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Research Article

Prevalence of Intestinal Parasitoses in Dogs within a Vulnerable Area of Ensenada, Buenos Aires Province, Argentina

María Inés Gamboa[1,2*]; Marcos J Butti[1]; Valeria V Corbalán[1]; Beatriz A Osen[1]; Antonela Paladini[1]; Estela B Bonzo[3]; Fiamma Lagala[1]; Nilda E Radman[1]

[1]Cátedra de Parasitología Comparada-Laboratorio de Parasitosis Humanas y Zoonosis Parasitarias, Facultad de Ciencias Veterinarias, Universidad Nacional de La Plata, Argentina

[2]Comisión de Investigaciones Científicas de la Provincia de Buenos Aires (CIC), Argentina

[3]Cátedra de Higiene, Epidemiologia y Salud Pública, Facultad de Ciencias Veterinarias, Universidad Nacional de La Plata, Argentina

***Corresponding author: María Inés Gamboa**
Cátedra de Parasitología Comparada-Laboratorio de Parasitosis Humanas y Zoonosis Parasitarias, Facultad de Ciencias Veterinarias, Universidad Nacional de La Plata, Calle 60 y 118 (1900) La Plata, Buenos Aires, Argentina
Tel: +54-221-4236663 int 413
Email: minesgamboa@fcv.unlp.edu.ar

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Abstract

The potential role of dogs as reservoirs of zoonotic infections is one of the major public health problems. Water emergency areas are generally vulnerable zones with a lack of care from the owners who do not have the basic sanitary service conditions. Dogs often feed on waste detecting coprophagia of human faeces facilitated by the final disposal of excreta in open-air defecation. The objective of this research was to determine the presence of intestinal parasitoses in dogs from a sanitary risk area inhabited by a vulnerable human population. Dog faeces were collected by an enema with a soapy solution and processed by the Telemann's sedimentation technique as well as the Sheather's flotation procedure besides a direct examination. A number of 703 (79.3%) analyzed dog faeces were parasitized from a total of 886. *Ancylostoma caninum* (57%), *Toxocara canis* (24%) and *Uncinaria stenocephala* (21%) were the most frequent species. The specific richness in the dog population was 17 species. The highest parasitosis frequency was observed among male dogs and those under one-year aged for the total parasitized ones with *T. canis*, *Cystoisospora canis*, *C. ohioensis*, and *Giardia* spp. High prevalences found in dogs from the present study could indicate that both diagnosis and treatment are not enough to achieve sustainable changes in vulnerable areas. Actions addressed to the environmental factor are essentials in order to avoid reinfections.

Keywords: Dogs; Intestinal parasitoses; Prevalence; Vulnerable population; Zoonoses

Impacts

- It was used 3 different techniques of detection of enteroparasitosis.
- It was detected high prevalence of canine enteroparasitosis (79.3%). *Ancylostoma caninum*, *Toxocara canis* and *Uncinaria stenocephala* were the most frequent species.

It was detected several zoonotic species. The highest frequency of parasitosis has been observed among male and under 1 year of age.

Introduction

Intestinal parasitoses show a high prevalence in dogs despite the empirical use of antiparasitic drugs and several control measures recommended by veterinarians. The potential role of dogs as reservoirs of zoonotic infections is one of the major public health problems. Water emergency areas are generally those where the population has unsatisfied basic needs [44], a vulnerability accompanied by low purchasing power. These are usually suburban areas with several animals per family. Dogs are careless, wanderers, underfed or malnourished, with the potential of consuming rats, waste, and sometimes human or other animal faeces as a product of the prevailing structural shortage [57]. Vulnerable zones are made up of informal dense urbanizations with precarious housing without access to suitable sanitary facilities occupied by populations of a low socioeconomic

