Revista Brasileira de Parasitologia Veterinária
Brazilian Journal of Veterinary Parasitology
ISSN 1984-2961 (Electronic)
www.cbpv.org.br/rbpv

Short Communication

Helminth eggs with zoonotic potential in the vicinity of public schools in southern Brazil

Ovos de helmintos com potencial zoonótico nas proximidades de escolas públicas no extremo sul do Brasil

Catia Cilene Santos de Mello[1], Leandro Quintana Nizoli[2], Alexsander Ferraz[2], Bruno Cabral Chagas[1], William James Domingues Azario[3], Marcos Marreiro Villela[1]*

[1] Departamento de Microbiologia e Parasitologia, Instituto de Biologia, Universidade Federal de Pelotas – UFPel, Pelotas, RS, Brasil
[2] Departamento de Veterinária Preventiva, Faculdade de Veterinária, Universidade Federal de Pelotas – UFPel, Pelotas, RS, Brasil
[3] Secretaria Municipal de Educação e Desporto, Prefeitura Municipal de Pelotas, Pelotas, RS, Brasil

How to cite: Mello CCS, Nizoli LQ, Ferraz A, Chagas BC, Azario WJD, Villela MM. Helminth eggs with zoonotic potential in the vicinity of public schools in southern Brazil. *Braz J Vet Parasitol* 2020; 29(1): e016419. https://doi.org/10.1590/S1984-29612019102

Abstract

The aim of this study was to evaluate environmental contamination by helminth eggs with zoonotic potential that were found in dog feces in the vicinity of elementary schools. Seventy-nine samples of dog feces were collected from 28 municipal schools located in five neighborhoods in Pelotas, Rio Grande do Sul (RS), Brazil. The samples were processed using the Willis-Mollay technique and analyzed using an optical microscope (40X), to identify any parasite eggs present. All neighborhoods were positive and 74.7% of the samples exhibited one or more helminth genera. The agent with the highest prevalence was *Ancylostoma* spp. (93.2%), followed by *Trichuris* spp. (18.6%), *Toxocara* spp. (11.9%) and *Toxascaris* (1.7%). These data show that there is a need for greater care towards controlling these helminths with zoonotic potential, including responsible pet ownership and daily activities to clean and collect dog feces in the vicinity of schools, because these are places where children play and study.

Keywords: Environmental contamination, larva migrans, hookworms, *Toxocara* spp.

Resumo

O objetivo deste estudo foi avaliar a contaminação ambiental por ovos de helmintos com potencial zoonótico, diagnosticados em fezes caninas depositadas nas proximidades de escolas primárias. Setenta e nove amostras de fezes foram colhidas em 28 escolas municipais localizadas em cinco bairros da cidade de Pelotas, Rio Grande do Sul (RS), Brasil. As amostras foram processadas pela técnica de Willis-Mollay e analisadas em microscópio óptico (40X), para identificar quaisquer parasitos presentes. Todos os bairros foram positivos e 74,7% das amostras apresentaram um ou mais gêneros de helmintos. O agente com maior prevalência foi *Ancylostoma* spp. (93,2%), seguido por *Trichuris* spp. (18,6%), *Toxocara* spp. (11,9%) e *Toxascaris* (1,7%). Esses dados mostram que há necessidade de maior cuidado no controle desses helmintos com potencial zoonótico, incluindo a posse responsável dos animais e atividades diárias de limpeza e colheita de fezes de cães nas proximidades das escolas, porque esses são locais onde as crianças brincam e estudam.

Palavras-chave: Contaminação ambiental, larva migrans, ancilostomídeos, *Toxocara* spp.

Received September 10, 2019. Accepted November 19, 2019.
*Corresponding author: Marcos Marreiro Villela. E-mail: marcosmvillela@bol.com.br

Close relationships between humans and domestic animals such as dogs and cats do not just become established in households. These animals also frequent public spaces that are used for leisure activities, especially by children, who thus are exposed to the risk of infection caused by some helminth species (Carvalho & Rocha, 2011).

