Research Article

Risk and protective factors associated with gastrointestinal parasites of dogs from an urban area of Córdoba, Argentina

Carlos Eugenio MOTTA[1] , María Romina RIVERO[2] , Carlos Daniel DE ANGELO[3] ,
Ana María SBAFFO[1] , Karina Ivana TIRANTI[1],*

[1]Department of Animal Pathology, Faculty of Agronomy and Veterinary, National University of Río Cuarto, Río Cuarto, Córdoba, Argentina
[2]National Council of Scientific and Technical Research, National Institute of Tropical Medicine, Puerto Iguazú, Misiones, Argentina
[3]National Council of Scientific and Technical Research, Subtropical Biology Institute, Puerto Iguazú, Misiones, Argentina

Abstract: Prevalence of gastrointestinal (GI) parasites and the effect of associated factors were evaluated in household dogs from an urban area of Córdoba, Argentina. A total of 493 fecal samples were collected during 2010 and 2013 and processed with Willis' salt, Sheather's sugar flotation, and formol-ether concentration techniques. Overall prevalence of GI parasites was 45.23% (95% CI 40.83–49.62), and *Ancylostoma caninum* (30.83%) was the most frequent parasite, followed by *Trichuris vulpis* (9.94%), *Cystoisospora* spp. (7.71%), *Toxocara canis* (6.90%), and *Giardia* spp. (5.88%). Independent variables were presence of at least one parasite element, named "all parasites", and each GI parasite more frequently detected was analyzed as separate outcomes. Logistic regression results showed an increased risk related to age (P = 0.0343) for all parasites and for *T. canis*, *Cystoisospora* spp., and *Giardia* spp. Statistically significant and protective variables were no ingestion of small animals, daily feces removal, and absence of shadow in house yards. Use of anthelmintics and daily feces removal for *A. caninum*, being female, and absence of shadow for *T. canis* resulted in protective factors. The role played by veterinarians and owners' commitment as key aspects, effective actions for prevention, control and treatment are required in the research area.

Key words: Dogs, parasites, prevalence, risk factors, protective factors

1. Introduction

Many parasitic, bacterial, fungal and viral pathogens provoke diseases, being transmitted to humans from domestic pets and vice versa. The domestic dog (*Canis familiaris*) maintains close contact with humans, and potential health risk to humans of enteric parasites harbored by pets remains a significant problem worldwide. In this sense, parasitic zoonotic diseases acquire special importance due to the morbidity burden (1). Therefore, a wide spectrum of investigations based on dogs' health, sanitary, and parasitic status have been conducted in the last decades and have been a substantial matter of study especially in endemic areas (2,3). Main risk factors described were age, sex, breed, neutering status, antiparasitic use, owners' socioeconomic status, and seasonal and spatial variation, from among others (4-8). In Argentina, previous studies determined the overall prevalence of GI parasitism in dogs' feces in different locations, with values ranging from 36% to 68% (8-12).

Age, breed, and seasonal and spatial patterns have been related to GI parasites of dogs, with high prevalence of *Ancylostoma caninum* in males. For other parasites, such as *Toxocara canis*, *Isospora canis*, *Isospora ohioensis* complex, *Hammondia–Neospora* complex, *Sarcocystis* spp., and *Giardia duodenalis*, prevalence decreased with increasing host age, while for *A. caninum* and *Trichuris vulpis*, an inverse pattern was found. Also, prevalence of three protozoa (*Isospora ohioensis* complex, *Sarcocystis* spp., and *Giardia duodenalis*) was significantly higher in purebred dogs, while *A. caninum* and *T. vulpis* were higher in mixed-breed dogs. Spatial patterns and seasonal variation have also been previously described in Argentina (8).

The aim of this study was to determine the prevalence of GI parasites in owned dogs from two municipalities of the Greater Rio Cuarto area, Córdoba, Argentina. Furthermore, the presence of at least one parasite and main parasite elements were evaluated regarding age, sex, household characteristics, and dogs' habits.

