



Article

Gastrointestinal Nematodes and Protozoa in Small and Large Ruminants from Rural Agro-Climatic Regions of Northern India

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Abstract: Gastrointestinal nematode parasites and gastrointestinal protozoan parasites are considered detrimental to the livestock population and manifest production-limiting effects. Small and large ruminants (cattle, buffalo, goats, and sheep) are important components of the rural economy of northern India. However, the epidemiology of gastrointestinal parasites in this agro-climatic region has not been studied extensively. In this study, the prevalence of gastrointestinal parasites was determined in 163 animals, including cattle ($n = 86$), buffalo ($n = 11$), goats ($n = 48$), and sheep ($n = 18$) from 26 sampling sites by copro-parasitological analysis. The prevalence values of 94.47% and 66.87% were recorded for the nematodes and protozoa, respectively. The group-wise prevalence of gastrointestinal nematode parasites was 95.3%, 90.9%, 93.7%, and 94.4% in cattle, buffalo, goats, and sheep, respectively, whereas for gastrointestinal protozoan parasites, the respective values were 70.9%, 54.5%, 60.4%, and 72.2%. Copromicroscopy revealed ten genera of nematodes—*Ascaris*, *Capillaria*, *Cooperia*, *Haemonchus*, *Nematodirus*, *Oesophagostomum*, *Ostertagia*, *Strongyloides*, *Trichostrongylus*, *Trichuris*, and one protozoan genus—*Eimeria*. The prevalence of *Trichostrongylus* spp. was highest in buffaloes, whereas in cattle, *Ascaris* spp. were predominant. In both goats and sheep, *Haemonchus contortus* was found to be predominant. The highest prevalence of gastrointestinal parasites was recorded in the rainy season. These findings indicate the prevalence of gastrointestinal parasites in the ruminant population in this region and necessitate the implementation of preventive and control strategies for effective animal health management.

Keywords: helminth parasites; ruminants; livestock; gastrointestinal nematode parasites; gastrointestinal protozoan parasites



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1. Introduction

Livestock farming remains the backbone of the rural economy in India [1–3]. It is recognized as the most important sub-sector of Indian agriculture and supports the basic needs and income of rural households in most parts of the country especially of northern hilly states [2,4–7]. According to the 20th Livestock Census 2019 report, the total livestock population in India is 536.76 million, of which 95.78% are from rural areas of the country [8]. The total number of cattle, buffaloes, goats, and sheep is 193.46 million, 109.85 million, 148.88 million, and 74.26 million which represent 36.04%, 20.47%, 27.74% and 13.83% of the total livestock population, respectively [8]. In Himachal Pradesh, the total livestock population is 4.41 million and constitutes an essential component of the livelihood of the rural population [8].

Helminth parasites represented by Platyhelminthes and Nematelminthes are considered a serious health risk for livestock worldwide [9–12]. These are responsible for distress, debilitating diseases, malnutrition, anemia, stunted growth, and allergic conditions in domesticated livestock and grazing animals [3,13–16]. Their parasitism results in reduced milk yields, decreased wool production, and low growth in most grazing herds and flocks. Overall, helminth parasites exert a significant impact on animal health, livestock productivity, the income of livestock owners, and food security [12,16]. Helminth parasites manifest their adverse effects on both small and large ruminants. Grazing ruminants are more severely impacted as compared to those kept in animal shed conditions [5,17,18]. The most widely prevalent gastrointestinal nematode parasite genera in domesticated ruminants are *Ascaris*, *Haemonchus*, *Strongyloides*, *Trichostrongylus*, *Cooperia*, *Ostertagia*, *Nematodirus*, *Trichuris*, *Capillaria*, and *Oesophagostomum*, among others [9,10,19–21]. Gastrointestinal protozoan parasites viz. *Balantidium*, *Eimeria*, and *Cryptosporidium* also infect ruminants, resulting in adverse effects on their health, productivity, and reproduction [22–26]. It is generally observed that tropical and sub-tropical conditions, high humidity, damp soil, high temperature, and a cold environment offer optimum conditions for the survival, multiplication, and transmission of gastrointestinal nematode parasites and gastrointestinal protozoan parasites [24]. Usually, these parasitic infestations of domesticated ruminants are seasonal in nature, with a higher frequency in the monsoon season and lower infection rates in the summer [27–29]. There is a considerable role of animal husbandry practices and conditions, i.e., animal sheds, floor conditions, crowding, cleaning routines, deworming practices, etc., in the transmission of veterinary parasitosis [10,30]. Further, helminthiasis of ruminants are influenced by immunity level, feeding pattern, water supply, grazing habits, humidity, temperature, rainfall, vegetation, husbandry facilities, and deworming practices [17,27,31]. This study was planned to determine the prevalence of gastrointestinal nematode parasites and gastrointestinal protozoan parasites in cattle, buffalo, goats, and sheep domesticated in the mountainous rural region of Himachal Pradesh, India.

2. Materials and Methods

2.1. Study Area

The study was carried out from February 2019 to July 2019 in the Sirmaur district of Himachal Pradesh, situated in the outer Himalayan Shivalik range of northern India (Figure 1). The study involved 26 villages (sampling sites) from five tehsils within district Sirmaur (Table 1).

Table 1. Sampling sites located in the Sirmaur district of the northern Indian state of Himachal Pradesh from where fecal samples of domesticated animals were collected and analyzed to determine the prevalence of gastrointestinal nematode parasites and gastrointestinal protozoan parasites.