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and educational level [1]. The lack of pet care by the owners is common in these populations. Dogs usually feed on waste and often perform coprophagy of human faeces facilitated by the final disposal of excrements at open-air defecation. Coexistence with domestic animals favors the presence and maintenance of infections, reinfections, and coinfections in these areas [22]. The problem is worsening by the movement of cysts, oocysts, eggs, and larvae until they are spread by rainwater or flooding. This is in addition to the presence of ditches where animals usually drink, feed on fish, toads, and frogs, paratenic hosts of parasitosis, and where excrements and wastewater from dwellings are disposed of [54]. The water quality for domestic use is generally inadequate with clandestine connections and the river water use for drinking, food production, and recreational activities hold a significant impact on human and animal health [38]. Canines can disseminate transmissible intestinal parasitoses to humans with their faeces [14]. Some helminthiases [27,65], several protozoonoses [68,72], and the parasitic algae *Blastocystis* spp. [48], are common findings on dogs and humans. Animal intestinal parasite nematodes of the genus *Toxocara* spp. Cause toxocarosis in humans, a disease of high seroprevalence in La Plata city [52], other regions of America [31], and the rest of the world [12,28,61].

Its neurological and ocular parasitic forms usually have serious consequences. Its presence in humans is favorably influenced by the residence place in suburban area [5,13,53]. The zoonotic enteroparasite *Giardia* spp. [39] causes acute or chronic diarrhea, and changes in the microbiome of canines, an effect still poorly studied [7]. Antiparasitic drugs reduce environmental contamination by parasitic dissemination forms and they are effective in their control. However, it is difficult to use all available information to evaluate the true drug efficacy such as the understanding of either possible geographic variations or drug resistance [40], among other variables. Sentinel animals can be used for surveillance of pathogen circulation. In this sense, dogs can act as bioindicators providing early evidence of emerging zoonotic diseases circulating in a certain area or region [8,24,63].

In Argentina, there is not enough information on intestinal parasitoses prevalence in dogs since most surveys were based on fecal samples collected from the ground [15,34,59,64].

The district of Ensenada, Buenos Aires Province, is composed of 14,660 households of which 10.3% have their basic needs unsatisfied [44]. The El Molino neighborhood is located right there, inhabited by a vulnerable population on the coast of the Río La Plata, a water emergency area. Its current layout is the result of an uninterrupted occupation process being the area a settlement area for people coming from other Argentinian provinces and neighboring countries without resources. This, together with the lack of infrastructure works and increasing deforestation increases its vulnerability.

Studies on canine intestinal parasitoses from this site report 92% [18], and 76.7% [54]. Special characteristics determined that this was selected as a sentinel area. Sentinel Sites located in areas of vulnerability are considered remote sensors that send alarm signals of selected variables. The variability effect on the stability of the population's livelihoods is reflected in higher levels of chronic and acute malnutrition, and other diseases, especially the transmissible ones. Sentinel surveillance allows the decision-maker to establish previsions and respond to emergencies in due time.

The objective of this work was to determine the presence of canine intestinal parasitoses in a health-risk area inhabited by a vulnerable population.

Materials and Methods

Study Area

The El Molino neighborhood (34° 55’ S, 57° 56’ W) within the Marginal Forest of Punta Lara, the southernmost gallery forest in the world, has specific hydrographic characteristics that contribute to the spread of parasites [9]. Floodings are not caused by rain but rather by southeast winds from the La Plata River which overcomes the estuary containment for its flat shores. These geomorphological features combined with a lack of river containment, channeling infrastructure, and an increase in the local resident population, raise the vulnerability of the area and favor the development of parasites. In addition, a clay-enhanced soil avoids the rainwater absorption also overlapping some cyclical flooding (Figure 1).

The dominant weather belongs to a plain wet-temperate type with mid-temperature conditions and mid-high rainfalls distributed regularly throughout the year. The relative moisture is high and the water balance shows a remarkable predominance of water surpluses over water deficits.

Regular rainy periods extend from October to April. Rainfalls slightly exceed 1000 mm per year. The lowest rainfalls are recorded during winter although there is a non-defined dry season. The mean annual temperature is 16ºC with mild winters and hot summers. Extreme temperatures are between 42ºC and -4ºC being January the warmest month with a mean of 22.5ºC and July the coldest with a mean of 9.7ºC.

