Original Article

Prevalence of Intestinal Parasites in Dogs (*Canis familiaris* Linnaeus, 1758) and Dzos (*Bos grunniens* Linnaeus, 1766) in Upper Humla, Nepal

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ABSTRACT

Background: Dogs and dzos are important domestic animals in human communities in high-altitude landscapes and are the potential carriers of different parasites. There is a risk of zoonotic parasite transmission between dzos and free-ranging dogs. Therefore, evaluating and managing the parasites could play a role in safeguarding health and overall well-being.

Objectives: The study aimed to investigate the prevalence and diversity of intestinal parasites in free-ranging dogs, owned dogs and dzos in Upper Humla, Nepal.

Methods: Fecal samples (n=151), including 109 from free-ranging dogs, 12 from owned dogs, and 30 from dzos, were collected. Microscopic examination of the fecal samples was conducted using direct wet mount and acid-fast staining methods.

Results: The overall prevalence of gastrointestinal (GI) parasites was 75.49%, with 75.23% in free-ranging dogs, 66.67% in owned dogs and 80% in dzos. Nineteen parasite species (18 confirmed) were recorded with nine species in dzos, seven in owned dogs, and 17 in free-ranging dogs. Triplet infections were more common in free-ranging dogs, while duplicate infections were more prevalent in owned dogs and pentuplet infections were more frequent in dzos. The dogs and dzos of Upper Humla were commonly infected with *Entamoeba* spp., ascarids, *Cryptosporidium* spp., *Eimeria* spp. and *Taenia* spp.

Conclusion: Intestinal parasites can substantially threaten human populations through zoonotic transmission. Controlling and managing the parasitic infection in dogs and dzos can help reduce the impact on human health.

Keywords: Agro-pastoralism, Cross-transmission, Gastrointestinal, *Cryptosporidium*, Zoonosis

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Introduction

Dogs (*Canis familiaris* Linnaeus, 1758) were among the first pets domesticated since prehistoric times for purposes (Bradshaw et al., 2017; Freedman et al., 2014). Sequencing of archaic DNA indicates that dog domestication began in Siberia about 23000 years ago (Perri et al., 2021). Free-ranging dogs include street dogs, stray dogs, and feral dogs that spend their lives in the wild outside human habitations, hunt with their groups, and breed without disturbances from humans (Boitani & Ciucci, 1995). These free-ranging dogs are opportunistic predators (Sarkar et al., 2023). They may also pose threats to wildlife (Thompson, 2013) and can facilitate the transmission of various zoonotic diseases (Butler et al., 2004), including several gastrointestinal (GI) parasitic species of zoonotic importance (Thompson, 2013; Khalifa et al., 2023; Sukupayo et al., 2023; Adhikari et al., 2023). The 2024 Worldostats estimates that out of approximately 900 million dogs globally, about 500 million are pet dogs with numerous breeds (Worldostats, 2024). The dzo (*Bos grunniens* Linnaeus, 1766) is a hybrid between the yak (*Bos mutus* GBIF Secretariat (2023a) and domestic cattle (GBIF Secretariat, 2023b). The male form is called a Jhoppa, and the female is referred to as a Jhuma.

Different types of endoparasites are responsible for causing health issues (Naqid, 2024; Ismael et al., 2024; Dalimi & Jaffarian, 2024) and death among pets (Morandi et al., 2020). Several endoparasites, such as *Toxocara canis*, *Ancylostoma* spp., *Giardia duodenalis*, *Cryptosporidium parvum* and *Toxoplasma gondii*, are zoonotically important (Oliveira-Sequeira et al., 2002; Ahmad et al., 2020; Firooz Jahantigh et al., 2020; David Ola-Fadunsin et al., 2023; Chamanara et al., 2024). Shelter conditions and scavenging feeding habits have been identified as significant contributing factors to the occurrence of endoparasite infections (Grandi et al., 2021; Adhikari et al., 2023).

Dzos are commonly found in a few Hindu-Kush Himalayan (HKH) regions, like Nepal Bhutan, and Tibet. In Humla, dzos are among the most preferred livestock types due to their adaptive nature and multi-purpose uses, including transportation in Upper Humla. Dzos frequently interact with free-ranging dogs. In these contexts, GI parasites can be cross-transmitted among owned dogs, free-ranging dogs and dzos in high-altitude landscapes. Due to the lack of veterinary health services for timely and routine vaccination of dogs or cattle, as well as the lack of awareness among the local population, there is a high risk of transmitting various GI parasites to nearby wild populations and humans. No studies have been conducted regarding GI parasites of dogs and dzos and their possible impact on zoonosis, although the HKH region is significant in the context of various microspecies associated with the hosts and the environment (Ghimire et al., 2020). Therefore, the current study aimed to study the prevalence of GI parasites in dogs and dzos in Namkha Rural Municipality and Simkot Rural Municipality in the Humla district of Nepal.

