Prevalence and Risk Factors of *Haemonchus contortus* Infection in Goats: Linked to FAMACHA, BCS, and Biochemical Markers

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Abstract
Haemochosis is an important gastrointestinal parasitism of particularly small ruminants like sheep and goats. It is becoming a major concern for both commercialized and smallholder goat farmers due to heavy production and economic losses. So, this study was carried out at Bharatpur Metropolitan City of Chitwan to determine the prevalence of *Haemonchus contortus* using a total of 412 individual fecal samples of goat for the coprological examinations. The results showed that prevalence of *H. contortus* was found to be 13.6%. Barbari breed was found highly infected (25%) followed by non-descript breeds (16.7%), Boer (15.8%), Khari (14%), Khapari (13.6%), and Jamunapari (7.5%) respectively. The infection was higher in female than male goats (15.9% vs. 8.8%). The animals with mixed types of feeding (stall and grazing) had a higher infection than the goats having stall feeding (18.7% vs. 11.3%). The goats having a history of regular deworming showed a lower prevalence than deworming with more or no history (4.4% vs. 35.8%). The egg per gram (EPG) showed no statistical difference in the scores for different body conditions scores (BCS) of goats but showed a difference in different FAMACHA scores. Blood-packed cell volume (PCV), albumin, and total protein (TP) were statistically different ($P = 0.00$) in *H. contortus* infected and uninfected goat. Similarly, these blood values showed significant differences in different FAMACHA scores of both *H. contortus* positive and negative goats. Thus, good management practice (GMP), and regular and cyclic deworming along with hematobiochemical status could be crucial for the prevention and adoption of specific treatment protocols at the farm for individual goats.

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Introduction

Goat farming is a prominent livestock industry that provides smallholder farmers, particularly those in rural Nepal, with a substantial source of income and food. Goats can adapt to diversified geographical and climatic conditions, low capital investment, and the ability to utilize marginal lands (Shrestha *et al*., 2020) [35]. Goat farming makes a substantial contribution to poverty reduction and revenue creation, particularly in marginalized populations and female-headed households. Farmers can increase their income and maintain financial stability by selling goats and goat products including meat, milk, and skins (Neupane *et al*., 2018) [29]. The reports from MoALD (2023) [27] show that around 13.99 million goats were living in Nepal. Despite the high numbers, Nepal mainly depends on India for a year-round meat supply and seasonal imports from Tibet. Goats have a comparative advantage over other animals due to their extensive feeding habits, small size, high adaptability to unfavorable environments, low maintenance cost, diverse utility, gestation period relatively short, and multiple offspring at a time.

(Degen, 2007; Dhakal *et al*., 2021; Khanal *et al*., 2024; Neupane *et al*., 2018) [10, 11, 1, 29]. They can transform household, industrial, crop residues, and other farm byproducts into valuable edible protein, contributing to national nutritional security because of most people's higher preference for goat meat (Poudel, 2018) [32]. In terms of national meat output, goat's meat is the third most after poultry and buff in terms of popular meat production (MoALD, 2023) [27].

Goat farming is the most profitable business among livestock industries, but gastrointestinal (GI) parasites are a common and major constraint. Nematodes, commonly known as roundworms, are the most common internal parasites that economically impact small ruminants (sheep and goats) globally (Roeber *et al*., 2013) [34]. Helminth in livestock industries significantly lower productivity in sheep and goats in the tropical and subtropical regions. Furthermore, the lack of good veterinary facilities and the suitable environment of the tropics facilitate the invasion and propagation of nematodes. Thus, nematode infection is common in Nepal (Adhikari *et al*., 2017) [1]. The infection is suspected by decreased weight gain and disease resistance, low production, and mortality in severe parasitism. Goats are a major source of nutrition and livelihood for small farmers in Nepal. However, the productivity of goats is being compromised due to the high prevalence of *H. contortus*. This voracious bloodsucker roundworm resides in the stomach leading to severe clinical lesions and signs. A single worm ingests up to 50 µL of blood per day, high infection levels can cause severe blood loss (more than 100 ml daily), followed by anemia and hypoproteinemia. It is important to address this issue because farmers in Nepal lack sufficient information about parasitic diseases and their effective control strategies which results in poor management and continuous economic losses. Previous research on *H. contortus* in Bharatpur Metropolitan City does not include a correlation between *H. contortus* infection with Faffa Malan CHArt (FAMACHA) and serum biochemistry. Farmers in Nepal lack easy handy diagnostic tools for *H. contortus* and its effective control strategies. This research aims to fill this gap by developing and validating a rapid and accurate diagnostic technique for *Haemonchus* species, tracing the prevalence and distribution of infections, and suggesting effective management practices to control parasitic infestations in goats. By doing so, this study aims to uplift livestock management systems, animal health, and productivity, thereby supporting Nepalese farmers' livelihoods.