Dogs and cats are the domestic animals that maintain closest the contact with humans. Consequently, sanitary control regarding these animals is essential for human health. Among the diseases that affect dogs and cats, gastrointestinal helminthiases require attention within veterinary practice and public health promotion (Acha & Szyfres, 2003; Hotez et al., 2008; Peruca et al., 2009). These parasitoses may lead to anemia, diarrhea, weight loss, enteritis and even dogs' and cats' death (Robertson & Thompson, 2002). Humans may also contract them when they have direct contact with these animals, drink/eat water/food contaminated by parasite eggs or are affected by active larval penetration through the skin. Cutaneous larva migrans (CLM) and visceral larva migrans (VLM) are among the infections that are transmitted to humans by dogs and cats (Santarém et al., 2004; Mendonça et al., 2013; Moura et al., 2013).

Since these parasitoses can have an impact on both human and animal health, it is fundamental to monitor areas where there are many domestic, semi-domestic and stray dogs, so as to evaluate the risks that they pose to the community. Therefore, the aim of this study was to evaluate environmental contamination by zoonotic agents in dog feces collected in the vicinity of elementary schools.

Samples of dog feces were collected within a radius of 50 meters from the entrance gate of every municipal elementary school located in five neighborhoods in Pelotas, Rio Grande do Sul (RS), Brazil. They were stored in plastic bags and kept under refrigeration at 4°C until the time of processing at the Laboratory of Parasitic Diseases (LADOPAR), which belongs to the School of Veterinary Sciences at the Federal University of Pelotas (UFPel). Parasite eggs were evaluated using the modified Willis technique (Willis, 1921) and their morphological characteristics were identified using an optical microscope (40X objective lens).

The results were expressed as descriptive statistics and were then tabulated and analyzed using the Microsoft Excel® software. The chi-square test (χ^2 test) was used to ascertain whether any statistically significant differences were present among the variables (feces, neighborhoods and distances between schools). P-values ≤ 0.05 were considered significant.

Twenty-eight (84.8%) out of the 33 schools that were investigated between March and September 2017 presented dog feces in their vicinity. Most of these schools (23) were located on the outskirts of the city, while five were downtown. Data provided by the Municipal Department of Education and Sports (SMED) showed that the schools under investigation represented 80.5% of the schools located in the urban area and had 20,812 students enrolled (Table 1).

Table 1. Number of schools and their students in neighborhoods under investigation in Pelotas, RS, Brazil.

Neighborhood	% of schools visited	% of students enrolled
Areal	77.8% (7/9)	84.3% (5012/5945)
Centro	85.7% (6/7)	96.7% (5426/5613)
Fragata	100% (7/7)	100% (4238/4238)
Laranjal	100% (3/3)	100% (1913/1913)
Três Vendas	66.7% (10/15)	52.4% (4223/8053)
Total	80.5% (33/41)	80.8% (20812/25762)

Seventy-nine samples of feces were collected in the vicinity of the schools investigated; 74.7% of these samples exhibited one or more helminth genera. The lowest

prevalence of contamination (60.0%) was found downtown, while the highest (80.0%) was found in a suburb called Areal. However, there were no significant differences among the neighborhoods regarding the frequency of findings of feces with parasites in the vicinity of the schools under investigation ($p \geq 0.05$) (Table 2).

Table 2. Number of samples of helminth-positive feces in the vicinity of schools and their neighborhoods in Pelotas, RS, Brazil.

Neighborhood	No. of schools	No. of stool samples collected	No. of helminth-positive stool samples	% of positive samples
Areal	7	20	16	80.0
Centro	5	10	6	60.0
Fragata	5	10	7	70.0
Laranjal	2	9	7	77.8
Três Vendas	9	30	23	76.7
Total	28	79	59	74.7

Lower prevalences of contamination were reported by La Torre et al. (2018) in samples of dog feces in Rome and Padua, Italy (9.7%), and by Medina-Pinto et al. (2018) in samples of dog feces collected from public parks in Yucatán, Mexico, where the positivity rate was only 11%. In RS, Lopes et al. (2014) found positivity similar to what is reported in the present study, i.e. contamination by parasite eggs in 71.6% of feces samples collected from public squares in municipalities located in the south of RS. In the same state, Scaini et al. (2003) observed 86.1% positivity in feces samples collected from streets in Cassino, a neighborhood in the municipality of Rio Grande, RS, while Villela et al. (2009) found 54.3% positivity in feces deposited on beaches located in Laranjal, in Pelotas, RS. The latter corroborate the data collected in the present study, since Laranjal was also evaluated in this study and exhibited contamination in 77.8% of the feces samples (Table 2).

Table 3. Occurrence of monoparasitism and polyparasitism caused by helminths in samples of dog feces collected in the vicinity of schools in Pelotas, RS, Brazil.

