

Gastrointestinal parasitic infections in Indian Gaddi (goat) breed bucks: clinical, hemato-biochemical, parasitological and chemotherapeutic studies

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Abstract The present study was envisaged with an aim to evaluate gastrointestinal parasitic infections in a herd of conserved Gaddi (goat) breed bucks (6–9 months of age) kept on semi-intensive feeding management. The individuals ($n = 20$) representing the herd were selected and divided into two groups; group I ($n = 10$), clinically ill and group II ($n = 10$), sub clinically infected individuals. The clinical examination revealed anemia, emaciation and rise in body temperature of the individuals of group I as compared to group II. The detailed copro-parasitological examination and copro-culture revealed the presence of eggs of *Moniezia expansa* and larvae of *Haemonchus* species, respectively in the fecal samples of both clinically and sub clinically infected individuals. The hemato-biochemical parameters proved vital indicators of the health of group I individuals and exhibited decline in the values of hemoglobin, packed cell volume and total erythrocyte count as compared to group II. Significant ($P < 0.05$) hypoproteinemia, hypoalbuminemia, hypoglycemia and

increased levels of alanine aminotransferase and aspartate aminotransferase were observed in infected individuals as compared to treated ones. The detailed parasitological, hemato-biochemical observations and clinical findings elucidated and supported the presence of concurrent gastrointestinal parasitism in the herd. The significant improvement was observed in the health status of the herd after 1 month of the therapeutic management, which was carried out using a combination of fenbendazole and praziquantel in both clinically and sub clinically infected individuals.

Keywords Biochemicals · Clinical findings · Gaddi bucks · Hematology · Gastrointestinal parasites

Introduction

In hilly temperate areas of Himachal Pradesh, India (lying in North-Western Himalayan region) Gaddi goat farming is a sustainable system adapted to local ecological and economic constraints. In general, purpose behind small animal farming (especially Gaddi goats) is low investment and input and good output as they help in sustaining poor farmer's family by providing wool, milk and meat (Lateef et al. 2005; Archana et al. 2009).

The helminth infections of gastrointestinal tract of small ruminants not only cause direct adverse effect on the health leading to morbidity and mortality but also indirectly effect economically involving cost of treatment and control of parasites (Nwosu et al. 2007). The concurrent helminth infections cause severe damage to the small ruminants worldwide (Perry et al. 2002). But, the grazing animals are more at risk of contracting gastrointestinal nematode and cestode infections (Mandal and Sharma 2008). The

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researches on small animal gastrointestinal parasitism had highlighted *Haemonchus* species infection as major constraint in small ruminant production worldwide including South-East Asia (Chauhan et al. 2003). On the other hand, moniezirosis also affects adversely the productivity of small ruminants and more specifically lambs become soft victims of the parasitic infestation leading to diarrhea, reduced weight gain, gastrointestinal disorders and even death (Elliott 1986; Yan et al. 2013). The parasitic infections and more specifically helminthic infections are major constraints in profitable small ruminant farming in western Himalayas and other hilly regions of India (Asrani et al. 1999).

In clinical and subclinical helminthic infections, the worms directly invade the host, suck blood, lead to anemia, hypoproteinemia and lower blood glucose levels or indirectly compete for essential amino acids or minerals (Ahmed et al. 2015). Thus, hematology and serum biochemical studies have been proved as important indicators of animal's health exhibiting the level of damage to host tissue and the severity of the infection (Otesile et al. 1991).

The mortality of two Gaddi breed bucks had prompted authors to envisage a study with an objective to assess the parasitological burden and variations in clinical and hemato-biochemical parameters in other individuals of the herd with most appropriate therapeutic management of the diseased individuals.

Materials and methods

Study area and herd description

The study was carried out at Palam Valley in Dhauladhar ranges of Himachal Pradesh (India), located at 32.12°N 76.53°E and at a height of 1470 m above main sea level. The study was carried out in post monsoon season.

The present study herd constituted of bucks only, with the age ranging between 6 and 9 months. The animals were reared under semi-intensive feeding management and were left for 6–7 h of grazing on natural pastures. They were also fed *ad lib* with seasonally available fodders and concentrate mixtures.

