Gastrointestinal helminth parasites of wild ungulates in Hirpora Wildlife Sanctuary, Kashmir, India

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Abstract Parasitic infection represents an emerging threat to wild ungulates and a challenge to their management. Although a lot of work has been carried out on helminth parasitic infestation of domestic ungulates of Kashmir but the data pertaining to this aspect of wild ungulates has been ignored. The study on gastrointestinal helminth parasitic infestation of wild ungulates was carried out during post livestock grazing period (November to May) of 2018/2019 in Hirpora Wildlife sanctuary (HWLS) to fill the gap in the existing literature. During the study fresh faecal samples of musk deer Moschus spp. (n = 44) and markhor Capra falconeri (n = 41) were collected and examined qualitatively and quantitatively for gastrointestinal helminth parasites. A total of seven helminth parasites were recorded which are arranged in the descending order of their overall prevalence as Haemonchus spp. (44.70%), Nematodirus spp. (40%), Trichuris spp. (37.64%), Strongyloides spp. (34.11%) Trichostrongylus spp. (28.23%) Moniezia spp. (23.52%) and Fasciola spp. (20%). The mean EPG (eggs per gram) of different parasites showed a considerable variation in both the wild ungulates. The highest mean EPG was that of Haemonchus spp. and the lowest mean EPG was that of Fasciola spp. in both hosts. A statistically significant difference was observed in the mean EPG of different parasites between two wild hosts (t = 3.606, p = 0.01).

Keywords Wild ungulates · Helminth parasites · Prevalence · EPG · Mixed infection · Hirpora wildlife sanctuary

Introduction

Understanding gastrointestinal parasitic (GIT) diversity is of crucial importance for management of wild animals as these parasites pose a threat to the populations of different wild ungulates (Daszak et al. 2000) and play a vital role in survival of some major mammalian taxa (Lafferty and Gerber 2002). Helminth parasitic load is considered as one of the crucial factors responsible for the decline of certain wild ungulates (Morgan et al. 2006) as it can cause major scale mortality of hosts (Borghare et al. 2009). Gastrointestinal parasites are preferably present in duodenum, ileum, caecum and large intestines (Soulsby 1982). These parasites have adapted themselves according to the feeding habits of the host, besides the continuous physiological changes related to feeding (Leonard 1987). These parasites drain the essential nutrients from the host rendering them weak and making them prone to other diseases (Loukopoulos et al. 2007). The parasites of the gastrointestinal system pass the infectious propagules with the faecal matter that requires a maturation period outside the body of host resulting in accumulation of the infectious agents in...
the habitat increasing their chances of transmission (Soulsby 1982).

The preliminary data on gastrointestinal parasitic infestation of hangul deer (*Cervus hanglu hanglu*) is available from Kashmir (Lone et al. 2016; Khurshid et al. 2021), but the baseline information on other wild ungulates is lacking. Keeping in view the global status of markhor *Capra falconeri* (Near Threatened, IUCN 2021) and musk deer *Moschus* spp. (Endangered, IUCN 2021), the present study was undertaken to examine the status of gastrointestinal helminth parasites of these two threatened mountain ungulates in the Hirpora Wildlife Sanctuary (HWLS) of Kashmir Himalaya for designing a future road map in parasitic disease monitoring and control in wildlife habitats.

**Materials and methods**

**Study area**

The study was conducted in HWLS (33°39’55” N and 73°39’40” E) located at a distance of 70 km from Srinagar in Shopian district of Kashmir at an altitude of 2546 mts (Ahmad et al. 2014) (Fig. 1). HWLS is surrounded by Lake Gumsar on north, Hirpora village on northeast, Rupri on east, Saransar on south and Pir Panjal pass on west. The steepness of mountain cliffs of HWLS varies, some of the peaks are very steep while as the others are moderately steep. The HWLS consists of different vegetation types like mixed coniferous forest, deciduous sub-alpine scrub forest and sub-alpine pastures. The sanctuary is known for Himalayan musk deer and Pir Panjal markhor (Ahmad et al. 2014). The sanctuary remains under the heavy grazing pressure of domestic livestock belonging to local shepherds, migratory herders from June to October (Bhat et al. 2019).