Parasite(s)	Absolute frequency (%)	Relative frequency (%)
Ancylostoma spp.	41	69.5
Trichuris spp.	3	5.1
Toxocara spp.	1	1.7
Ancylostoma spp. + *Trichuris* spp.	8	13.6
Ancylostoma spp. + *Toxocara* spp.	5	8.5
Ancylostoma spp. + *Trichuris* spp. + *Toxascaris* spp.	1	1.7

In 76.3% (45/59) of the samples, one parasite genus was detected, while in 23.7% (14/59) of them, polyparasitism was observed (Table 3).

Regarding the method used in the present investigation, the Willis technique was chosen because it presents good results in environmental contamination studies, showing significant difference when compared to the Faust, Sedimentation and direct examination technique (Táparo et al., 2006). Moreover, such technique has lower material costs, shorter execution time and only requires the use of supersaturated solution (Novaes & Martins, 2015). However, it should be noted that other authors have detected different results. According to Santana et al. (2015), the centrifugal flotation technique showed higher sensitivity towards detecting *Ancylostoma* spp. and *Toxocara* spp.

in dog feces. Therefore, the use of only one technique may not reveal the total parasitism present in the samples, wich can be considered one of the limitations of this research.

Ancylostoma spp. was the most prevalent parasite, since it was found in 93.2% (55/59) of the positive samples ($p < 0.0001$); followed by *Trichuris* spp., in 18.6% (11/59); *Toxocara* spp., in 11.9% (7/59); and *Toxascaris* spp., in 1.7% (1/59). Similar distribution was reported by Scaini et al. (2003) in Cassino, a neighborhood and beach in Rio Grande, RS, where the percentage prevalences were 71.3%, 32.5% and 9.3%, and by Villela et al. (2009) on the seashore in Laranjal, a neighborhood in Pelotas, RS, where they found 90%, 14.7% and 6.3%, in the cases of *Ancylostoma, Trichuris* and *Toxocara*, respectively. Other studies carried out in different Brazilian regions also found that *Ancylostoma* spp. was the helminth with the highest prevalence (Blazius et al., 2005; Labruna et al., 2006; Katagiri & Oliveira-Sequeira, 2008; Oliveira et al., 2009; Prates et al., 2009; Klimpel et al., 2010; Ferreira et al., 2013, 2016). Thus, precautions need to be taken since cutaneos larva migrans may be acquired in the vicinity of schools, given that the larvae of both *Ancylostoma braziliense* and *Ancylostoma caninum* have the capacity to penetrate through the skin (Bowman et al., 2010). Moreover, this is currently considered to be a neglected parasitic skin disease, and cutaneous larva migrans is one of the most common skin diseases reported in travelers returning from tropical regions (Leung et al., 2017; Reichert et al., 2018).

The possibility of *Toxocara* transmission, together with favorable environmental conditions, is a serious risk to public health (Capuano & Rocha, 2006; Santos et al. 2017). The nematode *Toxocara* spp. is usually identified as the causative agent of visceral larva migrans and ocular larva migrans (Sviben et al., 2009), and it still may result in central nervous system alterations (Hotez, 2014). A study carried out by Schoenardie et al. (2013) in the same region found that the seroprevalence of *T. canis* in children in Pelotas, RS, was 50.6%. These authors correlated their finding with the high rate of occurrence of stray dogs in this city (Schoenardie et al., 2013). Although no investigation of seroprevalence has been conducted among schoolchildren from the schools covered in this study, it is possible that a significant proportion of these individuals will be positive for anti-*Toxocara canis* antibodies, since parasite eggs have been identified in animal feces in the vicinity of the schools.

Concerning *Trichuris* spp. eggs, they may have belonged to the species *Trichuris vulpis*, which is a nematode that occurs in dogs, since dogs were often seen in the vicinity of these schools, even following children to the school gates. It should be highlighted that this finding requires attention because the zoonotic potential of *T. vulpis* has already been proven and its importance has been acknowledged, because this helminth may cause damage to digestive system (Traversa, 2011; Márquez-Navarro et al., 2012; Mohd-Shaharuddin et al., 2019).

Education for the population regarding hygiene measures and attention to the health of animals that are kept by the population not only enables changes in concepts and habits relating to zoonoses but also encourages responsible pet ownership. The importance of sanitary care for pets and responsible pet rearing in households needs to be emphasized, since lack of knowledge among the population regarding the risks involved in these diseases is one of the main factors that trigger these diseases (Peruca et al., 2009). From this, an educational folder about "environmental contamination by parasites and the importance of responsible pet ownership" was distributed to schools students.