Materials and Methods

Study area

This study was carried out in the Namkha Rural Municipality and Simkot Rural Municipality in the Humla district of Nepal (Figure 1). Humla district is the second largest in Nepal, covering an area of 5655 km^2 with a population of 55394 individuals (NSO, 2022). This region is characterized by its remoteness, rugged terrain, high mountains, and deep valleys and it is not connected to the rest of Nepal by road, making it one of the most isolated and challenging areas to reach in the country (Oli & Zomer, 2011; Lama et al., 2018). The district includes several rivers, including the Karnali River, which flows through Humla from its source in West Tibet. Upper Humla has a harsh climate, with snow for up to four months of the year, and experiences a tropical climate that includes wet and dry seasons, with most of the rainfall occurring during the monsoon season. The settlements are located between 2500-4000 meter. The common leopard, snow leopard, golden jackal, Himalayan black bear, Himalayan tahr, yellow-throated marten, leopard cat, Himalayan langur, rhesus monkey, chucker partridge, Himalayan griffon, danphe, bearded vulture, rock lizard and Himalayan pit viper are common wild faunae, whereas cattle, goats, sheep, dzos, yaks, horses, mules, donkeys, and chickens are common domestic fauna in the study areas (Lama et al., 2021; Thapa, 2023).

Sample collection, preservation and transportation

Before sample collection, a three-day primary field survey was conducted to investigate the study area and familiarize the investigators with the local population, husbandry practices, geographical location and population status of dogs and dzos. A detailed survey was conducted from October, 21 to October 30, 2022, with the assistance of trained local field associates. An opportunistic sampling technique was used to collect stool samples as it was challenging for the researchers to create a sampling frame in Upper Humla. A total of 151 fresh fe-