2. Material and methods

2.1. Study area

The study was conducted in the Great Rio Cuarto area located in the southern region of Córdoba province. This area includes two cities: Rio Cuarto and Las Higueras with a total population of 163,048 inhabitants (13) comprising 64.25 km² both. The area is characterized by temperate climate with well-defined four seasons. The study area was subdivided into six sections, five located in Rio Cuarto, the main city, and one in Las Higueras (Figure 1).

2.2. Sampling

Fecal samples from dogs were collected between March 2010 and November 2013 and preserved in 10% formalin. Demographic data and anthelmintic history were gathered for each animal sampled. A questionnaire was provided to the owners to collect information on factors likely to impact the prevalence of GI parasites. Variables gathered for each animal were the number of dogs in a household; presence of other animals; predation of other animals (birds, rodents) by the pet; ingestion of raw meat; presence of feces in household, if feces were observed the day the survey was done; if owner had previous knowledge of zoonotic diseases; whether the pet spent time inside or outside the house/household; outdoor activities, presence of grass or sand; presence of shadow; frequency of feces disposal; and antiparasitic treatment history. Dogs' age was categorized as less than one year and greater than or equal to one year.

2.3. Laboratory methods

Samples were processed in the parasitology laboratory of the Animal Pathology Department, Veterinary and Agronomy Faculty, National University of Rio Cuarto (UNRC). The methods used were Willis' salt flotation (14), Sheather's sugar flotation, and formol-ether concentration techniques (15).

Nematode eggs and protozoan oocysts were observed in both flotation methods, while cestode eggs and *Giardia* cysts were identified by observation of the sediment of the formol-ether concentration technique. *Giardia* cysts were recognized based on morphology characteristics. In order to differentiate *Ancylostoma* and *Uncinaria* eggs, previous studies using larvae culture showed that 100% of larvae of 3rd stage corresponded to the genus *Ancylostoma* (Motta, personal communication).
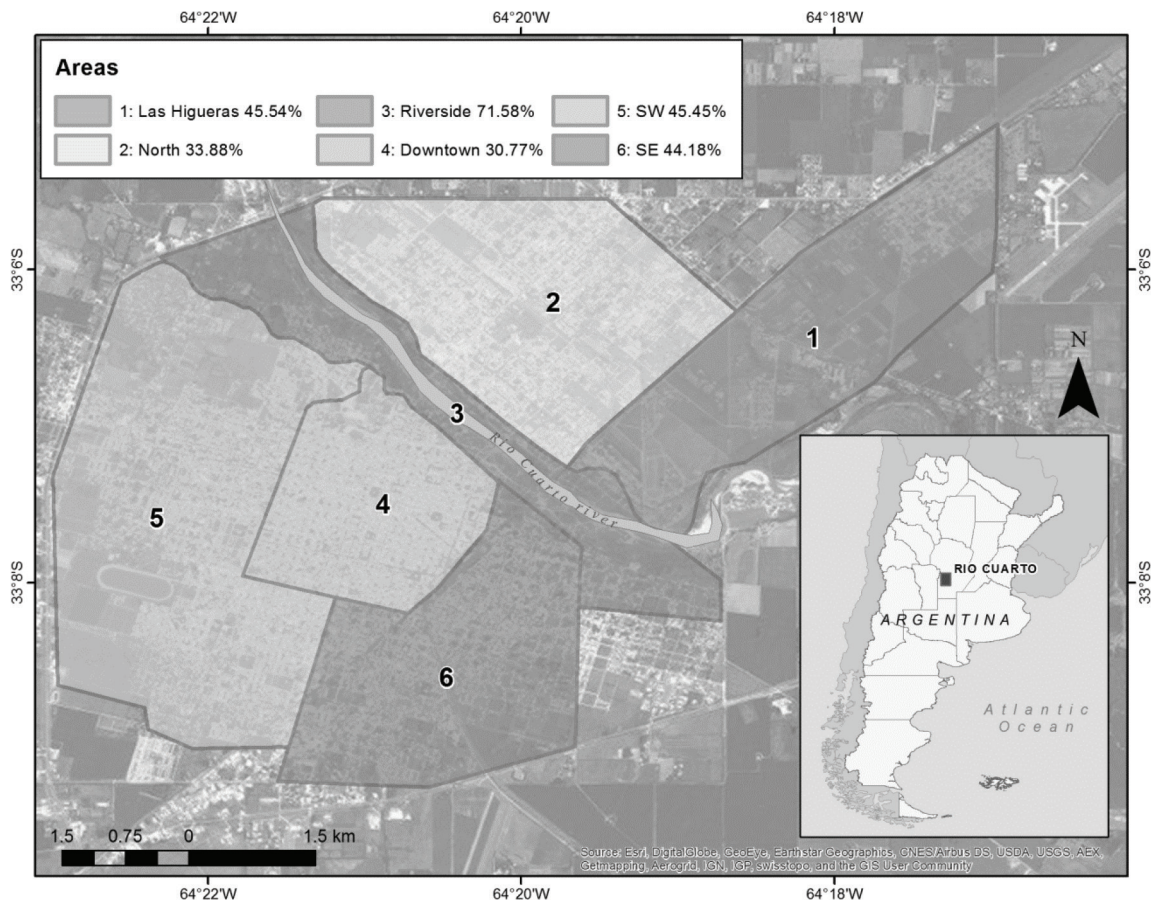