Clinical observations

The clinical inspection of the animals was carried out as per Lovatt (2010). The considerations during inspection involved: behavior, posture, gait and mucous membrane etc. Based on observations, the animals ($n = 20$) were divided into two groups: Group I ($n = 10$), animals with clinical infection and Group II ($n = 10$) having sub clinically infected animals. No animals could be considered for

non treated control as all the animals screened exhibited some level of infection.

Parasitological studies

Collection of fecal samples

The fecal samples were collected per-rectally from all the bucks of the herd in polythene bags (with individual identification number) and then transferred to the Department of Veterinary Physiology and Biochemistry, Dr. G. C. Negi College of Veterinary and Animal Sciences, CSK Himachal Pradesh Krishi Vishvavidyalaya, Palampur and kept at 4 °C until further analysis.

Coprological examination and morphometric analysis

The fecal samples were subjected to detailed classical parasitological analysis for presence of parasitic eggs: floatation concentration technique for eggs of nematodes, cestodes and coccidian oocysts and sedimentation technique for evaluation of eggs of trematodes (Soulsby 1982). The samples found positive for parasitic strongyle eggs were subjected to quantitative technique (McMaster egg counting technique) to get the EPG (eggs per gram of feces) (Soulsby 1982). The morphological examination was performed with light microscopy after concentrating the eggs by Sheather's sugar floatation technique (Soulsby 1982). The morphometric analysis targeting the size of the eggs was performed as per Moudgil et al. (2014). The length and width/breadth of the eggs were depicted in micrometers and were expressed considering mean \pm SD.

Copro-culture

Culturing of positive fecal samples was carried out as per the method of Soulsby (1982). The larvae were then recovered using Baermann's apparatus. The genus based identification of third stage larvae (L_3) of strongyles was done as per the key of Taylor et al. (2015).

Hemato-biochemical studies

The blood and sera samples were taken twice, one before and other after 30 days of treatment to assess the alterations before treatment and recovery after treatment. The hematological analysis of the whole blood of all the animals including hemoglobin level (Hb), total erythrocyte count (TEC), total leukocyte count (TLC), packed cell volume (PCV) and differential leukocyte count (DLC) was carried out on fully automated analyzer, Vetscan HM5, Abaxis Veterinary Diagnostics. Biochemical parameters

including aspartate aminotransferase (AST), alanine aminotransferase (ALT), total serum protein (TSP), albumin (ALB), globulin (GLO) and blood glucose (GLU) were analyzed by using commercial kits of Agappe Diagnostics.

Chemotherapeutic management

The concurrent helminthoses was managed by oral administration of drug Fentas-Plus® (combination of 1.5% fenbendazole and 0.5% praziquantel) (INTAS Pharmaceutical Ltd.) @ 1 bolus per 30 kg body weight of the animal in both clinically and sub clinically infected individuals. Quantitative evaluation i.e. eggs per gram (EPG) of feces was determined by using McMaster technique (Soulsby 1982) at day 10, 20 and 30 post treatment. A second dose of the anthelmintic was repeated after 14 days of first treatment due to very low effect of first treatment adjudged by quantitative estimation (EPG) of parasitic intensity. The anthelmintic efficacy of the drug used against strongyle infestation was assessed at days 10, 20 and 30 post-treatment by estimation of fecal egg count reduction (FECR) as per Rinaldi et al. (2014):

$$FECR (\%) = [1 - (\text{Arithmetic mean of FEC at follow-up} / \text{Arithmetic mean of FEC at baseline, day 0})] \times 100.$$

Statistical analysis

The analysis of variance (one-way ANOVA) of hemato-biochemical parameters among the different groups was carried out using Statistical software program (SPSS for Windows, Version 19.0, USA).

Results

While observing the animals from a distance, 13 animals were exhibiting the behavior of dullness with their heads down (depression). Out of these 13 animals, 11 were showing a gait with wide based stance, having their hind quarter soiled with diarrheic feces, whereas the gait and consistency of the fecal pellets of the rest of animals was normal. The conjunctiva mucous membrane of the 13 animals in question was pale, whereas the mucous membrane of the rest of the animals was slight pink to salmon pink. Based on these observations ten animals each exhibiting clinical or subclinical disease entity were selected in two groups to compare hematology and serum biochemical alterations in untreated and treated groups. The animals exhibiting the clinical signs were emaciated and were having an elevated body temperature ranging between 103.9 and 105.2 °F. On the other

hand, the sub-clinically diseased animals were not emaciated and had their body temperature within normal range.