There was high occurrence of helminth eggs with zoonotic potential in dog feces found in the vicinity of elementary schools in Pelotas, RS, Brazil. The results showed the role played by dogs in environmental contamination and emphasized the need for practices that prevent and control parasite infections. Such practices need to be implemented in association with responsible pet ownership and control over stray dogs, so as to decrease contamination in these areas and, consequently, decrease the population's exposure to these infections.

References

Acha PN, Szyfres B. *Zoonoses and communicable diseases common to man and animals: parasitoses*. Washington: PAHO; 2003.

Blazius RD, Emerick S, Prophiro JS, Romão PRT, Silva OS. Ocorrência de protozoários e helmintos em amostras de fezes de cães errantes da cidade de Itapema, Santa Catarina. *Rev Soc Bras Med Trop* 2005; 38(1): 73-74. http://dx.doi.org/10.1590/S0037-86822005000100018. PMid:15717103.

Bowman DD, Montgomery SP, Zajac AM, Eberhard ML, Kazacos KR. Hookworms of dogs and cats as agents of cutaneous larva migrans. *Trends Parasitol* 2010; 26(4): 162-167. http://dx.doi.org/10.1016/j.pt.2010.01.005. PMid:20189454.

Capuano DM, Rocha GM. Ocorrência de parasitas com potencial zoonótico em fezes de cães coletadas em áreas públicas do município de Ribeirão Preto, SP, Brasil. *Rev Bras Epidemiol* 2006; 9(1): 81-86. http://dx.doi.org/10.1590/S1415-790X2006000100010.

Carvalho EAA, Rocha RL. Toxocariasis: visceral *larva migrans* in children. *J Pediatr* 2011; 87(2): 100-110. http://dx.doi.org/10.1590/S0021-75572011000200004. PMid:21503372.

Ferreira FP, Dias RCF, Martins TA, Constantino C, Pasquali AKS, Vidotto O, et al. Frequência de parasitas gastrointestinais em cães e gatos do município de Londrina, PR, com enfoque em saúde pública. *Semina: Ciênc Agrár* 2013; 34(6): 3851-3858. http://dx.doi.org/10.5433/1679-0359.2013v34n6Supl2p3851.

Ferreira JIGS, Pena HFJ, Azevedo SS, Labruna MB, Gennari SM. Occurrences of gastrointestinal parasites in fecal samples from domestic dogs in São Paulo, SP, Brazil. *Rev Bras Parasitol Vet* 2016; 25(4): 435-440. http://dx.doi.org/10.1590/s1984-29612016081. PMid:27925072.

Hotez PJ, Bottazzi ME, Franco-Paredes C, Ault SK, Periago MR. The neglected tropical diseases of latin America and the Caribbean: a review of disease burden and distribution and a roadmap for control and elimination. *PLoS Negl Trop Dis* 2008; 2(9): e300. http://dx.doi.org/10.1371/journal.pntd.0000300. PMid:18820747.

Hotez PJ. Neglected infections of poverty in the united states and their effects on the brain. *JAMA Psychiatry* 2014; 71(10): 1099-1100. http://dx.doi.org/10.1001/jamapsychiatry.2014.1045. PMid:25103827.

Katagiri S, Oliveira-Sequeira TCG. Prevalence of dog intestinal parasites and risk perception of zoonotic infection by dog owners in São Paulo State, Brazil. *Zoonoses Public Health* 2008; 55(8-10): 406-413. http://dx.doi.org/10.1111/j.1863-2378.2008.01163.x. PMid:18811905.

Klimpel S, Heukelbach J, Pothmann D, Ruckert S. Gastrointestinal and ectoparasites from urban stray dogs in Fortaleza (Brazil): high infection risk for humans? *Parasitol Res* 2010; 107(3): 713-719. http://dx.doi.org/10.1007/s00436-010-1926-7. PMid:20532563.

La Torre F, Di Cesare A, Simonato G, Cassini R, Traversa D, di Regalbono AF. Prevalence of zoonotic helminths in Italian house dogs. *J Infect Dev Ctries* 2018; 12(8): 666-672. http://dx.doi.org/10.3855/jidc.9865.