Sampling

Sampling occurred within a framework of monthly educational healthcare workshops that took place from 2015 to 2019 at the “El Molino” neighborhood. Animals were spontaneously taken by their owners to be vaccinated, and they all provided several samples for diagnosis.

Socio-Environmental Data

Epidemiological data related to dogs were collected by specially designed forms in which all information concerning owner and animal backgrounds was recorded such as canine age, eating habits, and mobility within the neighborhood.

The delivery material included a consent-informed form allowing permission for interventions of clinical examination and sampling, vaccination (Rabies vaccine), and treatment, if relevant.

Dog Samples

Animals brought voluntarily by their owners were vaccinated and examined by echography at the same time as they provided samples for diagnosis. Dog faeces were collected by a soapy solution enema and processed by both the Telemann´s sedimentation technique and Sheather’s flotation besides a direct examination. Dissemination elements (eggs, larvae, cysts, and oocysts) were identified according to their morphology [60]. This study received approval from the Ethical Committee of the School of Veterinary Medicine at the National University of La Plata.

Results

From the 886 analyzed canine faeces, 703 (79.3%) were parasitized being *Ancylostoma caninum*, *Toxocara canis* and *Uncinaria stenocephala* the most frequent species (Table 1). The specific richness of the dog population was 17 species. A higher parasitosis frequency was observed in male canines (82.8%) than in females (75.4%), with significant differences (X^2 correct Yates= 6.9 p<0.01). The statistical association between both sex and parasite prevalence was only maintained in *A. caninum* (X^2 correct Yates=19, p<0.01), the most frequent species, analyzing this variable by each species. There was an association with dog ages being those under one-year aged -up to 1 year old- the most parasitized compared to those over one-year aged (X^2= 7.9 p<0.01). Significant differences were found with higher prevalence among the under one-year aged canines than the older ones for *T. canis* (X^2=50.1, p<0.01), OR: 3.524 (95% CI 2.4-5.0) pointing out they are 3.5 times more likely to host *T. canis* than those older ones analyzing the age distribution for each species. The same occurred with *Giardia* spp. (X^2=5.9, p=0.01), *Cystoisospora ohioensis* (X^2=7.2, p<0.01), and *C. canis* (X^2=11.5, p<0.01), OR: 4.4 (95%CI 1.5-12) indicating that under one-year aged dogs are 4.4 times more likely to host *C. canis* than older ones (Table 1). However, the association was inverse for *T. vulpis* (X^2=15.7, p<0.01), OR: 0.476 (95%CI 0.333-0.681), that is to say, juveniles are 2.1 less likely to have *T. vulpis* than older ones. In the case of *Capillaria* spp. (X^2=10.8, p<0.01), OR: 0.278 (0.129-0.602), under or equal one-year aged canines have 3.6 fewer chances to host *Capillaria* spp. than older ones.

There were no significant differences for intestinal parasitoses between pedigree and mongrel dogs (p>0.05). Positive cases of 44.2% were monoparasitized, 34.9% were infected by 2 species, 15.2% by 3 species, 4.7% by 4 species, and 1% by 5 species. That implies a 20.9% of polyparasitized dogs with a maximum of 5 species in co-infection. The monoparasitized (50.1%) harbored *A. caninum*, 17% *T. canis*, and 10.2% *U. stenocephala*.

Table 1: Prevalence of intestinal parasitoses over 886 canines from the El Molino neighborhood, Ensenada.