This study focuses on the prevalence and associated risk factors of *Haemonchus contortus* infection in goats within Bharatpur Metropolitan City. The primary objective is to evaluate the prevalence of *H. contortus* and identify factors contributing to its occurrence. Specifically, the study aims to investigate the relationship between the prevalence of *H. contortus* and key risk factors, including age, sex, breed of goats, feeding management practices, and deworming history. Furthermore, the research seeks to explore the association between the infection levels of *H. contortus* and various health indicators, such as FAMACHA scores, body condition scores (BCS), and haemato-biochemical parameters like packed cell volume (PCV), serum albumin, and total protein levels. These insights are crucial for understanding the dynamics of *H. contortus* infections and their impact on goat health, contributing to better management and control strategies.

Research Methodology

Duration of research study and site

The study was conducted for five months from April to August 2023. This research study was conducted in Bharatpur Metropolitan City (27°40'59.99" N 84°25'59.99" E). The mean temperature of Bharatpur is 23.2° C with rainfall of 94.8 inches per year.

Sample Size Calculation

According to MoALD, (2022) [27], the total population of Goats in Chitwan was 256,993. The number of samples required for the known population was estimated by using the formula given by (Almeda *et al*., 2010) [2].

$$n = \frac{Nz^2}{z^2+4Ne^2}$$

n = sample size, N = population size, e = level of error

As per the formula,

We have N = 256,993, and Z = 1.96 as a 95% level of confidence, e = 0.05 (5% error)

So, we calculated sample size (n) = $\frac{256993*(1.96)^2}{(1.96)^2+4*256993*(0.05)^2}$ = 383

The approximate sample size according to the formula was 383 and the actual sample size taken was 412 goats.

Sample Collection, Preservation, and Transportation

Observation and cross-sectional studies were carried out for the sample collection in different households. A questionnaire survey was carried out for age, sex, and deworming history of goats. Purposive random sampling was done for 412 fecal samples and blood samples. Fecal samples were collected immediately after the defecation by goats. It was done in the early morning. For the samples, about 3gm of feces were collected with great precaution using a spatula and transferred to sterile zip-locked polyethylene bags. The bags were labeled, preserved in 10% formalin, and transported to the parasitology lab FAVF, AFU, Rampur, Chitwan in a cool box and examined. The FAMACHA grade Card was used to grade the animals, and Evaluation was done using the body condition score (BCS).

About 5 ml of blood was drawn from the jugular vein of the goats using sterile needles. The blood collected was kept in an EDTA vial for PCV and a serum vial for blood biochemistry. The blood samples were then transferred to the lab using the cool box at 4^0 C.

Laboratory Examination of the samples

For fecal samples

The samples were processed and examined microscopically for the identification of the parasites. These techniques can be broadly classified into qualitative and quantitative methods. The qualitative and quantitative examination of the *H. contortus* eggs was done using different methods (Soulsby, 1982) [36]: Direct smear, sedimentation, flotation, and McMaster. Here are some commonly used fecal examination techniques, their principles, and benefits:

Direct smear methods

In this method, about 3gm of samples were crushed using mortar and pestle then a drop of iodine was placed in a clean and grease-free slide. The small number of samples was placed in an iodine drop and a smear was made. Then the smear was placed in the microscope and used magnification of 100X and 400X.