S. No.	Sampling Site	Tehsil	Number of Study Animals			
			Cattle	Buffaloes	Goats	Sheep
1.	Kolan	Rajgarh	2	0	1	0
2.	Nehar Bag	Rajgarh	2	0	2	0
3.	Choki	Rajgarh	2	2	0	0
4.	Kot	Rajgarh	1	3	2	0
5.	Deedag	Rajgarh	2	0	2	0
6.	Chichhrea	Rajgarh	2	0	1	0
7.	Jalag	Rajgarh	1	0	0	0
8.	Redigusan	Rajgarh	5	0	0	0
9.	Lana Machher	Pachhad	13	0	6	6
10.	Kakli	Pachhad	6	0	1	1
11.	Kheri	Pachhad	8	1	3	0

Table 1. Cont.

S. No.	Sampling Site	Tehsil	Number of Study Animals			
			Cattle	Buffaloes	Goats	Sheep
12.	Lana Bhalta	Pachhad	3	0	2	0
13.	Baru Sarera	Pachhad	10	1	3	2
14.	Lana Madag	Pachhad	4	0	2	0
15.	Manjithi	Pachhad	3	0	0	1
16.	Bongli Kech	Pachhad	2	1	1	3
17.	Bagroti	Pachhad	2	0	3	1
18.	Kotla Molar	Renuka	2	0	3	2
19.	Parara	Renuka	2	0	2	0
20.	Bachar Ka Bag	Dadahu	2	3	2	1
21.	Chaknal	Dadahu	2	0	2	0
22.	Nohra Dhar	Nohra	2	0	1	0
23.	Kapradi	Nohra	3	0	2	0
24.	Bharadi	Nohra	2	0	2	0
25.	Bogdhar	Nohra	2	0	2	1
26.	Kitta	Nohra	1	0	3	0
			86	11	48	18

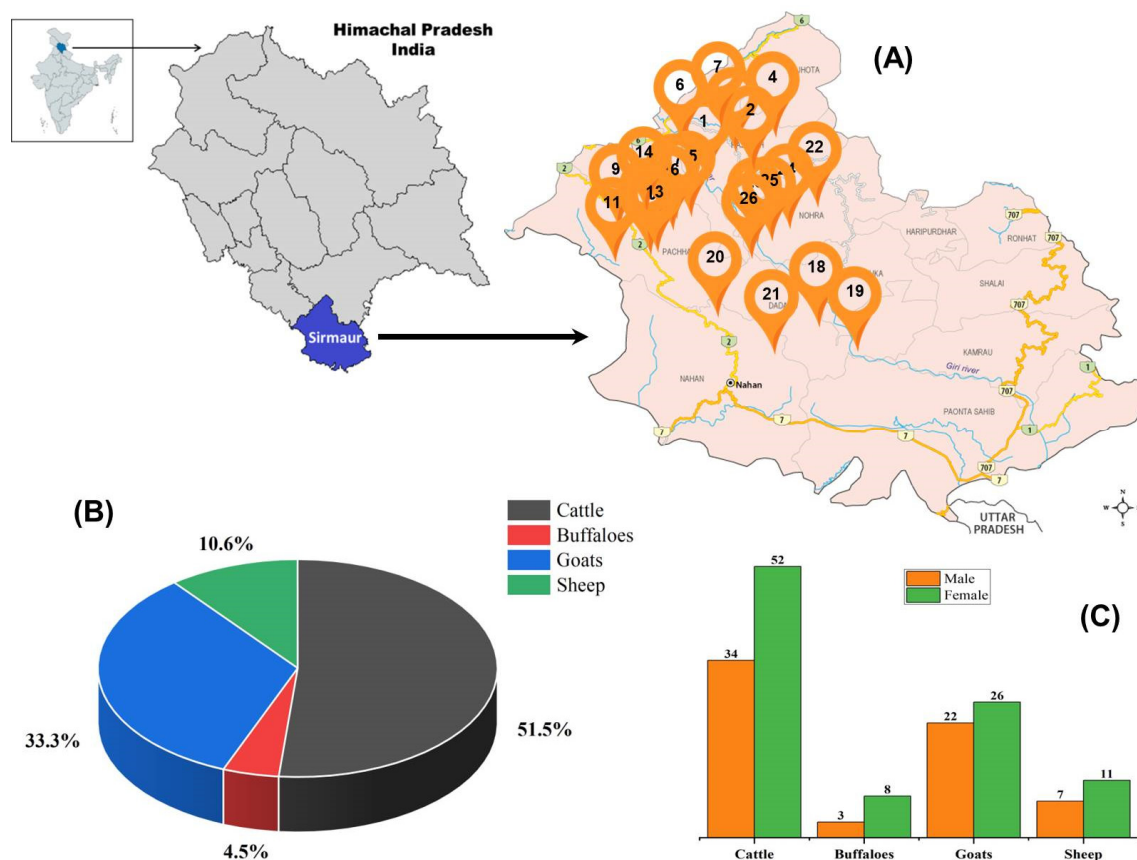


Figure 1. Geographical location, sampling locations and animals studied. (A) Sampling sites located in five tehsils of rural tropical Himalayan regions of Himachal Pradesh, India. (B) represents the distribution of ruminants in numbers and percentage, and (C) represents the gender-wise distribution of study animals from which fecal samples were collected for parasitological analysis.

2.2. Study Animals Selection

The domesticated ruminants viz. cattle (*Bos Taurus* Linnaeus, 1758), buffalo (*Bubalus bubalis* Linnaeus, 1758), goat (*Capra hircus* Linnaeus, 1758), and sheep (*Ovis aries* Linnaeus, 1758) were selected randomly from the sampling locations as mentioned in Table 1. The study animals were from the following breeds: cattle—Jersey, Sahiwal and Holstein Friesian; buffalo—Murrah; goat—Chegu and Gaddi and sheep—Rampur bushair and Gaddi. During the collection of freshly laid fecal samples, the livestock owners were made aware of the study objectives and provided a printed short questionnaire requesting them to furnish some basic information, i.e., age of animals, gender, breed, animal husbandry practices, farm size, animal shed floor type (cement concrete or soil), feedstuffs, fodders, grazing routines, cleaning and sanitation, and deworming history. The personal details of the livestock owners were kept anonymous and confidential.