Species	Total		Under one-year aged		Over one-year aged	
	N°	%	Nº	%	Nº	%
Ancylostoma caninum	497	57	244	54	253	58
Toxocara canis	210	24	152	33.6	58	13.4
Uncinaria stenocephala	184	21	88	19.4	96	22.2
Trichuris vulpis	160	18	60	13	100	23
Giardia spp.	69	8	47	10.4	22	5
Cystoisospora canis	65	8	47	10.4	18	4.2
Capillaria sp.	36	4	9	2	27	6.2
Cystoisospora ohioensis	28	3	20	4.4	8	1.8
Ascaris lumbricoides	10	1	2	0.4	8	1.1
Dipylidium caninum	7	1	3	0.7	4	0.9
Blastocystis spp.	7	1	4	0.9	3	0.7
Pentatrichomonas hominis	4	0.5	3	0.7	1	0.2
Sarcocystis spp.	3	0.1	3	0.7	0	0
Taenia sp.	2	0.1	0	0	2	0.5
Trichuris trichiura	1	0.1	0	0	1	0.2
Toxascaris leonina	1	0.1	0	0	1	0.2
Spirometra sp.	1	0.1	0	0	1	0.2

Cases of biparasitism were associated with *A. caninum-T. canis* (25.3%), followed by *A. caninum-U. stenocephala* (22.8%), and *A. caninum-T. vulpis* (20.4%). The most frequent combination was *A. caninum-U. stenocephala-T. vulpis* (15.6%), and *A. caninum-U. stenocephala-T. canis* (9.5%) in cases of polyparasitism. A statistical association was observed between the most frequent species *A. caninum* with *T. canis* (p=0.02), then *T. vulpis* (p<0.01), and *U. stenocephala* (p<0.01).

Discussion

The prevalence of intestinal parasitoses observed in the canine population (79.3%) implies a high infection risk for the human population due to the finding of dissemination forms of zoonotic parasites in their faeces. Other authors have recorded divergent values on intestinal parasitoses prevalence in canines of Argentina (52.4% [20] 82.1% [10] and other countries (87% [35] 57.4% [73] 38.3% [33] 40.1% [51] 63.5% [56].

In Argentina, many authors have based their studies on parasite detection in canine faeces collected from the ground [15,34,59,64]. This prevents the parasite prevalence calculation per host for which their results are not comparable with those of the present work performed with samples obtained from each animal beyond of giving valuable information on parasites circulating in the environment.

If we had worked with spontaneously excreted faeces, probably a higher number of parasites would have been found even in a single sample. Trophozoites in faeces could be destroyed by the action of a soap solution. Furthermore, a spontaneous elimination of either *Taenia* spp. or *Dipylidium caninum* proglotids can be lose by this technique [60]. Likewise, processing serial samples might increase positive diagnoses as indicated by Espinosa et al (1988).

Even so, fecal samples were observed fresh and processed by both flotation and sedimentation techniques which allowed to increase the recovery efficiency of parasitic forms [43]. A high specific richness (17 species) was detected which exceeded the reported values in the literature [10,20,33,35,51].

The most prevalent species were *Ancylostoma caninum*, *Toxocara canis*, *Uncinaria stenocephala*, and *Trichuris vulpis*, coinciding with that reported by other authors from Latin America [10,20,35], unlike works in Asia where cestodes predominate [51], and Europe with a high prevalence of *Toxascaris leonina* [56].

Intestinal parasitoses were more frequent among males and under one-year aged canines [20,73], with no breed differences in agreement with other studies. When analyzing the frequency by both parasitic species and age range, *T. canis*, *Cystoisospora canis*, and *Giardia* spp. were more frequent in puppies [33]. This could be due to the parasite-specific immunity is acquired by age probably as a consequence of successive exposures to parasites hence younger animals are more sensitive to parasitism [23,60]. Regidor-Cerrillo et al. (2020) found no association between parasitosis and canine age in Spain.

Many diagnosed parasitosis are zoonotic. Regarding, it is important to highlight that coincidentally with other research [74], the most prevalent species among analyzed canines in this study was *Ancylostoma caninum* capable of invading humans as both cutaneous larva migrans, and an emerging zoonotic intestinal parasitoses [21,29,50].