Figure. Overall canine parasite prevalence by sampling area, Río Cuarto, Córdoba, Argentina (n= 493).

2.4. Statistical analysis

Overall prevalence was defined as the percentage of fecal samples positive for any parasite species, and the specific prevalence as the percentage of fecal samples positive for a given parasite species. Multivariate logistic regression was used to quantify the association between parasites' presence with each variable after adjusting for the others. Only variables significant at P < 0.25 in the univariate analysis were considered eligible for inclusion in the multiple logistic regression analysis (16). Independent variables were the presence of at least one parasite in dogs' feces, defined as "all parasites", and GI parasites more frequently present were separate outcomes. Dummy variables were generated for any categorical variable with more than two levels. Backward elimination was used to determine the factors to be dropped from the multivariable model. The goodness of fit of the model was assessed with the Hosmer–Lemeshow statistic (16). All statistics calculations were performed with SAS Software (SAS System from Windows 9.0).

3. Results

A total of 493 fecal samples were collected between March 2010 and November 2013. Among these, 45.23% (223) (95% CI 40.83–49.62) included at least one parasite. The overall annual prevalence was similar throughout the years, except for the first year, when a higher value was observed (Table 1). Eight GI parasites were found in the study including *A. caninum* at 30.83% (152/493), *T. vulpis* at 9.94% (49/493), *Cystoisospora* spp. at 7.71% (38/493), *T. canis* at 6.90% (34/493), *Giardia* spp. at 5.88% (29/493), *Sarcocystis* spp. at 2.64% (13/492), *Eucoleus boehmi* at 1.42% (7/493), and *Dipylidium caninum* at 0.61% (3/493). *A. caninum* remained the most prevalent parasite during the study period. Regarding the area sampled, for all parasites, the highest prevalence was found in the riverside area of Rio Cuarto (71.58%, 68/95), followed by Las Higueras (45.54%, 51/112), southwestern Río Cuarto (45.45%, 20/44), southeastern Río Cuarto (44.18%, 19/43), northern Río Cuarto (33.88%, 41/121), and downtown Río Cuarto (30.77%, 24/78) (Figure). The distributions of variables gathered in this study are displayed in Table 1. Results from the final model of the logistic regression showed an increased risk with age for all parasites (for at least one parasite) and for *T. canis*, *Cystoisospora* spp., and *Giardia* spp. when analyzed as separate independent variables. No ingestion of small animals, daily feces removal, and no shadow in house yards were also associated with parasites' presence. At the same time, anthelmintic use and daily feces removal were protective factors for *A. caninum*. Female dogs, compared to males, were at a lower risk for *T. canis*. Absence of shadow in the yard was also a protective factor for this parasite. Regarding *Cystoisospora*, no ingestion of small animals was a protective factor, while for *T. vulpis* the only variable associated was no presence of shadow in the yard (Table 2).