2.3. Fecal Sample Collection

Freshly laid fecal samples from the study animals were collected in sterile polyethylene bags or sterile disposable 100 mL containers, labeled, and immediately brought to the laboratory under refrigerated conditions. Samples were processed and analyzed on the same day. However, in the cases where fecal samples could not be analyzed on the same day, they were kept under refrigeration (2–4 °C) until further analysis the next day.

2.4. Qualitative Analysis by the Saturated Salt Flotation Method

Fecal sample processing and microscopic identification of different life stages of gastrointestinal nematode parasites and gastrointestinal protozoan parasites were performed as per standard procedures and veterinary parasitology manuals [9,32–34]. A saturated salt solution of sodium chloride (specific gravity: 1.18–1.2) was prepared by suspending 400 g in 1000 mL of distilled water, followed by stirring for 30–60 min. Similarly, a saturated zinc sulphate solution (specific gravity: 1.18) was prepared by mixing 371 g of zinc sulphate in 1000 mL of distilled water and then thorough mixing. The saturated salt solutions were stored at room temperature under ambient conditions until experimental use. Each fecal sample (3.0 g) was thoroughly mixed in a 50 mL saturated salt solution with a glass rod, and the suspension was strained through a tea strainer. The suspension so obtained was gently transferred into 15 mL sterile glass test tubes and placed in a straight position. A clean glass coverslip was placed on the top of the convex surface of the test tube in such a way that the solution was in direct contact with the coverslip. After 20–30 min, the coverslip was carefully removed with forceps and placed on a glass slide for microscopic observations under the 10× and 40× objectives of a bright-field compound light microscope (Dewinter Optical, Inc., New Delhi, India). The nematode eggs and protozoan cysts were photographed with a camera (Leica Microsystems, Switzerland) attached to the microscope, and their dimensions (length and width) were determined by image processing software. Gastrointestinal nematode parasites and gastrointestinal protozoan parasites identified from fecal samples of ruminants on the basis of the presence of eggs, cysts, and characteristic shape, size, coloration, operculum, polar plugs, anterior and posterior ends, presence of blastomeres, and outer sheath.

2.5. Quantitative Analysis of Fecal Samples

The quantitative estimation of eggs per gram (EPG) counts of animal fecal samples was determined by Stoll's dilution method and the modified Wisconsin sugar flotation method [9,32,34]. Briefly, a 3.0 g fecal sample was suspended in 42 mL of distilled water and thoroughly mixed with a glass rod. The fecal suspension was strained through a tea strainer into a clean beaker and left to stand for 5 min. A drop of the suspension was placed on a clean glass slide, covered with a cover slip, and examined under a bright-field compound light microscope (Dewinter Optical, Inc., New Delhi, India). EPG counts were calculated by the formula: EPG count = Number of eggs counted × 100. In the modified Wisconsin sugar flotation method, a 3.0 g fecal sample was suspended in 10 mL

of Sheather's solution, mixed with a spatula and strained through a tea strainer to remove large debris and feed particles. The suspension was centrifuged at $2000\times$ rpm for 5 min, then test tube was filled with Sheather's solution to form a meniscus and covered with a glass cover slip. After 5–10 min, the cover slip was removed, placed on a clean glass slide, and observed under a bright-field compound light microscope (Dewinter Optical, Inc., New Delhi, India) at $10\times$ and $40\times$ objectives. Parasite egg counts in fecal samples were calculated as EPG count = number of eggs counted/3. Quantitative estimation of EPG counts in fecal samples of study animals was performed from two sampling sites, i.e., Lana Machher and Bongli Kech, from February 2019 to July 2019.

2.6. Meteorological Data

The data related to mean rainfall, average high temperature, and average low temperature in Sirmaur district during the period from February to July 2019 were retrieved from the websites of the Indian Meteorological Department, Shimla <http://weathershimla.nic.in/index.html> (accessed on 31 July 2019), and the Accuweather website <https://www.accuweather.com/> (accessed on 31 July 2019). The rainfall recorded in the months of February, March, April, May, June, and July was 138.4 mm, 29.6 mm, 54.6 mm, 27.6 mm, 55.0 mm, and 236.4 mm, respectively. The average high temperature ranged from 21.4 to 44.6 °C and the average low temperature was between 10.1 to 27.8 °C during the study period with peak values in June.

2.7. Statistical Analysis

Experiments were performed in triplicate, and the data were expressed as mean \pm SD. The results were analyzed using MS-Excel (Microsoft).

3. Results

The copro-parasitological evaluation of the study animals revealed the prevalence of ten genera of gastrointestinal nematodes and one genus of parasitic protozoa. These were identified as *Ascaris* spp., *Capillaria* spp., *Cooperia* spp., *Haemonchus contortus* (Rudolphi, 1803) Cobb, 1898, *Nematodirus* spp., *Oesophagostomum* spp., *Ostertagia* spp., *Strongyloides* spp., *Trichostrongylus* spp., *Trichuris* spp., and *Eimeria* spp. (Figure 2). The parasite eggs and oocysts were identified on the basis of distinguishing characteristics such as the presence of polar plugs in the case of *Trichuris* sp., operculum in the case of *Eimeria* oocysts, and embryos in eggs in strongyles (Figure 2). The characteristic features of all the identified genera are given in Table 2. The genera-specific prevalence of gastrointestinal nematode parasites in cattle, buffaloes, goats, and sheep is shown in Figures 3 and 4.

Table 2. Morphological characteristics and morphometric details of gastrointestinal nematode parasites and gastrointestinal protozoan parasites identified from fecal samples of domesticated ruminants of rural areas of Sirmaur, Himachal Pradesh.