Ancylostoma caninum and *Uncinaria* sp. own morphological and biological similarities. However, the prevalence was higher for *Ancylostoma caninum* than for *Uncinaria* sp. in this study. Analyzing the reasons for these occurs, it is likely they have influenced the mechanism of arrested larvae more frequent in *Ancylostoma* [32], its vertical transmammary transmission by colostrum and milk [32], and the site weather conditions since *Uncinaria* is optimally adapted to lower temperatures than *Ancylostoma caninum* [3,6,67.

Arrested larvae of *Toxocara spp* in pregnant canine females are mobilized causing infections by transplacental and then transmammary pathways. Others become adults in the maternal intestine causing patent infection with high dissemination of resistant eggs to adverse environmental conditions [5,19].

Females also become over-infected by ingesting immature worms present in faeces of their puppies. In this study, a higher prevalence of *Toxocara* canis was observed in under one-year aged animals in agreement with research conducted in Argentina and a global meta-analysis that included samples from more than 13 million animals [20,54,61]. Prevalence was 24% in males higher than in females (22%), but no significant differences.

A higher frequency of patent toxocarosis in males was previously reported by other authors [46,61]. Probably, it is due to a biological compensation since male canines can only disseminate by a patent intestinal infection [46] (Schnieder et al., 2011). Several authors postulate a possible immunosuppressive effect of testosterone that would enable this elimination pathway (Curi et al., 2017; Abdel Aziz et al., 2019).

The prevalence of this species was lower in under one-year aged animals since the acquired immunity associated with age probably decreases the *Toxocara* infection intensity and settlement (Greve, 1971; Abdel Aziz et al., 2019). Older male canines are removers of *T. canis* eggs in unusual instances and this occurs after a larvae incorporation by both ingestion of paratenic hosts and immunosuppression.

It is remarkable that a high seroprevalence of toxocarosis was observed in local children (32.3%), as mentioned by Archelli et al. (2019) in agreement with the high prevalence of *Toxocara canis* in canines. Human toxocarosis is currently more linked to both a geophagy behaviour and a lack of personal hygiene than either environmental contamination with eggs or contact with infected dogs [5,47,71].

There are reports of an association between seropositivity in children who played near their homes in areas with infected dogs [36]. Also, it could be inferred that people who perform working activities in contact with the ground have a higher risk of infection by this parasite as proven for *T. cati* [55]. Unlike Rostami et al. (2020), who reported higher prevalences of *T. canis* in deprived areas, this study was also carried out in a deprived area and a prevalence of 24% was found which is lower than that reported in an urban area by Radman et al. (2006). Probably, it is due to those authors performed a directed sampling.

On the other hand, Rostami et al. (2020), found lower frequencies (10% and 8%) in sampling sites located at the same latitude as the studying research area located at 34°49'S with 24% of canines infected by *T. canis*. Both the long-term prepatent period of *T. vulpis* [17], and the vertical transmission absence make this parasite more frequent in adult dogs as observed in this study and other areas [20,42.

Its role as a zoonotic infection agent is still debated [69], however, several cases of human intestinal parasitoses caused by this species have been reported [16,30,37,41,49]. Further studies are likely needed to clarify this but in a sympatric area such as the studied one where there could be a transmission between humans and canines [41].

However, both canine capillariids *C. aerophyla* and *C. boehmi* located in the respiratory tract are not vertically transmitted, their prepatent periods are extended, and their disposal is discontinuous [11]. This is consistent with a higher frequency in adult animals.

Although wandering canines often ingest either bird or rodent viscera parasitized by *Capillaria* spp., their eggs are eliminated with unaltered canine faeces which give rise to false-positive diagnoses. The absence of *Strongyloides stercoralis* in processed samples could be due to the circulation lack of this species in the area although it could also represent a false negative result since the appropriate diagnostic technique for the larvae recovery was not used.

It would be important to expand surveillance with the use of an appropriate methodology since the epidemiological site analysis indicates that all conditions required for the settlement of an autochthonous outbreak are present. The presence of residents from endemic areas, improper disposal of excreta, and both coprophilic and wandering canine habits are factors that are facilitated in addition to climate change.