4. Discussion

The overall prevalence of intestinal parasites in dogs detected in this study was high (45.23%) considering that all dogs lived in houses with owners. These values are similar to the ones reported in the literature (8,17). *A. caninum* (30.83%) was the most commonly found parasite in dogs' feces, agreeing with several studies (8,9,12,17,18). A marked decline for all parasites was observed in the years studied, with a prevalence value of 70.59% for 2010. In 2010 the majority of canines sampled were located in areas surrounding the river, where *A. canium* reached a prevalence of 50%. These areas consist of socioeconomically vulnerable human settlements, located along the river coast with poor infrastructure houses, low education level, no formal job, and lack of basic services such as potable water. At the same time, an important number of animals are present, mainly canines, but also small ruminants, swine, and equines which are used for sand extraction in the nearby river. Each family with horses had about five or six dogs; however, the variable area of the city sampled was not retained in the final model.

Results showed a low prevalence of *T. canis* compared to other studies (7-10,18-20). This could be explained with the fact that 53% of dogs sampled in the study were older than one year of age. Young dogs are at higher risk of infection for *T. canis* due to transplacental and lactogenic transmission (19).

With respect to *Giardia* and *D. caninum* infection, a lower prevalence was found compared with other studies (2,5,6,10,18,20–23). However, results should be interpreted with caution regarding the sensitivity of the diagnostic techniques used, intermittent shedding of *Giardia* cysts, observation of gravid proglottids for *D. caninum*, and differences in sample size.

Logistic regression models resulted in age being a risk factor associated with the presence of at least one parasite, while no animals' ingestion, daily feces removal, and absence of shadow in the yard were protective factors. Analyzing each factor in a separate manner, dogs younger than one year of age were at an increased risk for the presence of at least one parasite and also for *T. canis*, *Cystoisospora* spp., and *Giardia* spp. than animals of one year of age or more. This result agrees with previous studies (4,8,19,22,24-26). Immunity to coccidia and *Giardia* may be life-long in companion animals, but when exposed for the first time, older animals may be more likely to develop patent infections easily detectable on fecal examination (21).

Table 1. Population characteristics of parasites of dogs (n= 493), Greater Río Cuarto, Córdoba, Argentina.

	Positive, n (%)	Negative, n (%)	Total
Year 2010	84 (70.59)	35 (29.41)	119
2011	60 (40.82)	87 (59.18)	147
2012	34 (36.56)	59 (63.44)	93
2013	45 (33.58)	89 (66.42)	134
Sex Female	94 (39.33)	145 (60.67)	239
Male	129 (50.79)	125 (49.21)	254
Age < 1 year	42 (53.16)	37 (46.84)	79
≥ 1 year	181 (43.72)	233 (56.28)	414
Other animals in household Yes	171 (49.71)	173 (50.29)	344
No	52 (34.90)	97 (65.10)	149
Access outside household Yes	188 (47.35)	209 (52.65)	397
No	35 (36.46)	61 (63.54)	96
Access to public areas Yes	161 (48.49)	171 (51.51)	332
No	62 (38.51)	99 (61.49)	161
Owner takes dog for a walk Yes	70 (36.08)	124 (63.92)	194
No	153 (51.17)	146 (48.83)	299
Use of leash by owner Yes	58 (33.72)	114 (66.28)	172
No	165 (51.40)	156 (48.60)	321
Dog is a hunter Yes	128 (56.89)	97 (43.11)	225
No	95 (35.45)	173 (64.55)	268
Ingestion of raw meat Yes	42 (37.50)	70 (62.50)	112
No	181 (47.51)	200 (52.49)	381
Anthelmintics use Yes	88 (34.51)	167 (65.49)	255
No	135 (56.72)	103 (43.28)	238
Last treatment < 3 months Yes	51 (34.00)	99 (66.00)	150
No	172 (50.15)	171 (49.85)	343
Daily feces removal Yes	103 (35.03)	191 (64.97)	294
No	120 (60.30)	79 (39.70)	199
Cleaning of backyard ≤ 3 days Yes	71 (36.60)	123 (63.40)	194
No	152 (50.83)	147 (49.17)	299
Presence of feces Yes	194 (48.50)	206 (51.50)	400
No	29 (31.18)	64 (68.82)	93
Presence of grass or sand Yes	208 (47.16)	233 (52.84)	441
No	15 (28.85)	37 (71.15)	52
Presence of shadow Yes	163 (53.62)	141 (46.38)	304
No	60 (31.75)	129 (68.25)	189

Research on animals' ingestion evidenced hunting habits in the dogs, in accordance with the owners' knowledge. No ingestion of small rodents or birds was a significant protective factor for both the presence of at least one parasite element and *Cystoisospora* oocysts in feces. It has been described that stray dogs are more likely to be infected with *Cystoisospora* than dogs with owners because stray dogs must hunt for food and therefore have more exposure to infected paratenic hosts (27).