S. No.	Parasite Genera/Species	Family/Sub-Family	Dimensions (μm) Length \times Width	Characteristic Features *
1.	<i>Strongyloides</i> spp.	Strongyloididae	$87.2\ \mu\text{m} \times 47.6\ \mu\text{m}$	Oval, rounded ends
2.	<i>Trichostrongylus</i> spp.	Trichostrongylidae	$85.2\ \mu\text{m} \times 41.2\ \mu\text{m}$	Oval/oblong
3.	<i>Ostertagia</i> spp.	Trichostrongylidae	$78.6\ \mu\text{m} \times 45.1\ \mu\text{m}$	Oval, long shape
4.	<i>Nematodirus</i> spp.	Trichostrongylidae	$180.7\ \mu\text{m} \times 88.4\ \mu\text{m}$	Oval/oblong
5.	<i>Trichuris</i> spp.	Trichuridae	$75.2\ \mu\text{m} \times 35.5\ \mu\text{m}$	Ellipsoidal, two polar plugs
6.	<i>Haemonchus contortus</i>	Trichostrongylidae	$80.6\ \mu\text{m} \times 45.8\ \mu\text{m}$	Round/oval, blastomeres
7.	<i>Capillaria</i> spp.	Trichuridae	$47.2\ \mu\text{m} \times 30.6\ \mu\text{m}$	Oval, lemon-shaped

Table 2. Cont.

S. No.	Parasite Genera/Species	Family/Sub-Family	Dimensions (μm) Length \times Width	Characteristic Features *
8.	<i>Cooperia</i> spp.	Trichostrongylidae	75.8 μm \times 34.6 μm	Ovoid to ellipsoid
9.	<i>Oesophagostomum</i> spp.	Chabertiidae	76.2 μm \times 38.1 μm	Oval, with blastomeres
10.	<i>Ascaris</i> spp.	Ascarididae	44.3 μm \times 73.7 μm	Oval, wavy/folded surface
11.	<i>Eimeria</i> spp.	Eimeriidae	25.8 μm \times 41.8 μm	Ovoid or ellipsoid oocysts

* Parasite life stages were identified on the basis of distinguishing characteristics described in veterinary parasitology manuals and identification keys [9,32–34].

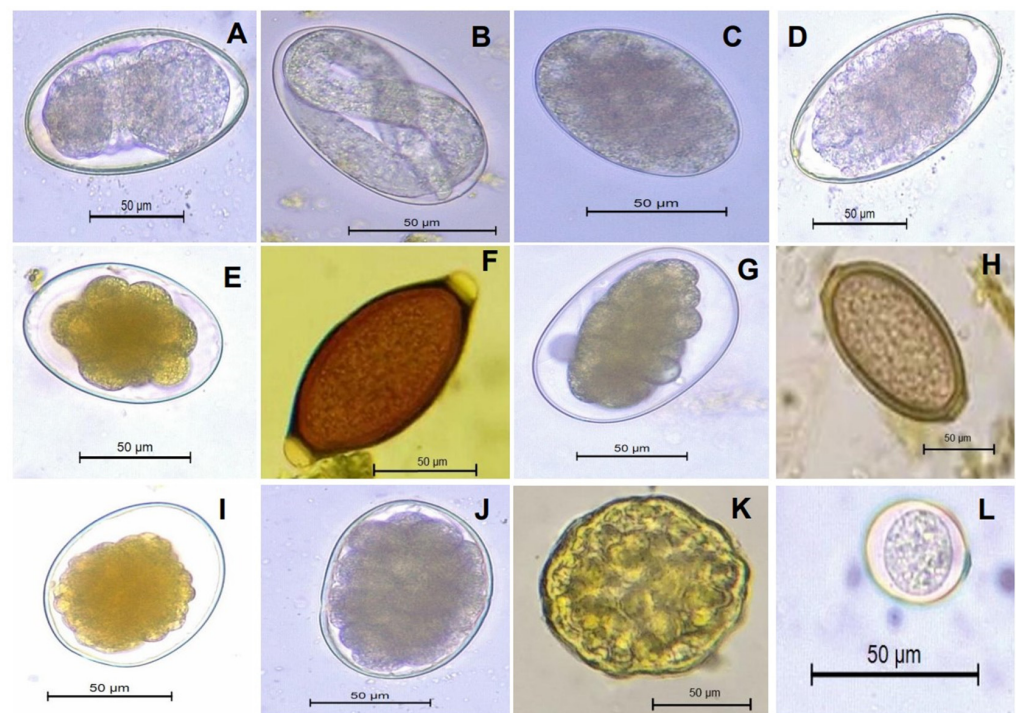


Figure 2. Morphological characteristics of diagnostic stages (eggs and oocysts) of gastrointestinal nematode parasites and gastrointestinal protozoan parasites observed by coprological analysis of the fecal matter of domesticated ruminants. (A,B) *Strongyloides* spp., (C) *Trichostrongylus* spp., (D) *Ostertagia* spp., (E) *Nematodirus* spp., (F) *Trichuris* spp., (G) *Haemonchus contortus*, (H) *Capillaria* spp., (I) *Cooperia* spp., (J) *Oesophagostomum* spp., (K) *Ascaris* spp., and (L) *Eimeria* spp. The fecal samples were observed at 400 \times magnification after processing and photomicrographs were morphometrically analyzed. Scale bar = 50 μm .

