenol (Glycerin: 2 parts; Distilled water: 1 part; Lactic acid: 1 part & phenol crystal (melted): 1 part) or glycerin (involving a series of upgrading mixtures of 70% alcohol & glycerin for the preparation of temporary mounts). The parasites were identified following Anderson et al. (2009) and Gibbons (2010) for the generic diagnosis of the worms, followed by consultation of the relevant literature available worldwide, for the validation of the species. The data obtained were analyzed following the formula of Tenoza & Zejda (1974), Margolis et al. (1982) and Bush et al. (1997) to study the following parameters.

\[
\text{Prevalence} = \left( \frac{\text{Total number of hosts infected}}{\text{Total number of hosts examined}} \right) \times 100
\]

\[
\text{Intensity of infection} = \frac{\text{Number of parasites collected}}{\text{Number infected hosts}}
\]

\[
\text{Density/Abundance} = \frac{\text{Number of parasites collected}}{\text{Number of hosts examined}}
\]
Index of infection: It is expressed by the formula of Tenoza & Zejda (1974)

\[ Z = \frac{A \times B}{C^2} \]

where, \( Z = \) Index of infection

\( A = \) Number of parasites collected

\( B = \) Number of hosts infected

\( C = \) Number of hosts examined

Ethical aspects: The protocol for collection of fishes and nematodes are in accordance with the norms of ethical committee, department of Biosciences, Himachal Pradesh University, Shimla and Himachal Pradesh forest department wildlife wing No. WL/ Research study / 234-36.

RESULTS

During a survey on nematode parasites of S. richardsonii, a total of 293 (92 ♂ + 201 ♀) fish hosts were examined from five collection sites of district Chamba: locality I (Chamba), locality II (Sukrala), locality III (Rari), locality IV (Saho) and locality V (Kheri) (Fig. 1) during three (in agreement with Himachal Pradesh Development report, 2014) different seasons; winter season (January-February, 2014), summer season (April-May, 2014) and rainy season (August-September, 2014) with the climate of Himachal Pradesh varying from semi tropical to the semi-arctic depending upon the altitude and the three seasons time period, with the rainy from July to September, winter from October to March and summer from April to June. The recovered nematode parasites (105 ♂ + 599 ♀) were of the two species of Rhabdochona Railliet, 1916; R. (Rhabdochona) denudata (49 ♂ + 283 ♀) and R. (Filochona) hellichi turkestanica (56 ♂ + 316 ♀).

The seasonal dynamics for both the species from random-sized hosts, at five different localities exhibited higher prevalence of nematode parasite in summer season (April-May) (55.55%, 61.11%, 84.62%, 86.67%, 40%) (Fig. 2) and the least prevalence during rainy season (August-September) (42.31%, 45.45%, 50%, 55%, 35%), (Fig. 3) with an overall prevalence of 49.25%, 54.76%, 65.95%, 71.83%, 37.87%. The intensity of infection, density or abundance and index of infection for both the species of nematodes were also observed as maximum during summer season with 1.8, 1.82, 4.09, 2.23, 1.16; 1, 1.1, 3.46, 1.93, 0.47; 0.56, 0.68, 2.92, 1.68, 0.19 (Fig. 5, 8, 11) respectively for R. (Rhabdochona) denudata and 2.3, 2.45, 2.72, 2.88, 1.83; 1.28, 1.5, 2.31, 2.5, 0.73; 0.71, 0.92, 1.95, 2.17, 0.29 (Fig. 5, 8, 11) respectively for R. (Filochona) hellichi turkestanica. The record for rainy season was 1.36, 1.40, 3.33, 1.64, 1.14; 0.58, 0.64, 1.67, 0.9, 0.4; 0.24, 0.29, 0.83, 0.49, 0.14 (Fig. 6, 9, 12) respectively for R. (Rhabdochona) denudata, 1916 and 1.82, 2.17, 2.55, 1.29; 0.91, 1.08, 1.4, 0.45; 0.33, 0.41, 0.54, 0.77, 0.16 (Fig. 6, 9, 12) respectively for R. (Filochona) hellichi turkestanica.

Of the five different localities, the observed prevalence was higher in the locality IV followed by locality III, locality II, locality I and of the least prevalence in locality V, while other parameters, viz., intensity of infection, density/ abundance and index of infection for R. (R.) denudata were higher in locality III followed by locality IV, locality II, locality I and locality V and for R. (F.) hellichi turkestanica these parameters were higher in locality IV followed by locality III, locality II, locality I and least values were observed in locality V (Tables 1 and 2; Fig. 2-13).

Of the above species, R. (F.) hellichi turkestanica dominated in all localities with higher values of intensity of infection (2.28), density or abundance (1.26) and index of infection (0.70), during the three seasons, whereas R. (R.) denudata exhibited the value of 2.04, 1.14 and 0.63 for the above said parameters. The infected hosts consisted mainly of females (114) as compared to 49 males, for both the species.
Table 1. Seasonal dynamics of *Rhabdochona (Rhabdochona) denudata* (Dujardin, 1845) Railliet, 1916.