Sedimentation methods

Using this procedure, water was added to a 100 ml beaker containing around 3 g of samples. The solution was stirred with a clean glass stirrer. Then the mixture was sieved into a clean beaker and the remnants in the sieve were discarded. The supernatant was removed after 30 minutes and fresh, clean water was refilled. This process was repeated until the supernatant was clear. Then the sediments left at the bottom of the beaker were examined microscopically.

Differential flotation methods

In this method, about 3 gm of samples were kept in 100 ml of clean and dry beaker and water was added to it. The solution was stirred with a clean glass stirrer. Then the mixture was sieved into a clean beaker and the remnants in the sieve were discarded. After 30 minutes, the supernatant was discarded, and clean fresh water was refilled. This process was repeated until the supernatant was clear. Then the supernatant was mixed with 42 ml saturated common salt solution (380 grams of NaCl/ L of water having a specific gravity of 1.2). The mixture was then placed undisturbed for 30 minutes. After that, the clean and dry cover was allowed to touch the surface of the mix then transferred to clean and grease-free slides and examined under the microscope.

Quantitative methods

For this method, MacMaster's counting technique was used. It is used to determine the fecal samples' egg per gram (EPG). In this method, about 3 gm of sample were ground using mortar and pestle then the grounded samples were poured with 42 ml clean and fresh water. The mixture was placed in 3 centrifuge tubes (14 ml in each tube) and centrifuged for 2 minutes at 2000 rpm. The supernatant was removed and the NaCl solution was added before straining. About 0.15 ml of the mixture was then placed on a Macmaster slide using a pipette and covered with a clean cover slip. Then the counting of eggs was done using a microscope. The EPG was calculated as:

EPG: Number of eggs counted in 2 chambers multiplied by 50

Blood samples analysis

The blood samples in the EDTA vial were used for hematological parameters and the blood samples in coagulants were used for blood biochemistry and were carried to the Pathology lab, AFU, Rampur.

PCV determination and blood biochemistry

For the determination of the PCV, the blood samples and the PCV determination were done following the micro-hematocrit technique described by Coles (1967) [9]. Serums were analyzed for the serum albumin and total protein in the Biochemistry lab, AFU, Rampur using a colorimeter. Serum Albumin analysis was done by the Bromocresol Green method (Doumas *et al*., 1971) [12] and Total Protein analysis was done by the Biuret method (Gornall *et al*., 1949) [49].

Scoring procedure

Body Condition Score (BCS)

Body condition scoring is a system for categorizing animals based on their degree of body reserves. Each animal is assigned number values based on its seeming external fat cover, muscular look, and apparent skeletal features, which are derived by subjective eye evaluation and/or physical examination (Ghosh *et al*., 2019) [13]. The scoring of the goats was done according to the table below:

Table 1: Body condition Score description for the assessment of goats

BCS of goats		Vertebral spines feeling	Ribs observation	Loin region
1	Very Thin	Easily visible as sharp	Easily feel with slight pressure	Fat layer absent
2	Thin	Smooth, non-sharped	Rounded, smooth, small pressure needed to feel	Smooth, Fat layer even
3	Good condition	Rounded and smooth	Smooth, even feel	Smooth, fat cover evenly
4	Fat	High pressure is needed, and sharp points absent	Individual ribs cannot be felt, but can still feel indent between ribs	Heavy fat
5	Obese	No vertebral feel and Smooth	Individual ribs cannot be felt. No separation of ribs felt	A thick covering of Fat.

FAMACHA Scoring in Goats

FAMACHA Card was used to determine the scoring of animals. The FAMACHA scoring procedure entails exposing the lower ocular mucous membranes and correlating them with the relevant color on the FAMACHA card. FAMACHA scorecards were used to assign individual animal scores. The assessor assessed the animals' conjunctival colors on a scale of 1 to 5 using the FAMACHA scorecard. The colors of each animal's eye mucous membranes were graded using the FAMACHA color scale as follows. 1 = red, non-anemic; 2 = red-pink, non-anemic; 3 = pink, somewhat anemic; 4 = pink-white, anemic; 5 = white, severe anemia (Kaplan *et al*., 2004) [19].

