

**Research Article:****EPIDEMIOLOGY OF ZONOTIC GASTROINTESTINAL HELMINTHS IN STRAY AND PET DOGS OF DHANGADHI, NEPAL****Laxman Bhatta** , **Shekhar Pokhrel**  and **Rupak Khatri-Chhetri\*** 

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**ABSTRACT**

Dogs act as reservoirs of zoonotic gastrointestinal helminths, posing a significant public health risk. A cross-sectional study was conducted in Dhangadhi City, where fecal samples from 400 dogs (200 pet, 200 stray) were examined for gastrointestinal helminths. Of all samples, 173 (43.25%) fecal samples showed the presence of at least one parasite. Stray dogs showed a significantly higher prevalence ( $p < 0.001$ ) of helminths (63.5%) than pets (23%). The zoonotic helminths identified included: *Ancylostoma* spp. (19.25%), *Toxocara* spp. (18.50%), *Taenia* spp. (5.75%), and *Dipylidium* spp. (9.75%). The helminth prevalence was slightly higher ( $p > 0.05$ ) in pet dogs under 1 year (25%) compared to those over 1 year (22.4%). Single helminth infection was statistically significantly ( $p < 0.05$ ) higher (77.45%) than concurrent mixed infection (22.54%). Helminth prevalence was slightly higher ( $p > 0.05$ ) in male dogs than in females. The prevalence of zoonotic helminths was statistically significantly greater ( $p < 0.05$ ) in non-dewormed dogs (73.9%) than in dewormed dogs (16.4%). Helminth infection was strongly associated with deworming history. The KAP survey response suggests most dog owners (88.5%) are aware of the zoonotic risk associated with dogs, and deworm their dogs within 3-6 months. Significant associations were found between gastrointestinal helminth infection and both playing behavior ( $p < 0.05$ ) and deworming practice ( $p < 0.001$ ). However, no statistically significant association was observed between the presence of helminth infection and zoonotic awareness, rearing system, and veterinary consultation practices. Responsible ownership and routine deworming practices should be followed to minimize the public health risk associated with zoonotic helminths of dogs.

**Keywords:** Canines, deworming, feces, parasite, prevalence**INTRODUCTION**

Humans and dogs have lived together for thousands of years, and dogs have played important social, economic, and cultural roles in society (McNicholas & Collis, 2000). In recent years, dog ownership in Dhangadhi has increased, and closer interaction between humans and dogs has increased the risk of zoonotic parasite transmission to humans. Dogs act as reservoirs of zoonotic diseases and also transmit to other healthy dogs and humans (Stull et al., 2015). These parasites can cause gastrointestinal distress, organ damage, and morbidity, which can lead to a significant socioeconomic burden. This dual role has become a pressing concern despite the numerous benefits of owning a dog.

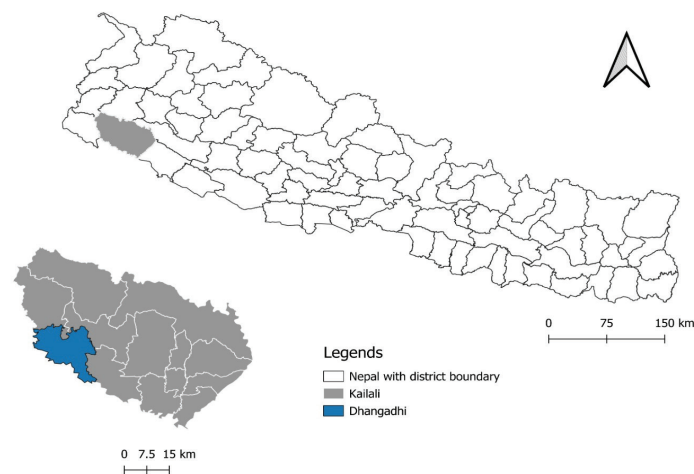
Information on the prevalence of zoonotic gastrointestinal parasites in dogs and their transmission to humans is valuable and can assist in designing control strategies, intervention methods, and public health planning using the “One-Health” approach (Mughini-Gras et al., 2016; Zinsstag et

al., 2020). However, limited studies have been conducted on zoonotic gastrointestinal parasites of dogs and their awareness level among pet owners of Dhangadhi, which creates a gap in controlling them and safeguarding public health. Therefore, this study aimed to investigate the various gastrointestinal zoonotic helminths in pet and stray dogs of Dhangadhi sub-metropolitan city and calculate the prevalence rate and its associated risk factors. Additionally, it assessed the knowledge, attitude, and practices of dog owners of Dhangadhi regarding gastrointestinal zoonotic parasites.