The sedimentation and concentration examinations of the fecal samples revealed the presence of strongyle and triangular anoplocephalid eggs with an EPG value of 2370 ± 226.32 (Table 1) for strongyle eggs, which ranged between 2000 and 2800 eggs in the fecal samples of clinically infected animals. The detailed morphometry of the strongyle and anoplocephalid eggs ($n = 10$ for each animal) revealed a size of $76.18 \pm 3.81 \mu\text{m} \times 41.6 \pm 4.63 \mu\text{m}$ (length \times breadth) and 69.96 ± 4.13 (diameter), respectively, recounting them to be the eggs of *Haemonchus* species and *Moniezia expansa* species. On the other hand, the eggs of strongyles and anoplocephalids were also recovered from fecal samples of sub clinically infected individuals with an EPG of 1170 ± 156.70 (ranging between 200 and 800 strongyle eggs). The decrease in EPG values were encountered at days 10, 20 and 30 post-treatments in case of both the groups.

For the identification, different morphological characteristics of the larvae ($n = 20$), viz. total length, sheath tail extension and number of intestinal cells were taken into consideration after fecal culturing. The average total length and average tail sheath extension of the larvae retrieved was 703.49 ± 26.11 and 72.47 ± 16.07 , respectively with 16 intestinal cells, depicting the larvae to be of *Haemonchus* species (Fig. 1). After culturing, approximately 63.29 ± 14.71 *Haemonchus* species larvae were recovered on an average in different positive fecal samples.

The hematological parameters (Table 2) showed significant decrease ($P < 0.05$) in hemoglobin concentrations ($3.6 \pm 0.70 \text{ g } \%$), packed cell volumes ($10.79 \pm 1.59\%$) and total erythrocyte counts ($9.38 \pm 1.88 \times 10^6/\mu\text{L}$) of clinically infected animals as compared to the sub-clinically infected individuals as well as animals belonging to treated groups. Significant ($P < 0.05$) hypoproteinemia ($3.19 \pm 0.36 \text{ g/dL}$), hypoalbuminemia ($1.16 \pm 0.79 \text{ g/dL}$), hypoglycemia ($36.12 \pm 4.42 \text{ mg/dL}$) and increased levels of alanine aminotransferase ($48.51 \pm 2.92 \text{ U/L}$) and aspartate aminotransferase ($110.15 \pm 2.98 \text{ U/L}$) were observed in infected individuals as compared to treated ones (Table 3).

The concurrent nematode and cestode infection was managed successfully by using the combination of fenbendazole and praziquantel. In case of strongyle infections, both groups I and II exhibited maximum fecal egg count reduction at day 20 post-treatment i.e. 94.09 and 95.72%, respectively (Table 1). But, slight decrease in fecal egg count reduction was observed in both the groups (90.29 and 92.31% in groups I and II, respectively) at day 30 post-treatment (Table 1).

Table 1 Anthelmintic efficacy against strongyle infections

	Group I				Group II			
	Pre-treatment	Post-treatment			Pre-treatment	Post-treatment		
		Day 10	Day 20	Day 30		Day 10	Day 20	Day 30
EPG (arithmetic mean \pm SD)	2370 \pm 226.32	1520 \pm 325.92	140 \pm 51.64	230 \pm 133.75	1170 \pm 156.70	420 \pm 187.38	50 \pm 52.70	90 \pm 73.79
Range	2000–2800	1000–2000	100–200	100–500	1000–1400	200–800	0–100	0–200
FECR (%)	–	35.86	94.09	90.29	–	64.10	95.72	92.31