The predominant copro-prevalence of *Ascaris* spp. (90%) and *Trichuris* spp. (87%) was observed in cattle (Figure 4). The prevalence of other parasite species ranged between 16.2 and 40.6% (Figure 3). In the case of buffalo, the prominent gastrointestinal nematode parasites were *Trichostrongylus* spp. (90%), *Capillaria* spp. (81.8%), *Strongyloides* spp. (81.8%), and *Trichuris* spp. (72.7%). As compared to cattle and buffalo, the dominant nematode parasite in the gastrointestinal tracts of goats was *Haemonchus contortus*, with a 95.8% prevalence. This was followed by *Strongyloides* spp. (89.5%) and *Trichostrongylus* spp. (83.3%). However, the prevalence of *Ascaris* spp. and *Trichuris* spp. was lower as compared to that of cattle (Figures 3 and 4). Similarly, in sheep, *H. contortus* was detected in the majority of the animals which corresponds to a 94.4% prevalence. The lowest prevalence was of *Nematodirus* spp. (27%), whereas that of *Ascaris* spp. was 66.6% (Figure 3). In total, 154 ruminants (94.47%) showed the prevalence of nematode life stages (eggs and early-stage larvae) in their fecal matter. In comparison, the overall prevalence of gastrointestinal

protozoan parasites was 66.87%. As depicted in Figure 5A, the prevalence of gastrointestinal nematode parasites in ruminants ranged between 90.9 and 95.3% with cattle being the highest among all the groups. On the other hand, the gastrointestinal protozoan parasite prevalence was highest in sheep, i.e., 72.2%, followed by cattle (70.9%), goats (60.4%), and buffalo, i.e., 54.5%. Overall, it was observed that the protozoal infestation in ruminants was lesser as compared to gastrointestinal nematode parasites.

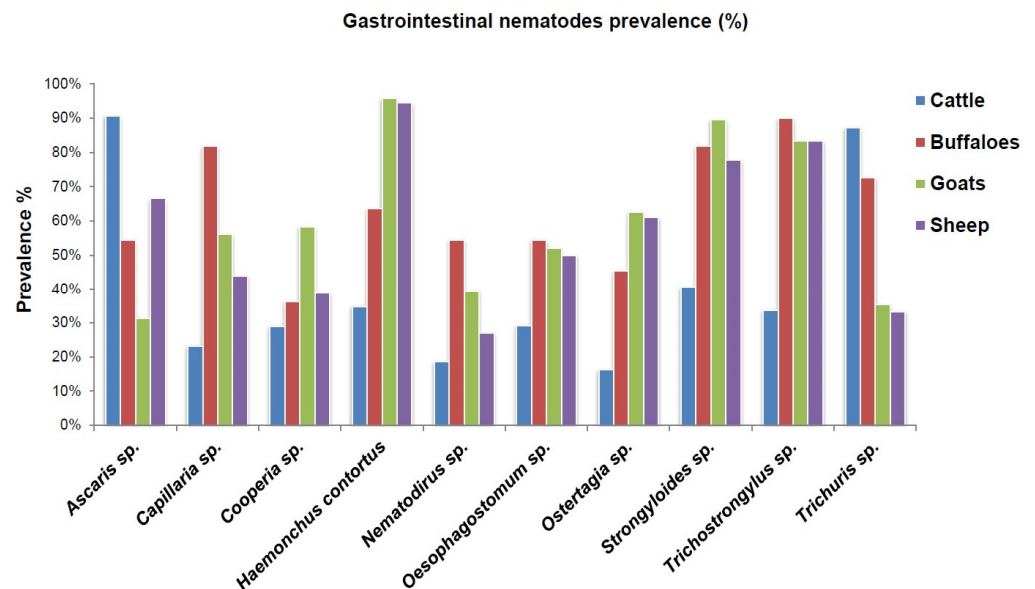


Figure 3. The copro-parasitological prevalence of gastrointestinal nematode parasites in cattle, buffaloes, goats, and sheep of rural areas of Himachal Pradesh, India.

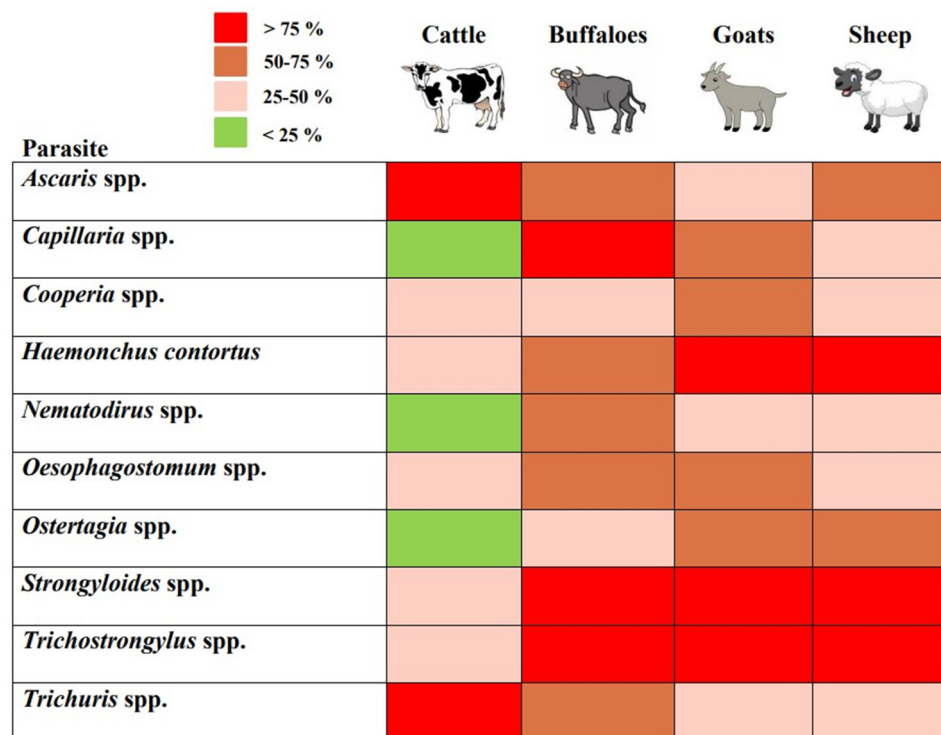


Figure 4. The heat map showing coprological predominance of gastrointestinal nematode parasites in cattle, buffaloes, goats, and sheep from the rural regions of Sirmaur, Himachal Pradesh. Here, rows represent nematode genera and column represents study animals.