## RESEARCH METHODS

### Study area:

The study was carried out in Dhangadhi sub-metropolitan city, which lies in Sudurpaschim Province of Nepal (Fig. 1). It has a total area of 3,235 km<sup>2</sup> and a total population of 911,155, according to the 2021 census.



**Fig. 1. Map of Nepal highlighting the Kailali district and the specific study sites within Dhangadhi, the study area for the present study.**

### Study design, sample size, and data collection:

A cross-sectional prevalence study was conducted in the study area from April to June 2023 to determine the prevalence of important zoonotic gastrointestinal parasites in pet and stray dogs. The sample size for the study was calculated by using the OpenEPI tool (Sullivan et al., 2009), with an expected prevalence of 58.75% (Yadav & Shrestha, 2017) and a desired absolute precision of  $\pm 5\%$ , and it was found to be 372, which was later rounded off to make 400.

A purposive sampling method was used for this study to cover the overall parts of Dhangadhi sub-metropolitan city. Freshly voided fecal samples (200 from pet and 200 from stray dogs) were collected from the dogs, and feces were placed in a zipper bag and transported in a cold box to the regional veterinary laboratory, Dhangadhi, for microscopic examination.

In addition to this, it also included a community survey with a semi-structured questionnaire to explore dog owners' general knowledge of dogs, their understanding of zoonotic diseases, housing, defecation practices, deworming practices, and other management strategies. A pilot survey of 10 owners was conducted, and some of the questions were modified according to their responses. The actual questionnaire survey was conducted only after written consent was taken from each dog owner, and their names and identification were kept secret throughout the study and not published anywhere. The interview was conducted in the local language, and the later answers were translated into English.

### **Procedure for fecal examination**

Fecal samples were examined using the floatation method (Eleni et al., 2011), which relies on the differences in specific gravities between eggs, fecal debris, and the floatation solution, since most zoonotic helminth parasites have specific gravities below 1.20. Briefly, feces (approximately 3 grams) were mixed with distilled water (10 ml) and filtered, and the filtrate was centrifuged at 2500 rpm for 5 minutes in a 15 ml tube. The supernatant was discarded, and floatation fluid was added to the centrifuge tube. The floatation fluid was prepared by mixing approximately 400 g of NaCl in 1000 ml of distilled water at 35 °C (Hansen & Perry 1994). The resulting liquid was further centrifuged at 2500 rpm for an additional 5 minutes. Finally, a coverslip was placed on top and left for 5 minutes, and transferred to a microscopic slide and examined under the microscope. Likewise, the sediment was also examined for parasite eggs. Eggs were identified by evaluating the characteristics of the eggs under a microscope (Soulsby, 1982).

### **Data management and statistical analysis**

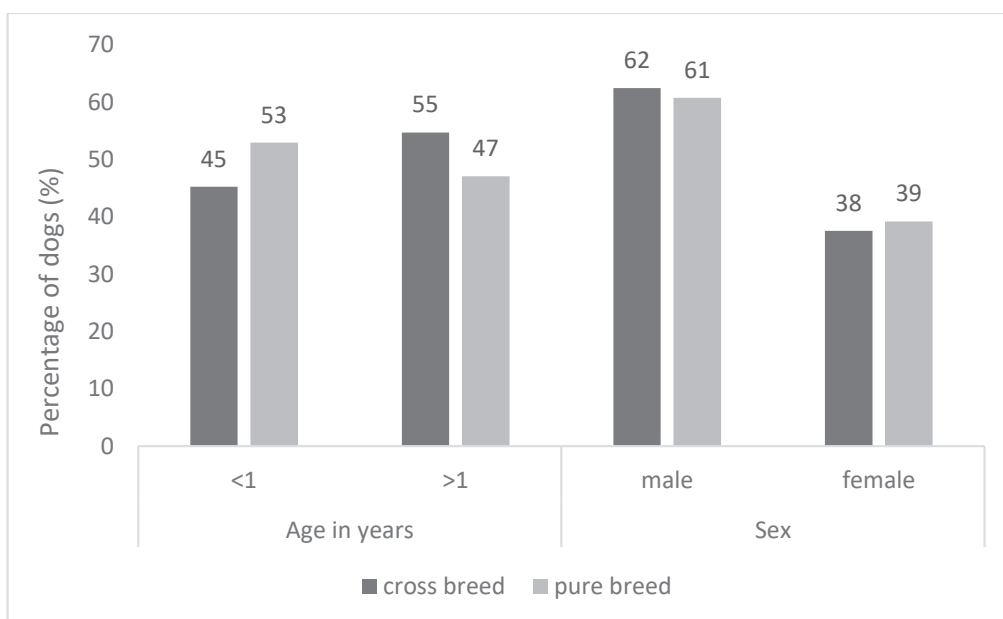
All statistical analyses (prevalence, Pearson chi-square test, odds ratio, relative risk, and 95% exact binomial confidence intervals) were calculated using R Statistical Software (version 4.5.2, R core team 2025). The significance of differences between groups was evaluated using Pearson's Chi-Square test, where a value of  $p < 0.05$  was considered significant at 95% level of confidence. Furthermore, a multivariate logistic regression model was fitted using dependent variables (presence of parasite- Yes/No) and predictor variables (Dog type, sex, age, breed, deworming history), and adjusted odds ratios (AOR) and 95% CI were estimated.