<table>
<thead>
<tr>
<th>Sr. No.</th>
<th>Localities</th>
<th>Seasons</th>
<th>Months</th>
<th>Number of hosts examined</th>
<th>Number of hosts infected</th>
<th>Number of parasite</th>
<th>Prevalence (%)</th>
<th>Intensity of infection</th>
<th>Density/Abundance</th>
<th>Index of infection</th>
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<tr>
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<td>April- May</td>
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<td>12 4 8</td>
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Table 2. Seasonal dynamics of *Rhabdochona* (*Filochona*) *hellichi turkestanica* (Skrjabin, 1917) Moravec et al., 2010.

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<th>Months</th>
<th>Number of hosts examined</th>
<th>Number of hosts infected</th>
<th>Number of parasite</th>
<th>Prevalence (%)</th>
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Figure 2. Locality-wise prevalence of *Rhabdochona (R.) denudata* (Dujardin, 1845) Railliet, 1916 and *Rhabdochona (F.) hellichi turkestanica* (Skrjabin, 1917) Moravec *et al.*, 2010 in summer season. Locality 1 = Chamba, Locality 2 = Sukrala, Locality 3 = Rari, Locality 4 = Saho, Locality 5 = Kheri.

Figure 3. Locality-wise prevalence of *Rhabdochona (R.) denudata* (Dujardin, 1845) Railliet, 1916 and *Rhabdochona (F.) hellichi turkestanica* (Skrjabin, 1917) Moravec *et al.*, 2010 in rainy season. Locality 1 = Chamba, Locality 2 = Sukrala, Locality 3 = Rari, Locality 4 = Saho, Locality 5 = Kheri.
Figure 4. Locality-wise prevalence of *Rhabdochona (R.) denudata* (Dujardin, 1845) Railliet, 1916 and *Rhabdochona (F.) hellichi turkestanica* (Skrjabin, 1917) Moravec et al., 2010 in winter season. Locality 1 = Chamba, Locality 2 = Sukrala, Locality 3 = Rari, Locality 4 = Saho, Locality 5 = Kheri.

Figure 5. Locality-wise intensity of infection of *Rhabdochona (R.) denudata* (Dujardin, 1845) Railliet, 1916 and *Rhabdochona (F.) hellichi turkestanica* (Skrjabin, 1917) Moravec et al., 2010 in summer season. Locality 1 = Chamba, Locality 2 = Sukrala, Locality 3 = Rari, Locality 4 = Saho, Locality 5 = Kheri.
Figure 6. Locality-wise intensity of infection of *Rhabdochona (R.) denudata* (Dujardin, 1845) Railliet, 1916 and *Rhabdochona (F.) hellichi turkestanica* (Skrjabin, 1917) Moravec et al., 2010 in rainy season. Locality 1 = Chamba, Locality 2 = Sukrala, Locality 3 = Rari, Locality 4 = Saho, Locality 5 = Kheri.

Figure 7. Locality-wise intensity of infection of *Rhabdochona (R.) denudata* (Dujardin, 1845) Railliet, 1916 and *Rhabdochona (F.) hellichi turkestanica* (Skrjabin, 1917) Moravec et al., 2010 in winter season. Locality 1 = Chamba, Locality 2 = Sukrala, Locality 3 = Rari, Locality 4 = Saho, Locality 5 = Kheri.
**Figure 8.** Locality-wise density/abundance of *Rhabdochona (R.)* denudata (Dujardin, 1845) Railliet, 1916 and *Rhabdochona (F.)* hellichi turkestanica (Skrjabin, 1917) Moravec et al., 2010 in summer season. Locality 1 = Chamba, Locality 2 = Sukrala, Locality 3 = Rari, Locality 4 = Saho, Locality 5 = Kheri.

**Figure 9.** Locality-wise density/abundance of *Rhabdochona (R.)* denudata (Dujardin, 1845) Railliet, 1916 and *Rhabdochona (F.)* hellichi turkestanica (Skrjabin, 1917) Moravec et al., 2010 in rainy season. Locality 1 = Chamba, Locality 2 = Sukrala, Locality 3 = Rari, Locality 4 = Saho, Locality 5 = Kheri.
Figure 10. Locality-wise density/abundance of *Rhabdochona (R.) denudata* (Dujardin, 1845) Railliet, 1916 and *Rhabdochona (F.) hellichi turkestanica* (Skrjabin, 1917) Moravec et al., 2010 in winter season. Locality 1 = Chamba, Locality 2 = Sukrala, Locality 3 = Rari, Locality 4 = Saho, Locality 5 = Kheri.