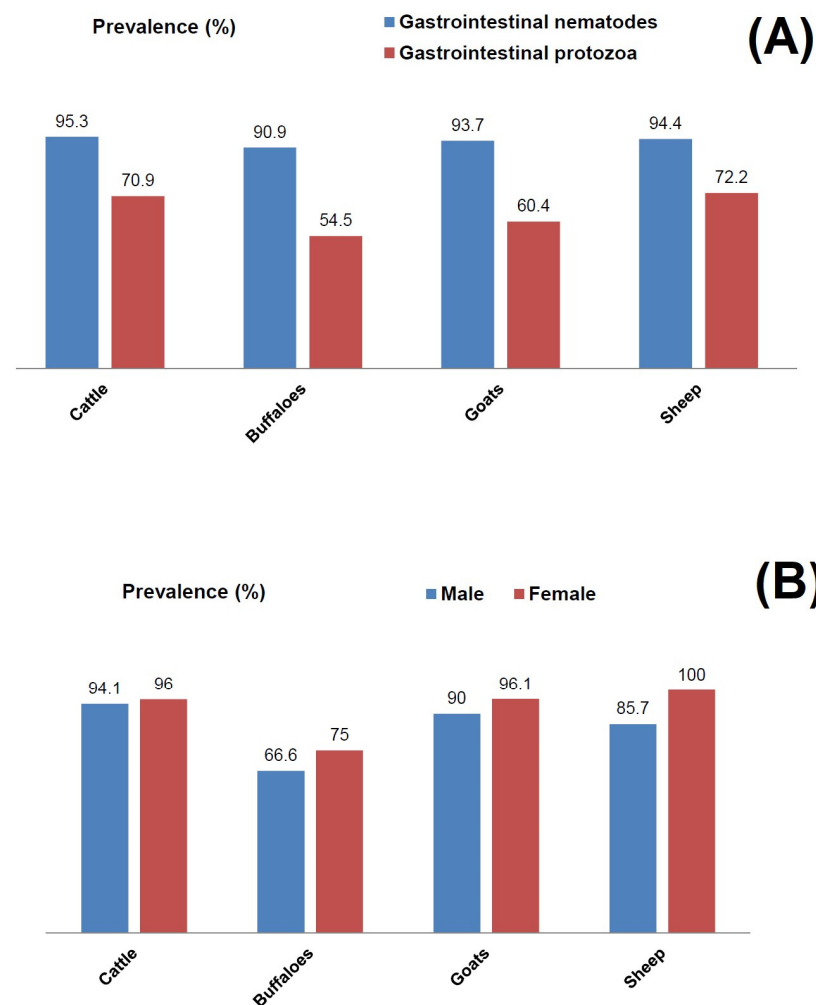


Figure 5. Prevalence of gastrointestinal nematode parasites and gastrointestinal protozoan parasites in domesticated ruminants. **(A)** Overall prevalence of gastrointestinal parasites in cattle, buffaloes, goats and sheep. **(B)** Gender-specific prevalence of gastrointestinal nematode parasites in fecal matters in male and female animals from the rural regions of Himachal Pradesh, India.

Fecal sample analysis of male and female ruminants revealed the presence of parasite eggs and cysts, but to a different extent (Figure 5B). In contrast to cattle, where both male and female animals had a comparable parasite prevalence, there was a higher parasite prevalence in female animals as compared to male counterparts in buffaloes, goats, and sheep. In the case of sheep, 85.7% of male animals had a positive gastrointestinal parasite prevalence, whereas all the female counterparts were positive for the prevalence of parasites (Figure 5B). A similar pattern was also observed with buffaloes and goats. As evident, the highest rainfall was recorded in July during the monsoon season and there were higher EPG counts in the study animals during this period. From the sampling site Bongali Kech, EPG counts in cattle were 27.5 ± 5.7 in February, which gradually increased to 72.5 ± 7.8 in the month of July (Figure 6). A similar increasing trend was observed in the cases of the other three animal groups. EPG counts were highest in the monsoon season, when rainfall and humidity were higher as compared to the winter and dry seasons. From the second sampling site Lana Machher, lower EPG counts were recorded in February and March, whereas 2–3-fold higher counts were observed in July.