The association between the presence/absence of gastrointestinal helminths and KAP-related variables in pet dogs was analyzed by the chi-square test. In addition, the association between zoonotic awareness and management practices (deworming practices and rearing system) was also analyzed.

## **RESULTS AND DISCUSSION**

### **Demographics**

Among 400 dogs, 346 were crossbreed dogs, and 54 were purebred dogs. Among cross-breed dogs, 45% were less than one year of age, while the remaining were one or more years old. However, 53% of pure-breed dogs were less than one year of age. The sex ratio was almost identical in both breed types, with 62% males in the cross-breed and 61% males in the pure-breed, respectively (Fig. 2).



**Fig. 2. Demographics of the dogs based on age, sex, and breed**

### Overall prevalence of zoonotic helminths

The overall prevalence of gastrointestinal parasites was found to be 43.25%, with a prevalence of 23% and 63.5% in pet and stray dogs, respectively (Table 1). Among those 173 positive cases, 77.45 % showed single parasite infection, whereas 22.54 % had infection with multiple parasites. There was a statistically significant difference in prevalence between pet and stray dogs ( $\chi^2 = 68.17$ ,  $p < 0.001$ ), where stray dogs are almost 3 times more likely in risk of getting helminth infection than pet dogs.

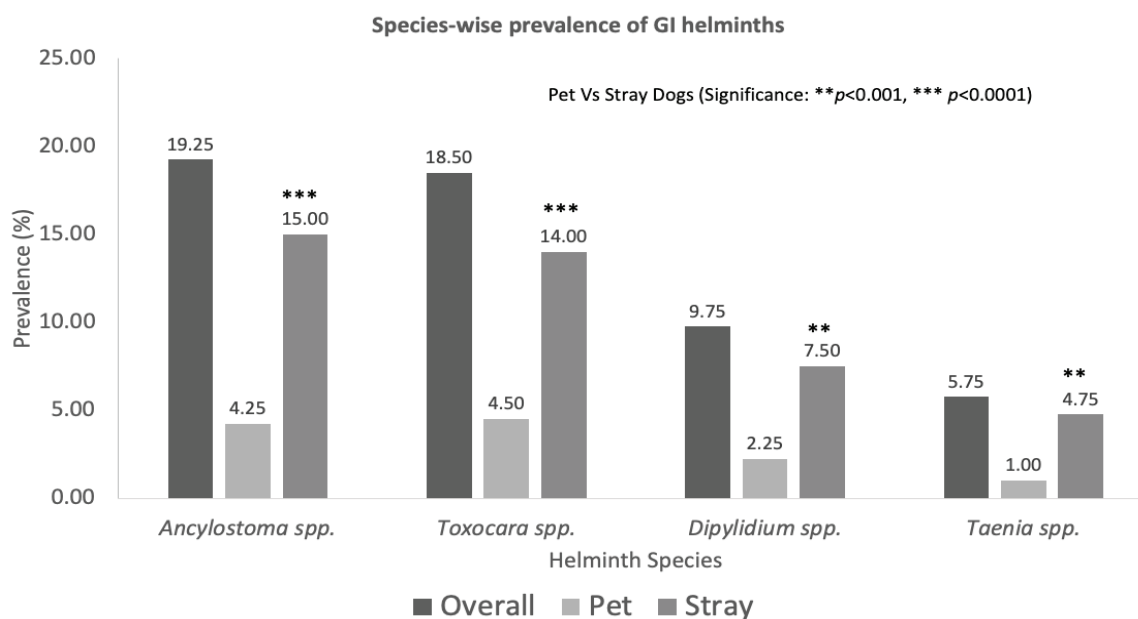
**Table 1. Prevalence of helminths in pet and stray dogs**