**Climate**

The climate of Kashmir valley is a typical warm temperate. The average temperature ranges from 30 °C (maximum) and 20 °C (minimum) in June–July to 4 °C (maximum) and −9 °C (minimum) in December–January. The absolute humidity ranges from 50 to 80% throughout the year and drops to about 55% at night during the winter and 64% during the summer. The whole valley remains covered with snow in winter. On the basis of temperature and

**Fig. 1** Map showing the study area (Hirpora Wildlife Sanctuary) and the distribution of sampling points of two hosts.
precipitation, four seasons are recognized in a year in Kashmir valley: winter (December to February), spring (March to May), summer (June to August) and autumn (September to November) (Dar et al. 2002).

Sample collection

During the period of investigation systematic surveys were carried in HWLS for the collection of samples. Fresh faecal pellets of markhor and musk deer were randomly collected on weekly basis from November to May of 2018/2019. In order to avoid contamination of samples, only that portion of the faecal sample was collected which was not in contact with the soil. Faecal pellets were placed in collection vials and zip lock bags which were carefully labelled with animal identification, species, date and place of collection along with GPS coordinates. In order to preserve the parasitic eggs/oocysts 4% formalin was added to collected samples. Preserved samples were transported to the Parasitology Research Laboratory, Department of Zoology, University of Kashmir where faecal samples were microscopically examined for helminth eggs and larvae using concentration methods by Zinc sulphate flotation, faecal sedimentation and Modified McMaster techniques (Soulsby 1982). Morphological characters like shape, size and color were used for identification (Soulsby 1982).

Data analysis

Percentages with their respective means ± SD (standard deviation) were employed to calculate the prevalence of helminth parasites. Eggs were estimated as number of eggs per gram of faeces with their respective median and percentile. Chi-square test was used to evaluate the significance in the EPG among different helminth species of two hosts. The data was analyzed using Statistical packages MINITAB software version 13.2 and SPSS-17 for windows. Confidence level was held at 95% and P < 0.05 for significance.

Results

The results revealed multiple combinations of various helminth parasites in markhor and musk deer. Out of 44 samples of musk deer and 41 samples of markhor examined, 21(47.72%) samples of musk deer and 19 (46.34%) samples of markhor were positive for one or more than one species of GIT helminth parasites with an overall prevalence of 47.05%. Nematodes were more prevalent in both the species of wild ruminants (Table 1). In musk deer the different helminth parasites recorded are arranged in the descending order of their prevalence as Haemonchus spp. (45.45%), followed by Nematodirus spp. (40.90%), Trichuris spp. (38.63%), Strongyloides spp. (34.09%), Trichostrongylus spp. 29.54%), Moniezia spp.(25%) and Fasciola spp.(20.45%). The most prevalent parasite in markhor was Haemonchus spp. (43.90%), followed by Nematodirus spp. (39.02%), Trichuris spp. (36.58%), Strongyloides spp. (34.14%), Trichostrongylus spp. (26.82%), Moniezia spp. (21.95%) and Fasciola spp. (19.51%) (Table 2). Haemonchus was the most prevalent and Moniezia the least prevalent parasite in both hosts. A considerable variation was observed in the mean EPG among different parasites and between two hosts. (Table 2). In case of musk deer the highest mean EPG (1605.14 ± 416.50) was that of Haemonchus spp. followed by Nematodirus spp. (1370.48 ± 267.27), Trichuris spp. (1337.57 ± 190.68), Strongyloides spp. (1225.14 ± 221.29) Trichostrongylus spp. (1196.64 ± 234.03), Moniezia spp. (1145.60 ± 87.17) and Fasciola spp. (732.61 ± 111.86). Similarly, in case of markhor the highest mean EPG recorded was that of Haemonchus spp.(1390.86 ± 131.76) followed by Nematodirus spp. (1338 ± 119.25), Trichuris spp. (1266.66 ± 102.34), Strongyloides spp. (997.21 ± 284.34), Trichostrongylus spp.(899.41 ± 195.45), Moniezia spp. (590.47 ± 123.03) and Fasciola spp.(225.39 ± 131.76). A statistically significant difference was observed in the mean EPG of different parasites among two wild hosts (t = 3.606, p = 0.01). Most of the positive samples of both the hosts were infected with the mixed infection of two or more than two species of parasites. Among 21 positive samples of musk deer, 19 samples (90%) (Table 4) were infected with mixed parasites and two with single parasite (one with Haemonchus spp. and one with Strongyloides spp. Similarly, among 19 positive samples of markhor, mixed infection was reported in 16 samples (84.21%) (Table 3) and single infection in three samples (two samples with Haemonchus spp. and one with Trichuris spp).