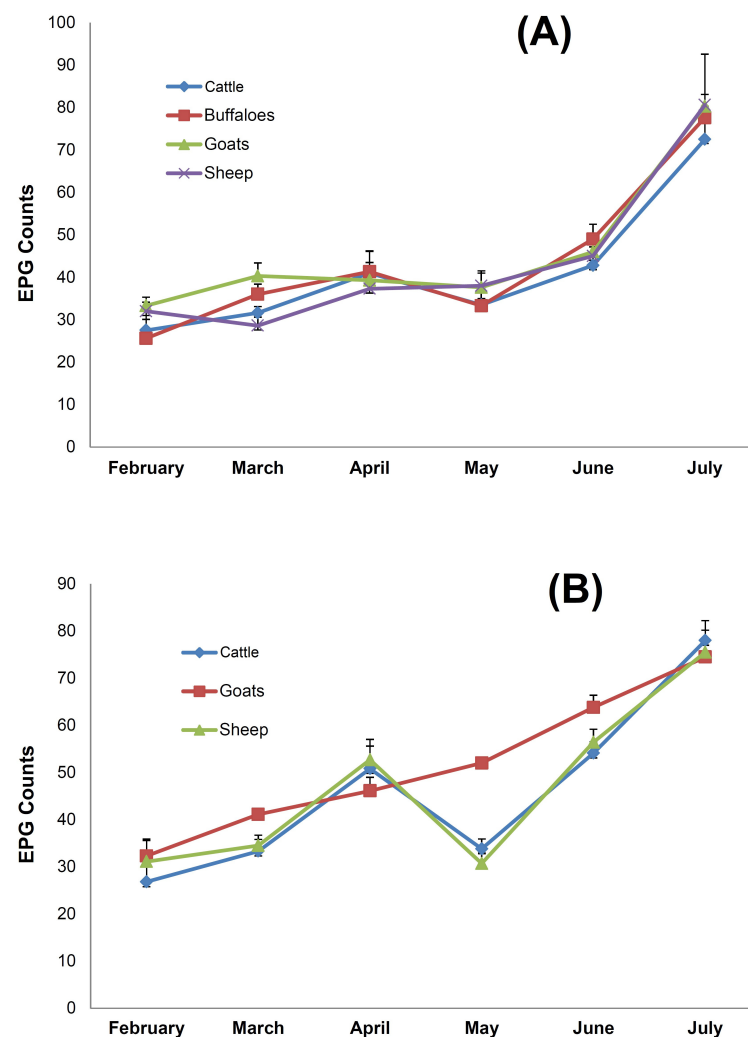


Figure 6. The correlation between meteorological parameters and eggs per gram (EPG) counts of gastrointestinal nematode parasites in domesticated ruminants. (A) Fecal EPG counts in ruminants from the sampling site Bongali Kech, and (B) fecal EPG counts in ruminants from the sampling site Lana Machher. Data represent the mean \pm SD of three independent experiments.

4. Discussion

Livestock farming is indispensable for the sustainability of the rural economy in India, including the northern state of Himachal Pradesh. However, gastrointestinal nematodes and gastrointestinal protozoa in domesticated ruminants are known to exert tremendous economic losses by adversely impacting animal health, productivity, yields, and reproduction [20,25,35–37]. Moreover, these infections are often neglected by livestock owners and result in significant economic losses in the long term [20]. The study areas of current investigation fall in the sub-mountain and low hills subtropical agro-climatic zone in the district of Sirmaur [1]. Fecal sample analyses of domesticated ruminants revealed an overall prevalence of 94.47% for gastrointestinal nematode parasites and 66.87% for gastrointestinal protozoan parasites. Ruminants were found to harbor a diverse community of gastrointestinal nematode parasites which include the species of *Ascaris*, *Trichuris*, *Strongyloides*, *Trichostrongylus*, *Haemonchus*, *Cooperia*, *Oesophagostomum*, *Ostertagia*, *Capillaria*, and *Nematodirus* as detected by coproscopical analysis. In terms of the genera-specific prevalence of gastrointestinal nematode parasites, the experimental data revealed the predominance of *Trichostrongylus* spp. and *Ascaris* spp. in buffaloes and cattle, respectively. The other three most dominant parasites in buffaloes were *Strongyloides* spp., *Capillaria* spp., and *Trichuris* spp., whereas *Strongyloides* spp., *Trichuris* spp., and *Haemonchus contortus* were among the

top four gastrointestinal nematode parasites in cattle. Jithendran [1] has also reported the predominance of the eggs of *Strongyloides* spp. in goats and sheep. Similarly, Choudhary et al. [38] observed the highest prevalence of *Trichostrongylus* spp. in cattle. A meta-analysis of the prevalence of gastrointestinal parasites in Indian buffaloes during the period 2011 to 2018 revealed a 46.67% average prevalence rate which was attributed to their low productivity over the years [39]. In the small ruminants, *Haemonchus contortus* had the highest prevalence (>94%) in both the goat and sheep populations. The other dominant parasites were *Trichostrongylus* spp. and *Strongyloides* spp. In other published reports, *H. contortus* was found to be predominant in both large and small ruminants' fecal contents [40–43]. Similarly, a high prevalence of *Haemonchus* spp., *Trichostrongylus* spp., *Bunostomum* spp., *Eimeria* spp. and *Strongyloides* spp. was reported from cattle in Ethiopia [44]. In the Andhra Pradesh state of India, a 39.1% prevalence of gastrointestinal nematodes was recorded in domestic ruminants with a predominance of *Haemonchus* and *Paramphistomum* [45]. In the case of gastrointestinal protozoan parasites, only one genus, i.e., *Eimeria* was identified from small and large ruminants. The incidence of *Eimeria* spp. to the extent of 11.4% in cattle and 4.7% in buffaloes was reported from the Gujarat state of India [46]. The prevalence of *Eimeria* spp. was also observed by Cruvinel et al. [47] and León et al. [48].