Deworming practice	Total (n)	Positive	Negative	Prevalence % (95% CI)	df	$\chi^2$ , p value	OR (95% CI)	RR (95% CI)
Stray	200	127	73	63.50 (56.4-70.2)	1	$\chi^2 = 69.73$ , $p < 0.0001$	5.83 (3.76-9.05)	2.76 (2.10-3.63)
Pet	200	46	154	23 (17.4-29.4)				
Total	400	173	227	43.25 (38.3-48.3)				

OR- Odds Ratio, CI- Confidence Interval, RR (Relative Risk)

### Species-wise prevalence of helminths

The highest prevalence was observed for *Ancylostoma* spp., accounting for 19%. In second place were *Toxocara* spp., *Dipylidium* spp., and *Taenia* spp., with prevalence rates of 18.5%, 9.75%, and 5.75%, respectively. The prevalence of all helminths was significantly higher in stray dogs compared to the pets (Fig. 3).



**Fig. 3. Prevalence of helminth species in pet and stray dogs**

### Sex wise prevalence of helminths

The prevalence of infection was almost similar in both male and female dogs in Dhangadhi city (Table 2). There was no significant association between sex and helminth infection ( $p=0.88$ ), and this is further supported by a low relative risk ratio ( $RR=1.02$ ). Among male dogs, stray dogs showed significantly higher prevalence (66.94) compared to pet dogs (23.4). Male stray dogs showed a higher risk ( $RR=2.86$ ) of infection compared to male pet dogs. Likewise, among female dogs, the prevalence was also significantly higher in stray dogs (58.23%) than pet dogs (22.03%), with almost three times higher risk of infection compared to female pet dogs ( $RR=2.86$ ).

**Table 2. Sex-wise prevalence of GI parasites in pet and stray dogs**

Deworming practice	Total (n)	Positive	Negative	Prevalence % (95% CI)	df	$\chi^2$ , p value	OR (95% CI)	RR (95% CI)
<u>Male</u>	262	114	148	43.51 (37.50-49.52)	1	$\chi^2 = 0.022$ , $p=0.88$ (M vs F)	1.03 (0.68-1.55)	1.02(0.81-1.29)
Stray	121	81	40	66.94 (58.55-75.33)	1	$\chi^2=53.05$ , $p<0.0001$ (SM vs PM)	6.63 (4-10.98)	2.86 (2.06-3.97)
Pet	141	33	108	23.4 (16.42-30.38)				
<u>Female</u>	138	59	79	42.75 (34.51-50.99)				
Stray	79	46	33	58.23 (47.36-69.10)	1	$\chi^2=19.55$ , $p<0.0001$ (SF vs SM)	4.93 (2.29-10.60)	2.64 (1.59-4.38)
Pet	59	13	46	22.03 (11.46-32.60)				

CI (Confidence Interval), OR (Odds Ratio), RR (Relative Risk), M (Male), F (Female), SM (Stray male), SP (Stray pet), SF (Stray female), SP (Stray pet)

### Age and breed-wise prevalence of helminths.

Regarding age, the prevalence of GI parasites was slightly higher in dogs under 1 year of age (25%) compared to older dogs (22.4%), but the difference was not statistically significant (Table 3). In terms of breed types, the prevalence was higher in crossbred (25.3 %). However, the purebred dogs had a low prevalence of only 16.7 %.