Data recording and analysis

Data recording was done, and entry was done at Ms-Excel 2010, and the data analysis was by using SPSS 20. For the analysis of the association between the prevalence of *H. contortus* in goats with their breed, sex, age, feeding, and deworming status; a chi-square test was performed (SPSS 20). ANOVA (Analysis of Variance) followed by DMRT (Duncan's Multiple Range Test) was done for the association of EPG with BCS and FAMACHA alterations in different parameters. ANOVA followed by an independent t-test was done for the association of biochemical parameters with *H. contortus* positive and negative goats.

Results

General Prevalence of *H. contortus*

Out of 412 fecal samples from goats 56 samples (13.6%) were found positive for *H. contortus.*

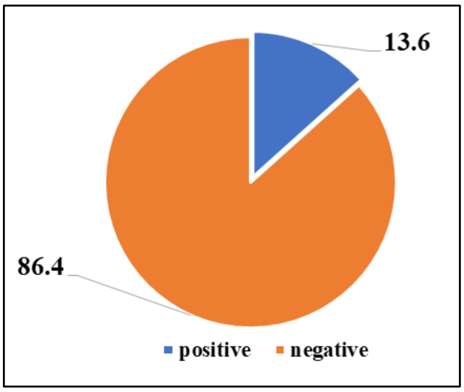

Fig 1: The overall prevalence percentage of H. contortus in goats of Bharatpur Metropolitan City

Breed-wise prevalence

The results showed no variation in the prevalence of *H. contortus* in goats among different breeds (*p* = 0.95). Out of 412 samples, the highest prevalence was found in Barbari and the lowest prevalence in Jamunapari.

Table 2: The prevalence of *H. contortus* in Goats of different breeds

Breed	Total samples	Positive number (%)	χ^2	df	*P* value
Barbari	12	3 (25)	6.62	5	0.95
Boer	190	30 (15.8)			
Jamunapari	107	8 (7.5)			
Khapari	22	3 (13.6)			
Khari	57	8 (14)			
Non-descript	24	4 (16.7)			
Total	**412**	**56 (13.6)**			

Sex-wise prevalence

There was a statistically non-significant difference in the prevalence of parasites according to the male and female goats (*P* = 0.30). However, females were infected more than male goats.

Table 3: Prevalence of *H. contortus* among male and female Goats in the Bharatpur Metropolitan City

Sex	Total samples	Positive number (%)	χ^2	df	*P* value
Female	276	44 (15.9)	4.41	1	0.30
Male	136	12 (8.8)			
Total	**412**	**56 (13.6)**			

Age-wise prevalence

The results showed no statistical difference in the prevalence of parasites in different age groups p = 0.68). But goats with age group less than 1 year were infected higher than other groups of goats.

Table 4: Prevalence of *H. contortus* in goats according to different age groups

Age (Year)	Total samples	Positive number (%)	χ^2	df	*P* value
Less than 1	202	25 (12.4)	115	3	0.68
1 to less than 2	107	10 (9.3)			
2 to less than 3	75	17 (22.7)			
3 and more	28	4 (14.3)			
Total	**412**	**56 (13.6)**			

Prevalence of parasites according to feeding methods

The results showed the prevalence of parasites in mixed types of feeding as 18.7% and stall feeding as 11.3%. However, there was no difference in the prevalence of parasites in different feeding methods (*p* = 0.27).

Table 5: Prevalence of *H. contortus* in goats according to types of feeding practices

Feeding methods	Total samples	Positive number (%)	χ^2	df	*P* value
Mixed (Stall and grazing)	128	24 (18.7)	4.77	1	0.27
Stall	284	32 (11.3)			
Total	**412**	**56 (13.60)**			

Prevalence of parasites according to deworming history

The results showed a statistically significant prevalence of *Haemonchus* in goats with different deworming histories (*p* = 0.03). The animals that were dewormed within 3 months were found to be infested by *H. contortus* by only 4.4%. However, the animals that were not dewormed within 3 months were found to be heavily infested with *H. contortus*.