Figure 11. Locality-wise index of infection of *Rhabdochona (R.) denudata* (Dujardin, 1845) Railliet, 1916 and *Rhabdochona (F.) hellichi turkestanica* (Skrjabin, 1917) Moravec et al., 2010 in summer season. Locality 1 = Chamba, Locality 2 = Sukrala, Locality 3 = Rari, Locality 4 = Saho, Locality 5 = Kheri.
**Figure 12.** Locality-wise index of infection of *Rhabdochona (R.) denudata* (Dujardin, 1845) Railliet, 1916 and *Rhabdochona (F.) hellichi turkestanica* (Skrjabin, 1917) Moravec et al., 2010 in rainy season. Locality 1 = Chamba, Locality 2 = Sukrala, Locality 3 = Rari, Locality 4 = Saho, Locality 5 = Kheri.

**Figure 13.** Locality-wise index of infection of *Rhabdochona (R.) denudata* (Dujardin, 1845) Railliet, 1916 and *Rhabdochona (F.) hellichi turkestanica* (Skrjabin, 1917) Moravec et al., 2010 in winter season. Locality 1 = Chamba, Locality 2 = Sukrala, Locality 3 = Rari, Locality 4 = Saho, Locality 5 = Kheri.
DISCUSSION

The present result showed that the high prevalence/incidence of infection (86.67%), intensity of infection (4.09, 2.88) and density of infection (3.46, 2.5) for both the species of *Rhabdochona* Railliet, 1916 (Nematoda - Rhabdochonidae) in summer season followed by winter and rainy season, was in agreement with Bhure *et al.* (2010), who while surveying the population ecology of *Rhabdochona* from *Labeo rohita* (Hamilton & Buchanan) indicated that a high incidence of infection (51.78%), intensity of infection (1.18%) and density of infection (0.61%) of *Rhabdochona* spp. occurred in summer season followed by winter season and rainy season. This type of result indicated that environmental factors and feeding habitats are influencing the seasonality of parasitic infection (Deshmukh *et al.*, 2013). Maheboob (2020) made a similar observation for high infections (Intensity, incidence, density and index of infection) of helminth parasite during summer season, followed by winter season and extremely low in monsoon season. It indicated that ecological factors and feeding habitat are affecting the seasonality of parasitic infection either directly or indirectly.

Ezenwaji *et al.* (2005) studied helminth endo-parasites of mochokids in a tropical rainforest river system observed prevalence, mean intensity and abundance of all the endo-parasites to be generally higher in the dry than in the rainy season as the relatively fast flow of water in lotic habitats would inevitably reduce host-parasite contact frequency resulting in low prevalence. Leela & Rao (2014) could observe that *R. garua* Agrawal, 1965 in *Glossogobius giuris* (Hamilton-Buchanan, 1822) from Lower Monair Dam (Andhra Pradesh) had lowest prevalence (28.92%), mean intensity (1.42) and relative density 0.41 for the premonsoon months, the highest prevalence (46.06%), mean intensity (2.32) and relative density (1.07) for the monsoon months (Agrawal, 1965). Kalse & Bhosale (2015) while studying biodiversity and prevalence of helminth parasites of Girna Dam fishes from February 2010- January 2012, observed that the incidence of nematode infections during 2010-11 was maximum (71.62%) in summer season, followed by (69.33%) in monsoon season and lower (63.51%) in winter season whereas for the year 2011-12 these were 37.5%, 32.60 and 35.05 respectively. Kumar *et al.* (2016) while studying the prevalence of a spirurid nematode *R. (F.) hellichi turkestanica* in *Tor putitora* (Hamilton-Buchanan, 1822) from Seer Khad, district Bilaspur (Himachal Pradesh) during May, August and November 2015, observed a high prevalence (85%) in May and November with mean intensity (4.79) and relative abundance (4.07) being high in the month of November only (Hamilton-Buchanan, 1822).

Hence, the present observations revealed that despite the identical prevalence, because of the sympatric relationship, the mean intensity, density/abundance and index of infection were 1.14-4.09, 0.40-3.46, 0.14-2.92 for *R. (Rhabdochona) denudata* and 1.29-2.88, 0.45-2.5, 0.16-2.17 for *R. (Filochona) hellichi turkestanica*.

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Author contributions: CRediT (Contributor Roles Taxonomy)

DCK = Deepak C. Kalia
YD = Yanchen Dolma
SK = Suman Kumari

Conceptualization: DCK, YD, SK
Data curation: SK
Formal Analysis: SK
Funding acquisition: SK
Investigation: SK, YD
Methodology: SK, YD
Project administration: DCK, SK
Resources: DCK, YD, SK
Software: SK
Supervision: DCK
Validation: DCK, YD, SK
Visualization: DCK, YD, SK
Writing – original draft: SK
Writing – review & editing: DCK, YD, SK

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