**Table 3. Age and breed-wise prevalence of GI helminths in pet dogs**

	Total (n)	Positive	Negative	Prevalence % (95% CI)	df	$\chi^2$ , p value	RR (95% CI)
<u>Age-group</u>							
≥ 1 year	152	34	118	22.37 (15.95-29.98)	1	$\chi^2 = 0.13,$ $p=0.72$	1.12 (0.63-1.98)
<1 year	48	12	36	25 (13.58-39.64)			
<u>Breed</u>							
Cross-breed	146	37	109	25.34 (18.54-33.20)	1	$\chi^2 = 1.53,$ $p=0.22$	1.52 (0.79-2.94)
Pure-breed	54	9	45	16.67 (7.87-29.11)			

CI (Confidence Interval), RR (Relative Risk)

### Deworming practice and prevalence of zoonotic helminths

The highest prevalence rate among dogs with a deworming history was found in dogs that had been dewormed more than six months prior, and they had a six times higher risk of infection than those dewormed within three months (Table 4). The prevalence of gastrointestinal parasites was directly related to the last time of deworming, and a highly significant association was found between deworming practice and prevalence ( $\chi^2 = 112.36, p < 0.001$ ). Dogs with no deworming recorded showed 8.3 times higher risk of helminth infection than dogs dewormed within three months.

**Table 4. Deworming interval and prevalence of zoonotic helminths in pet dogs**

Deworming practice	Total (n)	Positive	Negative	Prevalence % (95% CI)	p value	Odds Ratio (95% CI)	RR (95% CI)
Within last 3 months	39	3	36	7.7 (2.6-20.3)	$\chi^2 = 112.36,$ $p < 0.001$	0.81(0.19-3.46)	0.82 (0.22-3.12)
Within last 3-6 months	95	6	89	6.0 (2.9-13.1)			
More than 6 months ago	46	22	24	47.8 (33.8-62.2)			
No record	220	141	79	64.1 (57.6-70.1)			

CI (Confidence Interval), OR (Odds Ratio), RR (Relative Risk)

### Multivariate logistic regression results:

Multivariate logistic regression analysis showed that deworming history was significantly associated with gastrointestinal helminth infection. Dogs with no deworming record had 34.47 times higher odds of infection compared to dogs dewormed within the previous 3-6 months (AOR=34.47; 95% CI: 9.59-123.94;  $p < 0.001$ ). Similarly, dogs dewormed more than 6 months previously had significantly higher odds of infection (AOR= 12.02; 95% CI: 4.33-33.35;  $p < 0.001$ ). Dog type, sex, age, and breed were not significantly associated with infection in the adjusted model (Fig. 4).



**Fig. 4.** Forest plot showing adjusted odds ratios (AOR) and 95% confidence intervals for risk factors associated with gastrointestinal helminth infection in dogs based on multivariate logistic regression analysis. The vertical dashed line represents an odds ratio of 1. Variables with confidence intervals not crossing 1 were considered statistically significant.

### Knowledge, attitude, and practices (KAP) of pet owners

In total, 200 dog owners responded to our questionnaires. The major findings are summarized in Table 5. Significant associations were found between gastrointestinal helminth infection and both playing behavior ( $\chi^2 = 8.97$ ,  $p = 0.003$ ) and deworming practice ( $\chi^2 = 40.63$ ,  $p < 0.001$ ). However, no statistically significant association was observed between the presence of helminth infection and zoonotic awareness, rearing system, and veterinary consultation practices ( $p > 0.05$ ) (Table 6).

**Table 5. Knowledge, Attitude, and Practices among Pet Dog Owners in Dhangadhi, Nepal (n=200)**

Variable	Category	Number	Percentage (%)
Knowledge about zoonotic transmission	Yes	177	88.5
	No	23	11.5
Family members playing with their pets	Yes	142	71
	No	58	29
Housing system	Kennel outside the house	89	44.5
	Sharing house	92	46
	Allowed to wander freely	19	9.5
Regularity in deworming practices	Within 3 months	39	19.5
	Within 3 to 6 months	95	47
	More than 6 months	46	22
	No deworming	20	11.5
Veterinary Care	Contacted veterinary technicians	117	58.5
	Consulted registered private veterinarian	41	20.5
	Took to government hospital	42	21

**Table 6. Association between presence of gastrointestinal helminth infection and KAP-related variables among pet dogs (n=200)**

Query	Category	Positive n (%)	Negative n (%)	$\chi^2$ value	p-value
Knowledge about canine helminth zoonosis	Yes	38 (21.5)	139 (78.5)	0.49	0.482
	No	7 (30.4)	16 (69.6)		
Family members playing with their pets	Yes	40 (27.8)	104 (72.2)	8.97	0.003*
	No	5 (8.9)	51 (91.1)		
Rearing system	Indoor	40 (23.4)	131 (76.6)	1.11	0.575
	Outdoor	5 (19.2)	21 (80.8)		
	Free-roaming	0 (0.0)	3 (100.0)		
Regularity in deworming practices	Within 3 months	3 (7.7)	36 (92.3)	40.63	<0.001*
	Within 3 to 6 months	6 (6.3)	89 (93.7)		
	More than 6 months	22 (47.8)	24 (52.2)		
Veterinary Care	Consulted veterinary technicians	25 (21.4)	92 (78.6)	0.42	0.810
	consulted registered private veterinarian	11 (26.2)	31 (73.8)		
	Took to government hospital	9 (22.0)	32 (78.0)		