Discussion

Parasite infection and transmission from domestic livestock to wild ungulates is a concern for the conservation of threatened wild ungulates as they share habitat atleast seasonally with livestock in Kashmir Himalayas. The livestock in these habitats has been reported to have a high parasite infection (Bhat et al. 2019) but the transmission of such infection to wild ungulates has been largely unexplored except a few preliminary studies by Lone et al.(2016) and Khurshid et al. (2021). Our study shows that almost all the parasitic species reported for the two wild ungulate species have been seen in the domestic livestock that uses the study area seasonally (Bhat et al.
The changing environmental conditions and utilization of natural habitats for livestock grazing increase the risk of cross-transmission of parasites (Gottdenker et al. 2014). These parasites are reported by different workers in the captive wild animals from different areas (Fagiolini et al. 2010; Thwait et al. 2014). The prevalence of nematodes was high as compared to that of cestodes and trematodes. The highest prevalence of nematodes is because most of the nematodes reported in the study have a direct life cycle and do not involve intermediate hosts for transmission, which may account for their high transmission rate and prevalence (Soulsby 1982). This is in close agreement with several studies (Mckenzie and Davidson 1989; Pacon 1994; Shibashi et al. 2003; Santin et al. 2004) who have also reported higher prevalence of nematodes than trematodes and cestodes. Among the nematodes the most prevalent parasites recorded in these wild hosts were Haemonchus spp. which is in close approximation with the studies of several researchers of the world (Cook et al. 1979; Mason 1994; Naseimento et al. 2000). The low prevalence of cestodes and trematodes is because of limited availability of intermediate hosts which are important for their propagation (Soulsby 1982).

### Table 1: Prevalence of nematodes, cestodes and trematodes in Markhor and Musk deer in Hirpora Wildlife Sanctuary

| Host       | Number examined(n) | No. of positive samples | Nematodes (prevalence) | Cestodes (prevalence) | Trematodes (prevalence) |
|------------|--------------------|-------------------------|------------------------|-----------------------|-------------------------|
| Musk deer  | 44                 | 21                      | 21(47.72%)             | 11(25%)               | 9(20.45%)               |
| Markhor    | 41                 | 19                      | 18(43.90%)             | 9(21.95%)             | 8(19.51%)               |
| Overall    | 85                 | 40                      | 39(63.42%)             | 20(33.43%)            | 17(30.29%)              |

### Table 2: Prevalence and mean EPG of different parasites in Markhor and Musk deer in Hirpora Wildlife Sanctuary

| Parasite | Markhor (n = 41) | Mus Deer (n = 44) | Overall prevalence (%) |
|----------|------------------|-------------------|------------------------|
| Haemonchus spp. | 18(43.90%) | 1390.86 ± 131.76 | 20(45.45%) | 1605.14 ± 416.50 | 44.70 |
| Nematodirus spp.  | 16(39.02%) | 1338 ± 119.25 | 18(40.90%) | 1370.48 ± 267.27 | 40 |
| Trichuris spp. | 15(36.58%) | 1266.66 ± 102.34 | 17(38.63%) | 1337.57 ± 190.68 | 37.64 |
| Strongyloides spp. | 14(34.14%) | 997.21 ± 284.34 | 15(34.09%) | 1225.14 ± 221.29 | 34.11 |
| Trichostrongylus spp. | 11(26.82%) | 899.41 ± 195.45 | 13(29.54%) | 1196.64 ± 234.03 | 28.23% |
| Monezia spp. | 09(21.95%) | 590.47 ± 123.03 | 11(25%) | 1145.60 ± 87.17 | 23.52% |
| Fasciola spp. | 08(19.51%) | 225.39 ± 131.76 | 09(20.45%) | 732.61 ± 111.86 | 20% |