Values are shown as mean \pm SD

**Fig. 1** Microphotograph of larva of *Haemonchus* species (40X)

Discussion

As heavy parasitic burden was encountered while parasitological evaluation of the fecal samples, the clinico-physical observations of the present study could easily be correlated with the parasitological and hemato-biochemical findings. The clinical features exhibited by the infected

animals were in corroboration with various previous studies. The dullness, depression, emaciation, weakness, diarrheic feces, soiled hind quarters, fever and anemia had earlier been demonstrated in animals heavily infested with *H. contortus* (Soulsby 1982) as well as with *M. expansa* (Miglani et al. 1993; Kar et al. 2007). The dullness and depression of the animals observed in the present study could be attributed to hypoglycemia in the infected individuals, whereas, weakness and emaciation recorded could easily be correlated with hypoproteinemia leading to loss in body muscles during severe gastrointestinal parasitic infestations (Ahmed et al. 2015) and also due to reduced protein synthesis in the muscles in gastrointestinal nematode infections (Liu et al. 2007). Diarrheic feces and soiled hind quarters had also earlier been reported in small ruminants infected with *M. expansa* (Kar et al. 2007). Anemia and fever recorded in the present study had also earlier been reported in caprine hosts infected with haemonchosis (Sharma et al. 2001; Bordoloi et al. 2012) and monieziosis (Kar et al. 2007) and could be attributed to blood loss due to heavy parasitic infestation (Urquhart et al. 1996) and migrating larval stages (Soulsby 1982), respectively.

The parasitological examination involving qualitative and quantitative estimation of parasitic load and micrometry revealed presence of the eggs of *Haemonchus* species and *M. expansa* species. The findings were in line with the

Table 2 Pre and post-treatment hematology of clinically and sub-clinically infected bucks

Groups	Parameters			
	Hb (g %)	PCV (%)	TEC ($\times 10^6/\mu\text{L}$)	TLC ($\times 10^3/\mu\text{L}$)
Group I				
Pre treatment	3.6 \pm 0.70 ^a	10.79 \pm 1.59 ^a	9.38 \pm 1.88 ^a	18.25 \pm 6.99
Post treatment	7.29 \pm 0.83 ^b	17.79 \pm 1.61 ^b	14.28 \pm 1.51 ^b	19.06 \pm 5.02
Group II				
Pre treatment	6.93 \pm 0.56 ^b	15.28 \pm 1.51 ^{ab}	15.07 \pm 1.23 ^b	16.76 \pm 3.59
Post treatment	8.1 \pm 0.98 ^b	19.06 \pm 5.02 ^b	16.64 \pm 1.97 ^b	16.03 \pm 2.97

Values are shown as mean \pm SD

Values with different superscripts differ significantly ($P < 0.05$)

Table 3 Pre and post-treatment serum biochemical parameters of clinically and sub-clinically infected bucks

Groups	Parameters					
	TSP (g/dL)	ALB (g/dL)	GLO (g/dL)	GLU (mg/dL)	ALT (U/L)	AST (U/L)
Group I						
Pre treatment	3.19 ± 0.36 ^a	1.16 ± 0.79 ^a	2.03 ± 1.23	36.12 ± 4.42 ^a	48.51 ± 2.92 ^a	110.15 ± 2.98 ^a
Post treatment	6.67 ± 0.25 ^b	3.79 ± 0.37 ^b	2.88 ± 0.22	53.42 ± 3.55 ^b	36.64 ± 1.85 ^b	98.14 ± 1.56 ^b
Group II						
Pre treatment	5.21 ± 0.70 ^b	2.78 ± 0.26 ^{ab}	2.43 ± 0.45	48.77 ± 3.63 ^{ab}	38.19 ± 1.98 ^{ab}	97.92 ± 2.21 ^b
Post treatment	7.19 ± 0.48 ^b	3.99 ± 0.52 ^b	3.20 ± 0.47	54.22 ± 2.82 ^b	33.06 ± 1.99 ^b	93.02 ± 1.88 ^b

Values are shown as mean ± SD

Values with different superscripts differ significantly ($P < 0.05$)

observations of Mbuh et al. (2008) and Nwosu et al. (2007), who reported heavy infestation of *H. contortus* in the small ruminants especially goats in rainy season and with Ijaz et al. (2008) and Elshahawy et al. (2014), who reported heavy concurrent parasitism of *H. contortus* and *M. expansa* in goats along with other gastrointestinal parasites. The fecal culture studies carried out on the samples of the parasitologically positive animals exhibited the presence of larvae of *Haemonchus* species with similar features as were targeted by Wyk and Mayhew (2013) for the identification of *H. contortus* and other *Haemonchus* species in small ruminants and cattle.