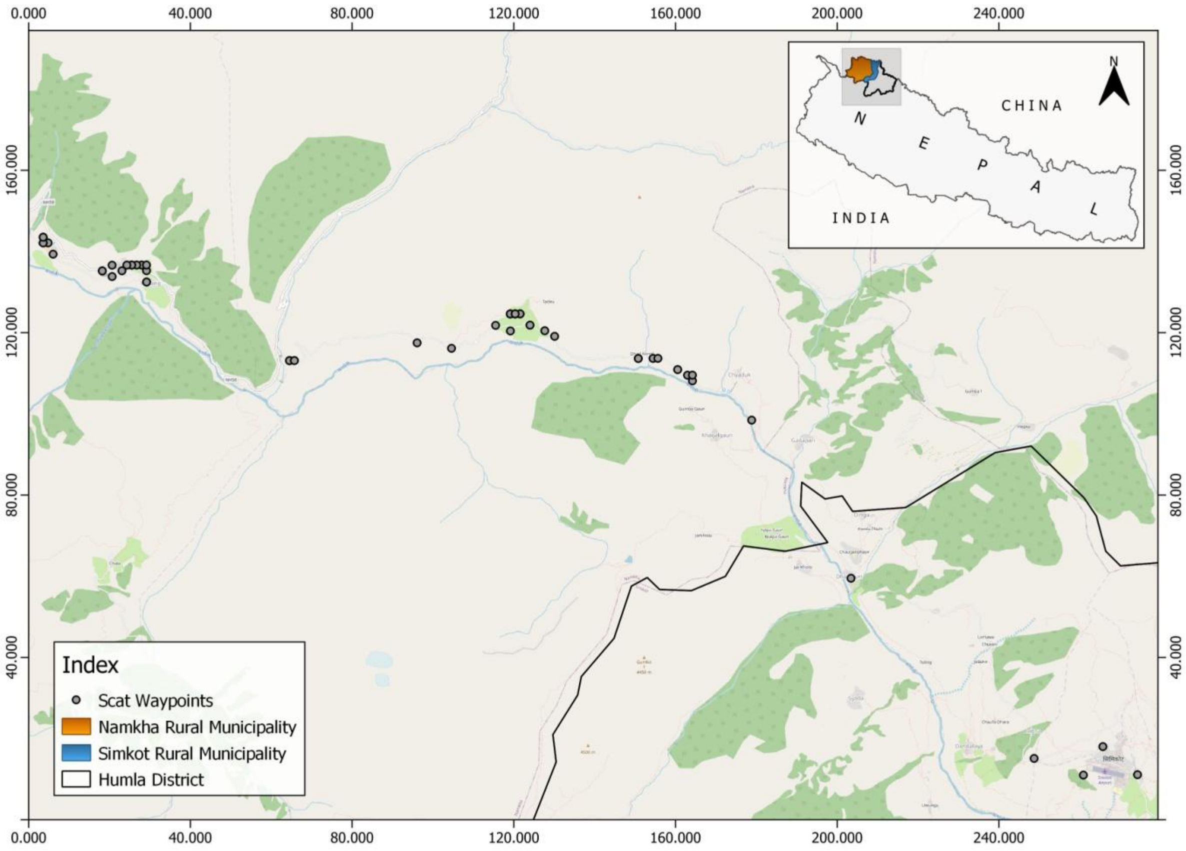


Figure 1. Map of the study areas

cal samples, collected opportunistically from the ground immediately after defecation, were obtained (Adhikari et al., 2022; Adhikari et al., 2023) from a total of 121 dogs, which included domestic dogs (n=12), free-ranging dogs (n=109) and dzos (n=30). Initially, the samples were macroscopically examined for blood, mucus, segments of worms and types of stool structures. The samples were then preserved in a 2.5% potassium dichromate solution in 20 mL sterile vials. A unique identification code was written on the vial of each fecal sample and finally, these samples were transported to the Research Laboratory in Kathmandu, Nepal.

Laboratory processing and examination

The fecal samples were analyzed using previously explained methods (Adhikari et al., 2022; Adhikari et al., 2023; Adhikari et al., 2024; Thapa Magar & Ghimire, 2023). These methods included direct wet mount techniques with or without using iodine staining and modified acid-fast staining techniques. A solution of iodine, which consisted of 10 g of potassium iodide dissolved in 100 mL of distilled water along with 5 g of iodine crystals, was utilized for the wet mount method (Zajac et al., 2021).

Approximately 2 g of a few stool samples were stirred and mixed carefully. A single drop of each sample was placed on a clean and dry glass slide, with or without Gram's iodine stain. The sample was covered with a coverslip and observed under a microscope at magnifications of 10× and 40× (Adhikari et al., 2023; Adhikari et al., 2024; Thapa Magar & Ghimire, 2024). The same sample was observed without using an iodine solution at the same magnifications.

The acid-fast staining test was performed primarily to identify *Cryptosporidium* spp.; therefore, a stool smear was prepared, air-dried, and heat-fixed. The smear was covered with carbol fuchsin (a mixture of phenol and basic fuchsin). The slide was placed over steam. After 5 to 10 minutes, the slide was cooled and gently rinsed with distilled water. Two to three drops of acid alcohol were applied to the slide, which was then rinsed with water. A drop of methylene blue was added to the slide for one minute and rinsed with water. Then, the slide was air-dried and observed under a compound microscope at a magnification of 100X using immersion oil (Ghimire & Bhattarai, 2019; Adhikari & Ghimire, 2021; Adhikari et al., 2020; Adhikari et al., 2022; Adhikari et al., 2023).

Parasite identification

The microscopic images of the detected parasitic stages, such as eggs, cysts, trophozoites and oocysts, were captured using a built-in camera on a mobile device (iPhone 11 Pro Max). Their identification and taxonomic position were confirmed using literature and online sources (Soulsby, 2012; Zajac et al., 2021; Adhikari et al., 2023; Sukupayo et al., 2023). *Entamoeba* spp. were identified by the smaller cysts with one to four nuclei, while *Entamoeba coli* were confirmed by the larger cysts with eight nuclei.

Data analysis

Data were tabulated, edited, and expressed in Microsoft Excel software, version 2010. The prevalence rates of parasites were calculated in each host, and their statistical significance was tested using P obtained from the chi-square tests and nonparametric Spearman's correlation. A P<0.05 (95% confidence interval [CI]) was considered statistically significant when comparing the prevalence rates among different subjects and variables. The intensity of each infection was calculated based on the observation of cysts, oocysts, trophozoites, or eggs of the parasite after direct wet mount. Therefore, the following symbolic indications were used for evaluating the intensity of particular parasitic species:

+1-3 per field; ++: 4-10 per field; +++: 11 or more (×400 field for protozoa); +1-3 per field; ++: 4-10 per field; +++: 11 or more (×100 field for helminths).

Results

In this study, out of 151 fecal samples, 114(75.49%) were positive for GI parasites with rates of 66.67% (8/12) in owned dogs, 75.23% (82/109) in free-ranging dogs, and 80% (24/30) in dzos. Nineteen different parasitic species were recorded, with nine species in dzos, seven in owned dogs and seventeen in free-ranging dogs (Figure 2 and Table 1). The prevalence of *Cryptosporidium* spp. was the highest among all three hosts. Similarly, GI parasites shared by three types of hosts were analyzed. These hosts shared *Cryptosporidium* spp., ascarid, *Entamoeba* spp., *Eimeria* spp., and *Taenia* spp. Interestingly, *Blastocystis* sp. was shared by only owned dogs and dzos, but *Cyclospora* sp. and *Neospora caninum* and unknown coccidia were shared by stray dogs and dzos (Table 1).