Our findings are in correlation with earlier reports by Choubisa and Jaroli [23], Singh et al. [49], Singh et al. [50], and Pinilla et al. [51], where the overall parasite prevalence ranged between 82 and 94% in small and large ruminants. Interestingly, Yusof and Isha [52] found an even higher prevalence, i.e., 89.2% of coccidiosis in a goat population of Malaysia. On the other hand, lower prevalence rates, i.e., 62.5% and 67.0% of helminth parasites, were reported by Jena et al. [53] and Shit et al. [54], respectively. Krishnamoorthy et al. [55] reported a high prevalence of gastrointestinal parasites in sheep (65%) and in goats (74%) in India. Their findings also indicated a higher parasite prevalence in goats from Himachal Pradesh. Marskole et al. [56] reported a higher parasite prevalence in cattle (75%) than buffaloes (70.4%). In contrast, Wadhwa et al. [57], Velusamy et al. [58], and Das et al. [43] observed a lower, i.e., 10–55%, prevalence of gastrointestinal nematode parasites and gastrointestinal protozoan parasites in ruminants. Similar reports on parasite prevalence rates are also available in the literature from different geographical regions of the world [36,37,59,60]. It was also observed that the female animals had a slightly higher infestation with gastrointestinal parasites as compared to their male counterparts. As compared to domesticated ruminants, the wild species of these animals usually exhibit a low parasite species diversity but higher intensity due to grazing habits, genetic factors, and variable population density [20]. Some possible explanations for the observed variability in parasite prevalence rates of gastrointestinal nematode parasites and gastrointestinal protozoan parasites in the study animals include geographical factors, agro-climatic conditions, grazing habits, deworming schedules, sampling procedures, procedural variations, and counting methods. Further, the contribution of animals' immunity, nutritional status, age, gender, and genetic makeup in imparting anti-gastrointestinal parasite resistance cannot be underestimated [20,35]. In previous reports, several species of gastrointestinal nematodes were detected from the ruminants of Himachal Pradesh and adjoining states, namely *Bunostomum trigonocephalum*, *Haemonchus contortus*, *Ostertagia circumcincta*, *Oesophagostomum columbianum*, *Trichostrongylus axei*, *T. colubriformis*, *Trichuris ovis*, and *Strongyloides papillosus* [1,61]. Among *Eimeria* species, *E. ninakohlyakimovae* was predominantly reported from goat populations [22]. The higher prevalence of gastrointestinal nematode parasites in fecal samples of buffaloes, cattle, goats, and sheep suggests that the agro-climatic conditions in the study area were supportive for parasite reproduction, development, and the survival of eggs and larvae inside water and soil. Another explanation for the higher parasite infestation in the ruminants is the poor animal husbandry practices adopted by the livestock owners in rural areas. The variability in the diversity of gastrointestinal parasites in cattle, buffaloes, goats, and sheep even though living in the same habitats and geographical areas might be due to a combination of environmental factors, host behavior, and host animals' intrinsic traits [20].

Previous studies carried out in the mountainous regions of northern Indian states revealed the occurrence of gastrointestinal nematode infestations in large and small ruminants. An extensive study by Jithendran [1] found 87.2% and 94.3% gastrointestinal helminths infection levels in cattle and buffaloes from Himachal Pradesh with the predominance of the species belonging to genera *Haemonchus*, *Bunostomum*, *Mecistocirrus*, *Strongyloides*, *Trichostrongylus*, and *Oesophagostomum*. Our study also found a high prevalence of these nematode genera, except *Bunostomum* and *Mecistocirrus* which were not detected. In sheep and goats, we have found a very high level of *Haemonchus*, *Strongyloides*, and *Trichostrongylus* infestations. Similar prevalence rates and trends were reported previously by Jithendran [1], except that *Bunostomum* and *Chabertia* were not found in fecal samples of small ruminants in the current study. In terms of the season-wise intensity of gastrointestinal parasites, the month of July witnessed peak levels of EPG counts in ruminants which is comparable to the data reported by Jithendran [1] where the highest parasite counts were observed in the period during June to September. Sharma et al. [61] also found an 86.1% prevalence of gastrointestinal nematodes with the highest incidence in the rainy season in Gaddi sheep from Palam valley of Himachal Pradesh. Among nematodes, *H. contortus* was predominant followed by *Ostertagia* and *Trichuris*, but the prevalence of *Trichostrongylus* and *Strongyloides* was found to be low as compared to our findings.

Environmental conditions such as rainfall, temperature, and relative humidity are some of the influencing factors that increase parasite loads in infected animals due to the enhanced rate of parasite eggs/larvae development [54,62]. Higher humidity in the monsoon months might be responsible for the enhanced survival, transmission, and development of the eggs and other life stages of gastrointestinal nematode parasites. Our findings are in accordance with reports published by Kemal and Terefe [63] and Patel et al. [64]. As less rainfall was received in the months of March, April, May, and June, correspondingly low EPG counts were recorded as compared to July, where the highest rainfall was witnessed during the period of the study. The influence of meteorological parameters especially temperature and rainfall on the prevalence of gastrointestinal nematode parasites quantitatively in both small and large ruminants was observed in this study. Previous reports by Jithendran [1], Nath et al. [65], and Das et al. [43] also supported our findings. Khajuria et al. [66] reported the lowest EPG counts during winters and the highest during monsoons. Kumar et al. [46] also found the highest prevalence of gastrointestinal nematode parasites in the summer season in both cattle and buffaloes. Similarly, Singh et al. [50] observed the highest parasite EPG counts during the monsoon season, although lower values were observed in summer. The higher prevalence of gastrointestinal nematode parasites during the rainy season might be associated with the appropriate molarities of various salts present in soils, which are a crucial factor for inducing ecdysis [34]. Considering the high morbidity and mortality associated with gastrointestinal parasites in livestock, different anthelmintic measures such as chemical drugs, phytochemicals-based herbal medicines, and nano-encapsulated anthelmintics should be evaluated on priority for an effective and sustainable management of gastrointestinal parasites [5,7,67].

5. Conclusions

This study found a high prevalence of gastrointestinal nematodes and gastrointestinal protozoan parasites in the ruminant population from Himachal Pradesh. Rainfall, humidity, temperature, and animal husbandry conditions, grazing, deworming, etc., were found to exert a considerable influence on the prevalence of these parasites. In order to alleviate the biological and economic impact of gastrointestinal parasites on livestock, an integrated, affordable, and combinatorial strategy based on the regular deworming of animals, improving the quality of animal nutrition, safer animal husbandry methods, improved housing, the use of non-protein nitrogenous additives (carbamide-molasses block), nematophagous fungi, and anthelmintic vaccines need to be implemented to minimize the productivity losses and economic impact of these parasites. The findings of this study will prove helpful during the strategic implementation of veterinary health management practices in this

geographical region. Collaborative efforts involving veterinary surgeons, para-veterinary officers, zoologists, and NGOs are required to ensure the control, prevention, and eradication of gastrointestinal parasites from livestock.

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