\*p&lt; 0.05 (indicates statistically significant difference)

## DISCUSSION

In the present study, zoonotic gastrointestinal helminths in pet and stray dogs in Dhangadhi Sub-Metropolitan City, Kailali District, Nepal were investigated by coprological examination. The prevalence of gastrointestinal helminth infections in the examined fecal sample was 43.25%. High prevalence of helminths in dogs has been reported from various regions of Nepal. Notably, 46.7% of dogs in Kathmandu (Satyal et al., 2013), 58.75% in Rupandehi (Yadav & Shrestha, 2017), 57.28% in Chitwan (BK et al., 2025), and 59.5% in Suryabinayak (Sukupayo & Tamang, 2023) were found to be infected. These variations arise from intricate interactions between host factors, environmental factors, and parasite factors (Oyesola et al. 2022).

Stray dogs showed significantly higher prevalence than pet dogs in both sexes, suggesting that they serve as a substantial reservoir of gastrointestinal helminth infection within the Dhangadhi metropolitan city. This is a prevalent global phenomenon, attributed to their scavenging behavior, delayed deworming, compromised health, insufficient veterinary care, and heightened exposure to contaminated soil and intermediate hosts (Martínez-Moreno et al., 2007; Robertson et al., 2000; Traub et al., 2002). In contrast, companion dogs are well managed with better nutrition, routine deworming, and veterinary care, are confined most of the time indoors, and thus the risk of exposure to zoonotic helminths and the infection burden is minimal (Howell et al., 2016). Similar observations have been found in several studies where free-roaming and stray dogs have been found to show a higher prevalence of zoonotic helminths compared to pet dogs (Cho et al., 1981; Davoust et al., 2008; Jovanovic et al., 2024; Otranto et al., 2017; Sukupayo & Tamang, 2023). It is noteworthy that after adjusting for multivariate logistic regression, there was no longer a statistically significant association between the prevalence of helminths in stray and pet dogs. This suggests that the crude association may have been confounded by variations in deworming practices between pet and stray dogs.

The frequency of GI zoonotic helminths was higher in crossbred pet dogs (25.3%) than in purebred pet dogs (16.7%). Purebred dogs typically receive good husbandry, veterinary care, and are given proper nutrition (Cho et al., 1981).

Helminth prevalence was slightly higher in dogs aged < 1 year (25%) compared to dogs aged  $\geq$  1 year (22.37%). Several factors, such as underdeveloped immunity in young animals, increase susceptibility to infection, particularly to hookworms, which can be transmitted through transplacental or transmammary routes (Bourgoin et al., 2022; Zajac & Conboy, 2012). However, in the present study, the difference was small, indicating both age groups could have been exposed to similar environmental sources of infection in the study area.

In relation to gender disparities, male domestic dogs exhibited a marginally higher prevalence of zoonotic helminth parasites (23.40%) compared to female pet dogs (22.03%). Stray dogs exhibited a comparable ratio, with 66.94% males and 58.22% females. Several other studies have reported a higher prevalence of GI helminths in males (Biu et al., 2012; Getahun & Addis, 2012; Mirzaei & Fooladi, 2012). The present study demonstrated that gastrointestinal helminth infection was not significantly influenced by sex, as both male and female dogs exhibited comparable prevalence, and differences were not statistically significant. This indicates that dogs of both sexes in the study area may have similar exposure to contaminated environments and infection sources.

In the present research, single helminth infections were more prevalent (77.45%) than concurrent mixed infections (22.54%). Similar findings have been reported: 31.4% - single infection and

18.5% - mixed (Katagiri & Oliveira-Sequeira, 2008), 73.8% - single and 12.8% - mixed (Swai et al., 2010), and 53.9% - single and 46.11% - mixed (Bastakoti et al., 2023). In contrast, a higher rate of concurrent mixed infection has also been reported elsewhere (Zewdu et al., 2010). This variation could be attributed to differences in knowledge about canine parasites, regular deworming practices, housing conditions, and other management practices (Cho et al., 1981; Minnaar et al., 2002).