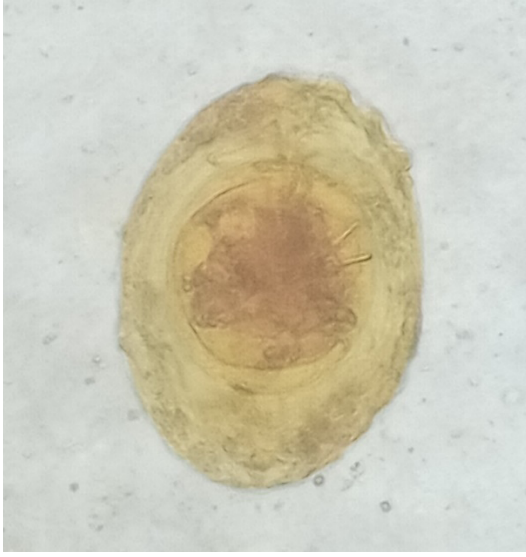
In owned dogs, ascarids and *Entamoeba* spp. showed the highest prevalence at low intensity. Interestingly, *Cryptosporidium* spp. showed the highest prevalence at both moderate and high intensities. In stray dogs, ascarids showed the highest prevalence at low intensity, whereas *Cryptosporidium* spp. showed the highest prevalence at either moderate or high intensity. In dzos, *Entamoeba* spp. exhibited the highest prevalence at low intensity, while *Cryptosporidium* spp. had the highest prevalence at moderate intensity, and *Taenia* spp. had the highest prevalence at high intensity (Table 2).

Data were analyzed using nonparametric Spearman's correlation between any two selected intensity sets for all total GI parasitic species to calculate r with a 95% CI and Gaussian approximation P (two-tailed). In domestic dogs, comparing ++ and +++ intensities resulted in an r of 1.000 (P<0.0001, 95% CI, 1.000%, 1.000%). In stray dogs, r was 0.4998 (P<0.05, 95% CI, 0.04437%, 0.7832%) when comparing + and ++ intensities. Similar results were obtained after comparing ++ and +++ intensities, generating an r of 0.9899 (95% CI, 0.9732%, 0.9962%, P<0.0001). In dzos, the comparison between ++ and +++ intensities generated an r of 1.000 (95% CI, 1.000%, 1.000%, P<0.0001). Interestingly, when comparing total intensities between ++ and +++, r was 0.7265 with a 95% CI, 0.3941%, 0.8908% and P<0.0005 (Table 2).

The current study also analyzed the concomitance of GI parasites in these hosts. Mixed infection of up to five different GI parasites was present in owned dogs, whereas seven GI parasites were present in free-ranging dogs or dzos, indicating a high concurrence of GI parasites (Table 3).