Table 6: Prevalence of *H. contortus* according to the deworming history of goats.

Deworming History	Total samples	Positive number (%)	χ^2	df	*P* value
0-3 months	292	13 (4.4)	47.401	1	0.03
3-6 months	120	43 (35.8)			
Total	412	56 (13.60)			

Egg per gram (EPG) in Body Condition Score (BCS) and FAMACHA scores

The result showed that EPG had no statistical difference in the different body condition scores of the animals. However, with increasing BCS, EPG decreased.

There was a significant difference in EPG with different FAMACHA scores. There was a significant increase in EPG with an increase in FAMACHA scores.

Table 7: Association of Egg per gram (EPG) with different Body Condition Scores (BCS), and FAMACHA Scores of Goats in the Bharatpur Metropolitan City

		Egg per gram (EPG)	*P* value
Body Condition Score	1	-	0.084
	2	1030.00±145.72	
	3	575.00±70.84	
	4	312.50±12.50	
	5	-	
FAMACHA Score	1	-	0.00
	2	312.50±12.50[a]	
	3	325.00±131.50[a]	
	4	460.00±62.58[a]	
	5	1342.86±175.74[b]	

*Note: Values are mean±SEM, and a-b (mean±SEM) with different superscripts within the column differ significantly at the 5% level by the Duncan's (DMRT) method.

Hematobiochemical alterations in goats

The result showed a statistical difference in blood PCV, Albumin, and Total protein in *H. contortus* positive and negative goat**.** The blood parameters decrease in *H. contortus* infected goats.

Table 8: Association of Biochemical values (PCV, Albumin, and TP)

Status	Packed cell volume	Albumin(g/dl)	Total protein (g/dl)
H. contortus (Negative)	27.94±.37[a]	2.04±.03[a]	5.60±.05[a]
H. contortus (Positive)	20.07±.85[b]	1.60±.04[b]	4.39±.13[b]
***P* value**	0.00	0.00	0.00

*Note: Values of PCV, Albumen, and TP are represented with mean±SEM where a-b (mean±SEM) with different superscripts within the rows differ significantly at the 5% level by the independent t-test

Blood PCV, serum albumin, and Total protein concerning *H. contortus* positive and negative goat in different FAMACHA scores

The result showed a significant difference in blood biochemical values (albumin, total protein, and PCV) in *H. contortus* positive and negative goats with different FAMACHA scores. The blood biochemical values decreased with an increase in FAMACHA scores.

Table 9: Blood biochemical values of *H. contortus* infected and uninfected goats in different FAMACHA scores

		H. contortus	Albumin	Total protein	Packed cell volume
FAMACHA score	1	-	-	-	-
		-	-	-	-
	2	Positive			
		Negative	1.76±.00	4.97±.00	22.00±.00
	3	Positive	2.29±.03	6.36±.06	32.65±.49
		Negative	2.32±.00	6.63±.00	34.00±.00
		P value	0.009	0.003	0.036
	4	Positive	1.79±.04	4.80±.16	23.20±1.04
		Negative	1.85±.03	5.00±.05	24.26±.33
		P value	0.052	0.325	0.433
	5	Positive	1.26±.00	3.08±.00	13.00±.00
		Negative	1.34±.03	3.69±.09	15.57±.58
		P value	0.007	0.006	0.001

*Note: The values were expressed in Mean ± SEM, and the *P* values were obtained by independent t-test

Discussion

The present study showed that there was a 13.6% prevalence of *H. contortus* in the Bharatpur metropolitan city. Our study results were similar to the study of Adhikari *et al*. (2017) [1] which showed the prevalence of *H. contortus* at 13.89% in the western region of the Chitwan district. A similar type of result was seen by Karki *et al* (2012) [20] in the Kalanki of Kathmandu which showed the prevalence of *H. contortus* to be 14.5%. The prevalence of the parasites found in the study was different from the study by (Tripathi & Subedi, 2015) [39] in Kapilvastu district, which was found to be 3.43%, 20.89% in Chitwan district by KC & Karki, (2012) [21].