Helminth infection was strongly associated with deworming history. The lowest prevalence of infection was found in dogs dewormed within the last 3 months. In contrast, dogs dewormed more than 6 months ago showed markedly higher prevalence, and dogs with no deworming record exhibited the highest prevalence and risk of infection. Importantly, the association remained statistically significant after adjustment for potential confounding variables. Canine anthelmintics were effective, with significantly fewer GI zoonotic helminths in dewormed (16.4%) than non-dewormed pet dogs (73.9%), consistent with a study in Kathmandu (Satyal et al., 2013). Prevalence was lowest in dogs dewormed every three months (9.3%) and highest in those dewormed at intervals exceeding six months (44.2%), reflecting diminishing anthelmintic effectiveness (Bahrami et al., 2011). The findings emphasize the important role of routine deworming in reducing helminth infection in dogs.

In the present study, all four helminths showed significantly higher prevalence in strays than in pets, and are high-risk for zoonotic transmission, especially *Ancylostoma* and *Toxocara*. *Ancylostoma* spp. (19.25%), *Toxocara* spp. (18.50%), *Dipylidium* spp. (9.75%), and *Taenia* spp. (5.75%) were the most common parasites identified in the current research. These helminth parasites are common findings in coprological examination of dogs worldwide (Mahdy et al., 2012; Singh et al., 2022; Swai et al., 2010; Zewdu et al., 2010).

The KAP survey response suggests most dog owners (88.5%) are aware of the zoonotic risk associated with dogs. Despite this high awareness, gastrointestinal helminth infection was still detected among pet dogs, suggesting that awareness alone may not ensure total control. Most owners responded to administering anthelmintics within 3-6 months. A highly significant association was observed between deworming history and parasitic infection, where the dogs with irregular or absent deworming practices showed significantly higher infection prevalence. Playing behaviour was also significantly associated with infection status, indicating that increased interaction and environmental exposure may facilitate transmission of infection (Tylkowska et al. 2024).

The findings of the present study highlight the need for greater public and dog owner knowledge of zoonotic gastrointestinal helminth infections and the dangers they pose. Interventions in public health awareness should encourage responsible pet ownership, which includes routine deworming, appropriate feces management, and cleanliness practices. Some limitations of the present study should be taken into consideration while interpreting the findings. The present study has unequal representation of sub-groups, which may have affected the robustness of comparisons between categories. However, the observed distribution likely reflects the actual demographic composition of the sampled population. No dog-level metadata (body condition, exact roaming range) or environmental samples were obtained in the present study, which should be taken into account during future parasite surveys in dogs of Dhangadhi. Only the floatation method using saturated sodium chloride was used for identifying the helminths, which may have missed some trematodes, heavy eggs, or protozoa. Future research employing multiple parasitological techniques, such as Baermann's method and sedimentation methods, may help to identify more parasite species, which might have been missed by the present

floatation method. Moreover, only microscopic examination was done in this study. More robust diagnostic techniques, such as Polymerase Chain Reaction, can help confirm the exact parasite species (Tavares et al., 2011; Verweij, 2014). The class of anthelmintic drugs administered to the dogs could not be verified in the study. Future studies should try to address this limitation, as variation in the efficacy of anthelmintics and anthelmintic resistance may also affect prevalence.

### CONCLUSION

The findings of the present study highlight the significance of stray dogs in the transmission of GI helminths in the Dhangadhi sub-metropolitan city. Therefore, appropriate control strategies should be implemented to mitigate the risk of transmission. Regular deworming of pet dogs, client education on responsible dog ownership, improved sanitation, and proper disposal of feces should be advocated. Moreover, the stray dog population should be managed by periodic mass deworming that can help reduce infection pressure and minimize environmental contamination.

### ACKNOWLEDGEMENTS

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### AUTHOR CONTRIBUTIONS

**LB:** Conceptualization, Investigation, Writing – original draft; **SP:** Data curation, Writing – original draft, Writing – review & editing; **RKC:** Conceptualization, Methodology, Formal analysis, Writing – review & editing.

### CONFLICT OF INTEREST

The authors declare that they have no known competing financial interests or personal relationships that could have appeared to influence the work reported in this paper.

### ETHICAL APPROVAL AND PERMITS

In the present study, naturally voided fecal samples were collected from the dogs. Therefore, separate approval was not applied for ethics board approval.

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