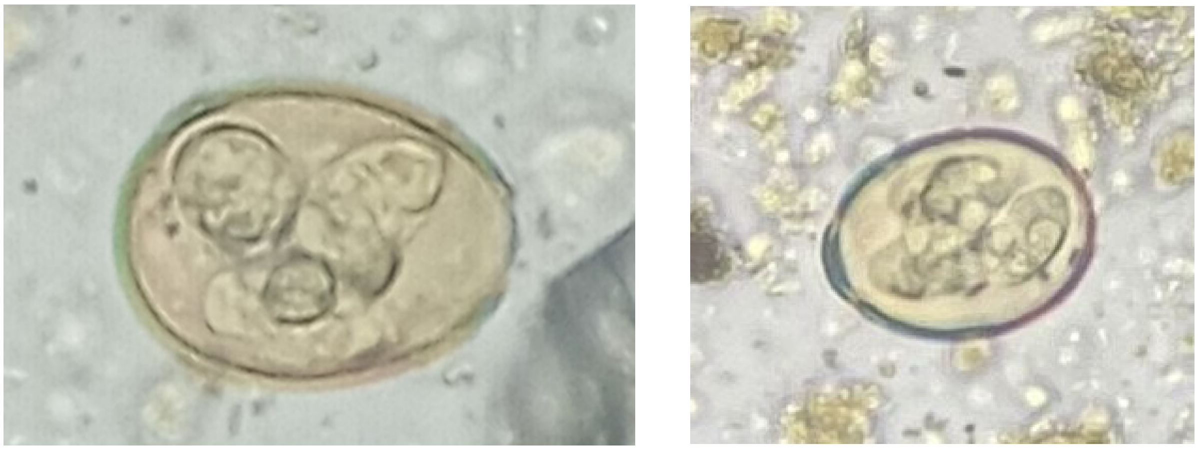
Discussion

The current research was the first to record the prevalence of various GI parasites in dogs and dzos in the complex landscapes of Upper Humla, one of the best representative areas of HKH. The rates of GI parasites in owned dogs vary across different geographical regions; for example, Brazil (11.3%, n=400) (Arruda et al., 2021), Spain (48.8%, n=252) (Mateo et al., 2023), Kenya (65%, n=100) (Mulinge et al., 2020), the high mountain areas of Columbia (43.9%, n=41) (Peña-Quistial et al., 2020) and Algeria (61.07%, n=131) (Ziam et al., 2022). Similarly, the prevalence of GI parasites in free-ranging dogs is variable; for example, Bangladesh (95%; n=60) (Das et al., 2012), India (99%; n=101) (Traub et al., 2014) (90.7%; n=108), (Sudan et al., 2015), Nepal (95.7%; n=332) (Adhikari et al., 2023), South Africa (82.5%; n=240) (Mukaratirwa & Singh, 2010), and Vietnam (55.5%, n=200) (Ng-Nguyen et al., 2015). The

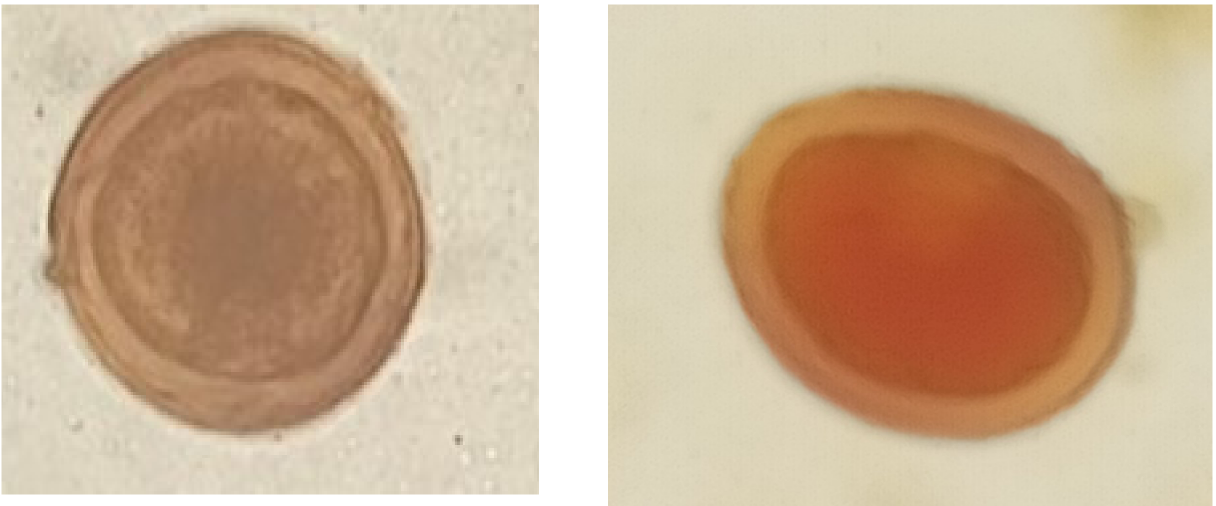


A. Egg of *Hymenolepis nana* (Iodine staining, ×400)

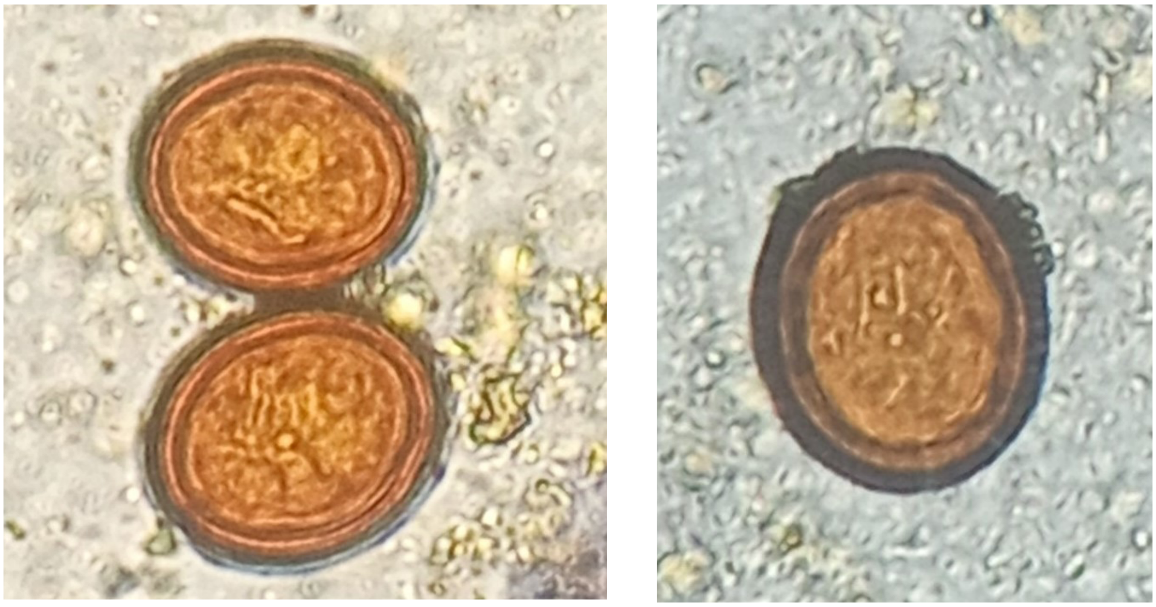
B. Egg of *Trichuris* sp. (Iodine staining, ×400)