The significantly ($P < 0.05$) lower concentrations of hemoglobin, packed cell volumes and total erythrocyte counts (Table 2) in the parasitic infested animals could be correlated with blood sucking ability of the *Haemonchus* and blood loss due to leakage attributed to severe damage caused by the heavy parasitic burden (Soulsby 1976; Urquhart et al. 1996). Significant ($P < 0.05$) hypoproteinemia and hypoalbuminemia (Table 3) in the infected animals could be attributed to protein losing gastroenteropathy in haemonchosis (Soulsby 1982) and malabsorption of proteins from damaged intestinal mucosa in concurrent gastrointestinal infections (Ahmed et al. 2015). The *Haemonchus* species infested animal loses large amount of serum protein in gut lumen and about 210–340 ml of serum protein cleared with feces per day in infected individuals (Dargie 1975; Bordoloi et al. 2012). Hypoalbuminemia could be attributed to osmotic sensitivity of albumin to fluid losses in parasitic infections especially in haemonchosis and monieziosis leading to its selective loss due to small size (Tanwar and Mishra 2001). Hence, fractional catabolic rate of albumin significantly increased resulting in hypoalbuminemia. Significant ($P < 0.05$) hypoglycemia observed in the present study had earlier been reported in goats infected with *M. expansa* (Kar et al. 2007).

Marked increased levels of enzymes i.e. alanine aminotransferase and aspartate aminotransferase in the

present study was in corroboration with the findings of Sharma et al. (2001) and Bordoloi et al. (2012). Significant increased levels of alanine aminotransferase and aspartate aminotransferase were observed in infected individuals as compared to treated ones due to traumatic damage of abomasal and intestinal lining mucosa caused by adult parasites (Al-Zubaidy et al. 1987; Sharma et al. 2001) or damage of deep abomasal muscular layers by the *Haemonchus* species larval stages (Charleston 1965; Sharma et al. 2001), which could have resulted in rise in the levels of enzymes (Bordoloi et al. 2012).

The management of the concurrent gastrointestinal parasitism was carried out with the application of a combination of benzimidazole (fenbendazole) and acetylated quinoline (praziquantel), which are highly effective against nematodal and cestodal infections, respectively (Taylor et al. 2015). Fenbendazole and praziquantel proved effective against haemonchosis with high fecal egg count reductions at day 20 post-treatment. The efficacy of fenbendazole in case of clinically infected animals was slightly less and was depicting slight resistance at day 20 post treatment, which could be attributed to the fact that immature and resistant individuals could have reached the adult stage. But, slight decrease in fecal egg count reduction reported at day 30 post-treatment in both the groups could be incriminated to reintroduction of the animals to the infected pastures.

Conclusion

It is evident from the present study that for the control of further infection of the animals and for avoiding development of resistance against the effective anthelmintics, the introduction of animals to the infective pastures should be checked and appropriate drug dosage formulation should be carried out. The variation in hematology and serum biochemistry of different groups signified that alteration in these parameters could prove a valuable indicator of health

indices of the infected individuals especially in case of gastrointestinal infestations.

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Authors contribution Aman Dev Moudgil has planned the study and carried out the coprological investigations and treatment of the animals. Ankur Sharma has also helped in treatment of the animals. Madan Singh Verma was involved in haemato-biological studies of the samples. Ravindra Kumar and Pradeep Kumar Dogra helped in review and preparation of manuscript. Pallavi Moudgil has carried out the statistical analysis and prepared and reviewed the manuscript.

Compliance with ethical standards

Conflict of interest The Authors declare that they have no conflict of interest.

Ethical approval The findings recovered in the present study were the result of therapeutic and prophylactic treatment carried out in Gaddi goats and did not involve any experimentation in the precious animals. So, in order to carry out the treatment and preservation of the diseased animals no ethical approval from any agency was sought.

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