**t = 3.606, p = 0.01**

### Table 3: Mixed infection of parasites in musk deer in Hirpora Wildlife Sanctuary

| Multiple parasites | +ve samples (%) |
|--------------------|-----------------|
| Haemonchus spp., Trichuris spp., Nematodirus spp., Strongyloides spp., Monezia spp. | 6 (28.57) |
| Trichuris spp., Fasciola spp., Haemonchus spp., Nematodirus spp., Trichostrongylus spp. | 5 (23.5) |
| Trichuris spp., Monezia spp., Haemonchus spp., Nematodirus spp., Trichostrongylus spp. | 4 (19.04) |
| Trichostrongylus spp., Haemonchus spp., Nematodirus spp., Strongyloides spp., Fasciola spp. | 2 (9.52) |
| Fasciola spp., Trichuris spp., Haemonchus spp., Strongyloides spp., Nematodirus spp., Trichostrongylus spp. | 1 (4.76) |
| Haemonchus spp., Trichuris spp., Strongyloides spp., Trichostrongylus spp., Fasciola spp., Monezia spp. | 1 (4.76) |
| Total | 19 (90) |

2019). The changing environmental conditions and utilization of natural habitats for livestock grazing increase the risk of cross-transmission of parasites (Gottdenker et al. 2014). These parasites are reported by different workers in the captive wild animals from different areas (Fagiolini et al. 2010; Thwait et al. 2014). The prevalence of nematodes was high as compared to that of cestodes and trematodes. The highest prevalence of nematodes is because most of the nematodes reported in the study have a direct life cycle and do not involve intermediate hosts for transmission, which may account for their high transmission rate and prevalence (Soulsby 1982). This is in close agreement with several studies (Mckenzie and Davidson 1989; Pacon 1994; Shibashi et al. 2003; Santin et al. 2004) who have also reported higher prevalence of nematodes than trematodes and cestodes. Among the nematodes the most prevalent parasites recorded in these wild hosts were Haemonchus spp. which is in close approximation with the studies of several researchers of the world (Cook et al. 1979; Mason 1994; Naseimento et al. 2000). The low prevalence of cestodes and trematodes is because of limited availability of intermediate hosts which are important for their propagation (Soulsby 1982). Similar results have been
reported in several other studies world across (Vengust 2003; Chroust and Chroustova 2004).

The presence of parasites such as *Haemonchus* spp., *Moniezia* spp. and *Trichuris* spp. have been also recorded to infect wild ungulates across the globe (Suzy et al. 1987; Rana et al. 2015; Bajwa et al. 2019; Eslami et al. 1981). The high prevalence of *Haemonchus* spp. and *Nematodirus* spp. revealed in the study is because the infectious agents of these species have the tendency to withstand the harsh (dry and cold) environment for a long period (Poole 1956). *Moniezia* spp. reported in the study had already been reported in cervids of Pakistan. The EPG in the present study is low as compared to the EPG recorded by Lone et al. (2016) in hangul deer (*Cervus elaphus hanglu*) in Dachigam National Park Kashmir, India. This may be due to low group size, scattered population and less congregation of wild ungulates in HWLS. The EPG of different parasites showed considerable variation in two hosts and between different parasites. The highest egg count was that of *Haemonchus* spp. and lowest of *Fasciola* spp. This is because of the fact that production of eggs by the parasite depends on several factors which includes variation in the number of eggs produced by different parasites, individual host immunity and the severity of infection (Hudson et al. 1992; Zajac and Conboy 2012). Some of the parasites like *Haemonchus* spp. are prolific and are capable of producing 5000 eggs per day while as others like *Trichostrongylus* produces only few hundred (Stein et al. 2002a, b). The mixed infection in most of the infected samples and the spectrum of helminths identified in the current study in which *Haemonchus* spp. was most prevalent and is in agreement with previous studies conducted in different parts of the world (Ezenwa 2003; Waruira et al. 2005; Obanda et al. 2019).