C. Oocysts of *Eimeria* spp. (Iodine staining, ×400)



D. Eggs of Ascarid (Iodine staining, ×400)



E. Eggs of tapeworm (Iodine staining, ×400)

Figure 2. Representative parasites in the fecal samples of dogs and dzos

prevalence of GI parasitic rates in dzos has not been reported elsewhere; however, similar species, yaks, have been studied. For example, the GI parasitic rates in yaks ranged from 1.3% to 82.5% (n=40-733) in China (Qin et al., 2019; Chen et al., 2022).

It is difficult to explain such discrepancies in the prevalence rates among different hosts. However, many factors, such as geographical and environmental variations, types of hosts, their breeds and characteristics, history of antiparasitic treatment and its routine strategies, study design, sampling methods, sample collection techniques, microscopic assay tools and techniques, and other unknown variables can play a role (Soulsby, 2012; Uiterwijk et al., 2019; Adhikari et al., 2023; Adhikari et al., 2024). Among these variables, coproscopic techniques play a direct role in parasite detection efficiency. For instance, the employment of different coproscopic techniques can produce a variety of results (Kotwa et al., 2021). In this context, the direct wet mount technique may be less specific; however, the modified Ziehl-Neelsen technique produces highly specificity results for coccidian parasites. Although the modified Ziehl-Neelsen technique is less sensitive compared to enzyme-linked immunosorbent assay (ELISA) and polymerase chain reaction (PCR) techniques, it is advantageous as it indicates only active *Cryptosporidium* infections. Therefore, in the absence of gold-standard techniques, staining methods represent the best option for *Cryptosporidium* detection.

Several underlying causes contribute to the high prevalence and parasite loads in the studied population. First, Upper Humla is a rural area with limited access to healthcare facilities, including veterinary care. This lack of infrastructure for regular medical care, as well as for timely deworming of humans, livestock, and dogs, could facilitate the spread of disease among these hosts. In contrast to developed countries where authorities and people engage in preventive measures and seek veterinary care when needed, such practices are often neglected in poorer and less developed areas. Dzos' feces are used for fuel and manure. Dogs are free-ranging, and with the frequent movement of people and livestock in these municipalities, they pose significant health threats. This is further evidenced by the presence of seventeen species of GI parasites in free-ranging dogs, compared to seven and nine GI parasites in owned dogs and dzos, respectively, in the current study.

Secondly, both the Namkha and Simkot areas lack proper management of owned and free-ranging dogs, with inadequate sewage and fecal management, allowing dogs to pose a significant health threat to people and livestock. Thus, free-ranging dogs are important bridge hosts in the current study sites among domestic pets, livestock, and nearby wildlife. For example, free-ranging dogs come into contact with wildlife, including jackals, foxes and wild cats, such as snow leopards. In addition, they also interact with pet dogs. Although owned dogs are often confined to homes during the day, they are allowed to roam at night. During these nocturnal excursions, they

Table 1. Prevalence of GI parasites in domestic dogs, stray dogs and dzos (P<0.0001)