Regarding practices of removing feces in the household, daily feces removal was a protective factor for the presence of at least one parasite element and *A. caninum*. Accordingly, it has been reported that removing feces in the household reduces the contamination of the environment, thus decreasing the risk of infection for humans and other dogs (28). Another protective factor detected in this study was the absence of shadow in the yard. The effect was observed for the presence of all

Table 2. Final logistic regression model for all parasites and main parasites found in 493 dogs in Greater Río Cuarto, Córdoba, from March 2010 to November 2013.

	OR	95% CI	P
All parasites			
Age < 1 year vs ≥ 1 year	1.749	1.042; 2.936	0.0343
Animals ingestion No vs Yes	0.583	0.382; 0.890	0.0123
Daily feces removal Yes vs No	0.537	0.347; 0.829	0.0051
Shadow presence No vs Yes	0.565	0.374; 0.854	0.0068
Ancylostoma caninum			
Anthelmintics use Yes vs No	0.528	0.341; 0.816	0.0040
Daily feces removal Yes vs No	0.456	0.296; 0.702	0.0004
Toxocara canis			
Age < 1 year vs ≥ 1 year	5.381	2.461; 11.765	< 0.0001
Sex Female vs Male	0.435	0.197; 0.960	0.0393
Shadow presence No vs Yes	0.146	0.043; 0.493	0.0019
Cystoisospora spp.			
Age < 1 year vs ≥ 1 year	2.788	1.261; 6.164	0.0113
Animals ingestion No vs Yes	0.270	0.127; 0.576	0.0007
Trichuris vulpis			
Shadow presence No vs Yes	0.330	0.156; 0.697	0.0037
Giardia spp.			
Age < 1 year vs ≥ 1 year	3.559	1.610; 7.865	0.0017

parasites and *T. vulpis.* Shady environments are suitable to eggs' survival, and higher proportions of viable eggs were recovered in samples from shady areas than sunny ones (29).

Other protective factors were anthelmintic use for *A. caninum* and dogs' sex for *T. canis,* with bitches being at a lower risk for being positive to *T. canis* than males. In the current epidemiological investigation, the number of male dogs was almost double the number of females examined. This unequal distribution of sex in the studied population might be considered when comparing results. In addition, people usually neuter more females than males, so predation on paratenic hosts could be greater in males or these results could be masked by confounding. No data were gathered regarding the dogs neuter status. Hence, these results should be interpreted with caution.

Most of the owners (56.72%) reported not deworming regularly their dogs; however, the use of anthelmintics was a protective factor for Strongylidae eggs in feces. Lack of knowledge of dog owners about the zoonotic potential of intestinal parasites and methods of control seems to be the main reason for the apparent negligence in treating pets (18). However, no data were gathered regarding the drugs used for treatment, but generally broad spectrum drugs were used. The results could also be misleading due to response bias.

Among the factors associated in this study, daily removal of dogs' feces and treatment are easily modifiable, supporting the conclusion that treatment and control measures against infection with GI parasites must be applied. Also, the high level of parasite infection described highlights the need of monitoring parasites by routine fecal examination, strategic deworming programs, and hygiene measures.

In conclusion, the present study showed that age was associated with an increased risk of GI parasites in dogs. No ingestion of small animals, daily feces removal, and absence of shadow in the house yards were protective factors. The limitation of one stool sample being collected and examined may cause underestimation of the prevalence of zoonotic parasites in pets. Given the clinical importance of intestinal parasites affecting pets, their ubiquitous presence, and the zoonotic impact, public education is crucial for reducing risk exposure in both humans and companion animals.

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