The difference in results may be due to natural resistance different management practices in animal husbandry differences in nutritional status, different geological conditions, drug treatment, and age factors (Barger, 1999; Barnes *et al*., 2001; Chaudary *et al*., 2007; Gupta *et al*., 1987; Lindqvist *et al*., 2001; Mandonnet *et al*., 2003; Mulugeta *et al*., 2011; Pal & Qayyum, 1993; Preston and Allonby, 1987; Soulsby, 1982) [6, 7, 8, 16, 24, 26, 28, 33, 36].

The study showed there was no significant difference in the prevalence of *H. contortus* in different breeds. The case was different in the study of the prevalence of *H. contortus* in different breeds of goats by Lashari *et al* and (Tasawar *et al*., 2010) [37] where prevalence was significantly different in the different breeds of goats. The variation in the prevalence of *H. contortus* in animals may be due to the differences in species of animals, genetic differences, geological differences, nutritional and management differences, and due to the seasonality and climatic condition differences during the study by different researchers.

The prevalence of the *H. contortus* in the study according to age was not significantly different (P = 0.68). Our study results were supported by the study done by Adhikari *et al*. (2017) [1] and Tasawar *et al*. (2010) [37] which also shows no statistical difference between the prevalence of *H. contortus* in different age groups. The higher prevalence of parasites at a young age below 3 years is due to the lower immunity due to low exposure to parasites at young ages. The young animals were kept mixed with adults; not segregating from adults may cause a higher chance of exposure to parasites and lower immunity in young animals makes it critical for the infection of the parasites.

The study showed a significant effect of the deworming history on the prevalence of parasites (P = 0.03). The animals with a deworming history within 3 months had the least infection whereas the goats with no deworming history within 3 months were heavily infected. The result was similar to the study by Adhikari *et al*. (2017) [1]. The variation in the prevalence of the parasites may be due to the effective deworming by anthelminthic drugs.

The study showed no statistical difference in EPG in different Body condition scores of goats. There was a lower parasitic load in higher BCS which is similar to the reports by Idika *et al* (2012) [17] and Ogbudu *et al* (2022) [30] which stated that helminth attack was more in goats with poorer BCS than those with higher body condition scores. There was a significant difference in EPG in different FAMACHA scores. There was an increase in EPG with an increase in FAMACHA score. The study was similar to the findings of Bandhaiya *et al* (2020) [5] and Bala *et al* (2015) [4] which stated that the increased parasitic load in animals decreases the whole blood increasing the FAMACHA score.

The result showed there was a significant decrease in packed cell volume, albumin, and total protein in *H. contortus* infected goats which was similar to the study of Awad *et al* (2021) [3] which demonstrated a decrease in PCV to 18±.19, albumin to 1.7 ± 0.45g/dl, and total protein to 5.02± 0.59 g/dl. Decreased in biochemical values in this research may be due to the bleeding of the abomasa due to the injuries caused by the *H. contortus*. It is estimated that an adult *H. contortus* can suck 0.05 ml of blood/day, in addition to causing leakage of blood from the site of attachment (Urquhart *et al*, 1996) [40].

This study showed that blood parameters like PCV, Serum albumin, and total protein were found to differ in different FAMACHA scores (P<0.001). Similar findings were obtained by Malum (2014) [25] in Nigeria. Low-packed cell volume is a sign of anemia (Kaneko *et al*., 1997) [18]. However, in goats, PCV is influenced by sex and altitude (Tibbo *et al*., 2005) [38], management, age, breed, health status, ambient temperature, and physiological statuses such as excitement, muscular exercise, pregnancy, estrus, parturition, water balance, and transportation (Kaneko *et al*., 1997) [18].

Conclusion

The overall prevalence of *H. contortus* in goats was 13.60. Timely deworming the goats significantly reduces the infection rate while, age, sex, breed, and feeding management were not found to have significant differences in *H. contortus* infection. The higher the EPG, the higher the FAMACHA score, and the lower the PCV, Albumin, and total protein. Biochemical markers and FAMACHA Scores serve as reliable tools for diagnosing infection severity and monitoring goat health.

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