Parasites	No. (%)		
	Domestic Dogs (Positive)	Stray Dogs (Positive)	Dzos (Positive)
Cryptosporidium spp.	8(66.67)	64(58.72)	19(63.33)
Ascarid	4(33.33)	37(33.94)	17(56.67)
Entamoeba spp.	4(33.33)	23(21)	17(56.67)
E. coli	0	19(17.43)	0
Eimeria spp.	2(16.67)	13(11.92)	14(46.67)
Balantidium coli	0	11(10.1)	0
Sarcocystis spp.	0	9(8.26)	0
Taenia spp.	1(8.33)	8(7.33)	6(20)
N. caninum	1(8.33)	8(7.33)	0
Acanthocephalid	0	4(3.67)	0
Isospora spp.	0	3(2.75)	0
Strongyloides sp.	0	2(1.83)	0
Trichuris sp.	0	2(1.83)	0
Cyclospora sp.	0	1(0.92)	7(23.33)
Ancylostoma	0	1(0.92)	0
H. nana	0	1(0.92)	0
Blastocystis sp.	2(16.67)	0	1(3.33)
Giardia sp.	0	0	1(3.33)
Unknown coccidia	0	1(0.92)	3(10.00)

Note: P<0.0001 (chi-square tests) when comparing the rates of GI parasites among specific hosts, including domestic dogs, stray dogs and dzos.

may encounter free-ranging dogs, potentially facilitating the spread of parasites between these two groups. The existence of a domestic and sylvatic cycle of tapeworms and other infestations is linked to wild carnivores (Abdybekova et al., 2012; Jannat et al., 2020), indicating a critical zoonotic risk factor.

It is important to note that the sharing of common niches and direct contact in the grazing environments of livestock, like dzos, can result in the spread of parasites. The roles of dzos in the transmission of GI parasites must be addressed in this context. Although the dried form of dung is usually used as fuel and manure, handling during drying processes or distribution in the fields may pose a risk for potential GI parasite transmission. For example, the shared GI parasites recorded in free-ranging and owned dogs in this study included *Entamoeba* spp., ascarid worms, *Cryptosporidium* spp., *Eimeria* spp., *Taenia* spp., and *N. caninum*. Furthermore, dzos have also been found to harbor these parasites, indicating that a route of GI parasite transmission exists in the study area.

Cryptosporidium spp. are significant coccidian parasites, with up to three mixed species of other GI parasites shared among the three hosts. This suggests that these coccidian species infect all three hosts with high intensity. A review has identified 12 species or genotypes of *Cryptosporidium* spp. in yaks, including *C. parvum*, *C. hominis*, *C. ubiquitum*, *C. bovis*, *C. ryanae*, *C. baileyi*, *C. andersoni*, *C. canis*, *C. struthionis* and *C. xiaoi*, with

Table 2. Intensity of intestinal parasites (%) in domestic dogs, stray dogs, and dzos

Parasitic Species		No. (%)											
		Domestic dogs (++ vs +++: P<0.0001, 95% CI, 1.000%, 1.000%)			Stray Dogs (+ vs. ++: P<0.05, 95% CI, 0.044%, 0.783%)(++ vs +++: P<0.0001, 95% CI, 0.9732%, 0.9962%)			Dzos (++ vs +++: P<0.0001, 95% CI, 1.000%, 1.000%)			Total (++ vs +++: P<0.0005, 95% CI, 0.39%, 0.891%)		
		+	++	+++	+	++	+++	+	++	+++	+	++	+++
Protozoa	*Cryptosporidium* spp.	3(25)	1(8.33)	4(33.33)	20(18.34)	14(12.84)	30(27.52)	3(10)	6(30)	10(33.34)	26(17.21)	21(13.90)	44(29.13)
	Entamoeba spp.	4(33.33)	0	0	23(21.10)	0	0	17(56.67)	0	0	44(29.13)	0	0
	E. coli	0	0	0	19(17.43)	0	0	0	0	0	19(17.43)	0	0
	Eimeria spp.	2(16.67)	0	0	13(11.92)	0	0)	14(46.67)	0	0	29(19.20)	0	0
	B. coli	0	0	0	11(10.1)	0	0	0	0	0	11(7.28)	0	0
	Sarcocystis spp.	1(8.33)	0	0	9(8.26)	0	0	0	0	0	9(5.96)	0	0
	N. caninum	1(8.33)	0	0	8(7.34)	0	0	0	0	0	9(5.96)	0	0
	Cyclospora sp.	0	0	0	1(0.92)	0	0	7(23.33)	0	0	8(5.29)	0	0
	Isospora spp.	0	0	0	3(2.75)	0	0	0	0	0	3(1.98)	0	0
	Blastocystis sp.	2(16.67)	0	0	0	0	0	1(3.33)	0	0	3(1.98)	0	0
	Giardia sp.	0	0	0	0	0	0	1(3.33)	0	0	1(0.66)	0	0
	Unknown coccidia	0	0	0	1(0.92)	0	0	3(10)	0	0	4(2.65)	0	0
Nema-toda	*Ascarid*	4(33.33)	0	0	35(32.11)	2(1.83)	0	17(56.67)	0	0	56(37.08)	2(1.83)	0
	Ancylostoma	0	0	0	1(0.92)	0	0	0	0	0	1(0.66)	0	0
	Strongyloides sp.	0	0	0	2(1.84)	0	0	0	0	0	2(1.32)	0	0
	Trichuris sp.	0	0	0	2(1.84)	0	0	0	0	0	2(1.32)	0	0
Cestoda	*Taenia* spp.	1(8.33)	0	0	8(7.34)	0	0	6(20)	0	0	15(9.93)	0	0
	H. nana	0	0	0	1(0.92)	0	0	0	0	0	1(0.66)	0)	0
Acantho-cephala	*Acanthocephalid*	0	0	0	4(3.67)	0	0	0	0	0	4(2.64)	0	0