Applying larval recovery techniques would be also very useful for the diagnosis of both canine and feline pulmonary verminosis such as *Angyostrongylus vasorum*, *Aleurostrongylus abstrusus*, and other Metastrongylidae. The presence of *A. lumbricoides* and *T. trichiura* eggs indicates both the fecalism of human faeces on behalf of canines and environment fecal contamination, a behavior already observed in surveys on nearby areas [10,22].

In Greece, Kostopoulou et al. (2017), found *Giardia* spp. as the most prevalent parasite in dogs (25.2%), and cats (20.5%), by using an immunological test. However, results suggested a limited zoonotic risk when performing genotyping. Probably, the frequency of *Giardia* spp. in tested dogs is higher than that detected by the methodology used in the present work (7.8%).

The high prevalence of this protozoonoses in local children (20.9%) deserves that genotyping studies are carried out to corroborate whether they correspond to zoonotic genotypes.

The finding of Taenidae family eggs suggests feeding on raw meat, viscera, or animal carcasses. Hydatidosis is endemic in Argentina with a high prevalence in livestock (12.7%).

Even though there are not enough studies on the *Echinoccocus granulosus* prevalence in dogs, Taenidae family eggs have been detected in different areas of the country in canine fecal matter extracted from the ground [15,58,62,64,66]. Our results, the high prevalence of echinococcoses in livestock, and the presence of one hydatidosis case in a resident [4], allow us to suppose that the disease could be present in the area and canines would act as reservoirs.

Canines as bioindicators allowed to evidence the presence of *Spirometra* sp., a human sparganosis agent, caused by the plerocercoid of these cestodes widely distributed in South America [45]. The risk of acquiring sparganosis increases when frogs are included in the diet, and their commercialization contributes to its spread. In this study, its prevalence was 0.1%, lower than 6% previously reported in the area [18].

The finding of various zoonotic parasitosis in canines provides data on the circulation of different parasitic genera. Affected animals act as an infection source for others and humans. Intestinal parasitoses must be addressed by One Health like other transmissible diseases.

Unsatisfied basic needs added to working and educational precariousness condition pollution and dissemination of parasitic forms in suburban areas. The area under study had the opportunity to receive a different assistance degree, diagnosis, and supply of antiparasitic treatments.

However, obtained results indicate that all efforts made were insufficient for a sustainable reduction of canine intestinal parasitoses. Probably, the flooding area facilitates the dispersion of parasitic resistance forms. The soil filtration mechanism eventually concentrates them on the surface which combined with resistance strategies of each parasite and its high biotic potential facilitates that more infective forms are available to their hosts. If the environmental situation continues and there is no further monitoring of the infection sources, it is expected that parasitosis will remain at current levels or will increase.

As the ground loses its absorption capacity, the polluted area after each flood may increase.

High prevalences found in the present study in canines could indicate that both the diagnosis and treatment are not enough to achieve sustainable modifications in certain areas. Actions directed to the environmental factors are essential in order to avoid reinfections.

Author Statements

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Ethical Approval

This study received the approval of the Ethical Committee of the School of Veterinary Medicine of the National University of La Plata and has no conflicts of interest.

Competing Interests

The authors declare no confict of interest.

Authors Contributions

María I. Gamboa: Socioenvironmental-data and fecal collection, laboratory analysis and preparation of manuscript.

Beatriz A. Osen: Collection of fecal samples, socioenvironmental-data and laboratory analysis.

Nilda E. Radman: Parasitological and socioenvironmental-data collection, laboratory analysis and preparation of manuscript.

Marcos J. Butti: Collection of fecal samples and laboratory analysis.

Valeria V Corbalán: Collection of fecal samples, socioenvironmental-data collection and laboratory analysis.

Antonela Paladini: Collection of fecal samples, socioenvironmental-data collection and laboratory analysis.

Estela Bonzo: Statistical analysis.

Fiamma H Lagala: Collection of fecal samples, socioenvironmental-data collection and laboratory analysis.

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