The parasitic infestation decreases the response of the hosts towards secondary infection by causing immune suppression (Abee et al. 2012). The gastrointestinal helminth fauna drain the essential nutrients rendering the animals weak, which affects the reproductive potential and survival rate of hosts (Toft 1982; Panayotova- Pencheva 2013). The helminth parasites lower the body mass, reduce fecundity and increase mortality among wild ungulates (Stein et al. 2002a, b). The nematodes may significantly affect the general health of endangered animals by causing various morbidities and mortalities and play a vital role in population regulation of wild ungulates (Roche 1988). *Strongyloides* spp. cause several ill effects like diarrhea, anorexia, weight loss, anemia and dyspnea (Gupta et al. 2011). *Haemonchus* spp. is known to cause intestinal ulceration leading to loss of blood, which makes the host weak and susceptible to other diseases besides causing significant morbidity and mortality (Davidson et al. 1980). The parasitic load can cause the death of hosts in severe cases which results in their population decline (Zhang et al. 2008). Intestinal parasitosis is known to cause disease and death in some wild hosts as reported by several workers (Solórzano-García and Pérez-Ponce de León 2017). The present study needs further elaboration for understanding the impact of GIT helminth parasites of the disease dynamics of threatened mountain ungulates in Kashmir.

### Conclusion

The presence of gastrointestinal helminth parasites in markhor and musk deer with known ill impacts may threaten the survival of these two threatened mountain ungulates. This study has opened a vision for monitoring and managing of wildlife parasitic disease in livestock dominated protected areas. The livestock entering the wildlife protected areas should be screened and vaccinated to reduce the level of infection and transmission to wild ungulates. Detailed investigation of the intensity of

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**Table 4** Mixed infection of parasites in Markhor in Hirpora Wildlife Sanctuary

| Host: Markhor | Multiple parasites | + ve samples (%) |
|--------------|--------------------|------------------|
| *Haemonchus* spp., *Trichuris* spp., *Nematodirus* spp., *Fasciola* spp., *Trichostrongylus* spp., *Strongyloides* spp., *Moniezia* spp. | 5 (26.31) |
| *Trichuris* spp., *Fasciola* spp., *Haemonchus* spp., *Nematodirus* spp., *Trichostrongylus* spp., *Strongyloides* spp | 3 (15.75) |
| *Trichuris* spp., *Moniezia* spp., *Haemonchus* spp., *Strongyloides* spp., *Nematodirus* spp. | 3 (15.75) |
| *Trichostrongylus* spp., *Haemonchus* spp., *Trichuris* spp., *Nematodirus* spp., *Strongyloides* spp. | 2 (10.52) |
| *Trichostrongylus* spp., *Haemonchus* spp., *Nematodirus* spp., *Moniezia* spp. | 1 (5.26) |
| *Nematodirus* spp., *Haemonchus* spp., *Trichuris* spp., *Strongyloides* spp. | 1 (5.26) |
| *Nematodirus* spp., *Haemonchus* spp. | 1 (5.26) |
| Total | 16 (84.21) |
infection and evaluation of mode of transmission at length in future is needed for the strengthening the management of the protected area and the threatened wild ungulates.

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Declarations

Conflict of interest The authors declare no competing interests. Equipment brands, chemicals and other trade names are mentioned here solely for the convenience of the reader and imply no endorsement by the authors.

Ethics approval All procedures and methods used in this study regarding the use and care of animals were carried out in accordance with accepted international animal use and care guidelines.

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