Table 3. Co-infection of intestinal parasites in domestic dogs, stray dogs and dzos (P<0.0001)

Acanthocephala	No. (%)		
	Domestic Dogs	Stray Dogs	Dzos
Single	1(8.33)	17(15.60)	2(6.67)
Double	3(25)	22(20.18)	5(16.67)
Triple	1(8.33)	25(22.93)	1(3.33)
Quadruple	2(16.67)	12(11)	6(20)
Pentuple	1(8.33)	4(3.67)	7(23.33)
Sextuple	0	1(1.22)	2(6.67)
Septuple	0	1(1.22)	1(3.33)

Note: P<0.0001 when comparing the rates of co-infection within a single host and among domestic dogs, stray dogs and dzos.

the first three species having zoonotic potential (Ryan et al., 2021; Geng et al., 2021). In this context, these coccidia can serve as zoonotic sources for nearby hosts via rainfall-mediated transportation of infected dung at high altitudes.

Regarding mixed infections, the current Dzo population showed a higher GI parasitic load. For example, pentuplet parasitic infections were more prevalent than quadruplet, duplet, triplet, sextuplet, and septuplet GI parasitic infections. Although the pathogenic consequences of this heavy infestation of GI parasites cannot be ignored, multiple coinfections may also lead to positive, negative, or null effects. These effects may primarily occur by altering host susceptibility to other parasitic species (Viney & Graham, 2013). For example, helminth infections can downregulate host resistance to Mycobacterium tuberculosis, HIV, and *Plasmodium* spp. (Salgame et al., 2013), as well as to vaccine-induced immunity for BCG (Elias et al., 2001) and TT(Sabin et al., 1996). However, poly parasitized cats showed reduced Toxocara loads (Serrano & Millan, 2014) indicating negative interactions due to mixed infections. In cases of null effects, there will be no influence of one parasite on others. Further studies should be conducted to investigate how this coinfection affects the dzos and associated interacting hosts at high altitudes in the future. Notably, out of 1415 human pathogens, 62% have been identified as zoonotic in origin (Billinis, 2013) and wild animals serve as principal reservoirs for many zoonoses that can affect domestic animals and humans (Thompson, 2013). Therefore, understanding the interactions of intestinal pathogens within these populations, including feral dogs and domestic animals like dzos, will provide guidelines for managing spillover mechanisms.

Conclusion

This is the first study documenting GI parasites in dogs and dzos in Humla. This microscopic study documented intestinal parasites in dogs and dzos at high altitudes in Nepal, representing HKH regions that are predominant for GI parasitic infections. The hosts documented in this study shared GI parasites, including *Entamoeba* spp., ascarid worms, *Cryptosporidium* sp., *Eimeria* spp., and *Taenia* spp. These GI parasites are critically zoonotic and may be transmitted among the associated human population. However, further studies are needed to investigate how these GI parasites exist, survive, spread, and cause pathologies, as well as their epidemiology in complex landscapes such as the current study area in the HKH.

Ethical Considerations

Compliance with ethical guidelines

The necessary permission for collecting fecal samples was issued by the Department of Forest (Permit 321-2078/079), Babarmahal, Kathmandu, as well as by Namkha Rural Municipality (Permit 288-2079/080) and Simkot Rural Municipality (Permit 818-2079/080) in Humla district, Nepal. The study was conducted using stool samples defecated on the surface, and no experimental infections in dogs and dzos were established during the research. None of the animals were touched or harmed in the study.

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Authors' contributions

Laboratory survey and writing the original draft: Dharma Acharya; Funding acquisition and resources: Rinzin Phunjok Lama; Laboratory analysis: Tirth Raj Ghimire; Methodology: Dharma Acharya and Rinzin Phunjok Lama; Supervision: Rinzin Phunjok Lama, and Tirth Raj Ghimire; Conceptualization, data analysis, review, editing and final approval: All authors.

Conflict of interest

The authors declared no conflict of interest.

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