Labruna MB, Pena HFJ, Souza SLP, Pinter A, Silva JCR, Ragozo AMA, et al. Prevalência de endoparasitas em cães da área urbana do município de Monte Negro, Rondônia. *Arq Inst Biol (Sao Paulo)* 2006; 73(2): 183-193.

Leung AKC, Barankin B, Hon KLE. Cutaneous larva migrans. *Recent Pat Inflamm Allergy Drug Discov* 2017; 11(1): 2-11. http://dx.doi.org/10.2174/1872213X11666170110162344. PMid:28078983.

Lopes TV, Fernandes CPM, Michelon L, Hijano A, Félix SR, Schons SV, et al. Parasitas zoonóticos em fezes de cães de praças públicas em municípios da região sul do Rio Grande do Sul, Brasil. *Rev Bras Hig Sanid Anim* 2014; 8(2): 242-250.

Márquez-Navarro A, García-Bracamontes G, Álvarez-Fernández BE, Ávila-Caballero LP, Santos-Aranda I, Díaz-Chiguer DL, et al. *Trichuris vulpis* (Froelich, 1789) infection in a child: a case report. *Korean J Parasitol* 2012; 50(1): 69-71. http://dx.doi.org/10.3347/kjp.2012.50.1.69. PMid:22451737.

Medina-Pinto RA, Rodríguez-Vivas RI, Bolio-González ME. Zoonotic intestinal nematodes in dogs from public parks in Yucatán, México. *Biomedica* 2018; 38(1): 105-110. http://dx.doi.org/10.7705/biomedica.v38i0.3595. PMid:29668139.

Mendonça LR, Figueiredo CA, Esquivel R, Fiacconed RL, Pontes-de-Carvalho L, Cooper P, et al. Seroprevalence and risk factors for *Toxocara* infection in children from an urban large setting in

Northeast Brazil. *Acta Trop* 2013; 128(1): 90-95. http://dx.doi.org/10.1016/j.actatropica.2013.06.018. PMid:23845771.

Mohd-Shaharuddin N, Lim YAL, Hassan NA, Nathan S, Ngui R. Molecular characterization of *Trichuris* species isolated from humans, dogs and cats in a rural community in Peninsular Malaysia. *Acta Trop* 2019; 190: 269-272. http://dx.doi.org/10.1016/j.actatropica.2018.11.026. PMid:30500371.

Moura MQ, Jeske S, Vieira JN, Corrêa TG, Berne MEA, Villela MM. Frequency of geohelminths in public squares in Pelotas, RS, Brazil. *Rev Bras Parasitol Vet* 2013; 22(1): 175-178. http://dx.doi.org/10.1590/S1984-29612013000100034. PMid:24252968.

Novaes MT, Martins IVF. Avaliação de diferentes técnicas parasitológicas no diagnóstico de helmintoses caninas. *Rev Bras Med Vet* 2015; 37(Supl. 1): 71-76.

Oliveira VSF, Melo DPG, Fernandes PR, Schulze CMB, Guimarães MS, Silva AC. Ocorrência de helmintos gastrintestinais em cães errantes na cidade de Goiânia-Goiás. *Rev Patol Trop* 2009; 38(4): 279-283.

Peruca LCB, Langoni H, Lucheis SB. Larva *migrans* visceral e cutânea como zoonoses: revisão de leitura. *Vet Zootec* 2009; 16(4): 601-616.

Prates L, Pacheco LS, Kuhl JB, Dias MLGG, Araújo SM, Pupulin ART. Frequência de parasitos intestinais em cães domiciliados da cidade de Maringá, PR. *Arq Bras Med Vet Zootec* 2009; 61(6): 1468-1470. http://dx.doi.org/10.1590/S0102-09352009000600033.

Reichert F, Pilger D, Schuster A, Lesshafft H, Guedes de Oliveira S, Ignatius R, et al. Epidemiology and morbidity of hookworm-related cutaneous larva migrans (HrCLM): results of a cohort study over a period of six months in a resource-poor community in Manaus, Brazil. *PLoS Negl Trop Dis* 2018; 12(7): e0006662. http://dx.doi.org/10.1371/journal.pntd.0006662. PMid:30024875.

Robertson ID, Thompson RC. Enteric parasitic zoonoses of domesticated dogs and cats. *Microbes Infect* 2002; 4(8): 867-873. http://dx.doi.org/10.1016/S1286-4579(02)01607-6. PMid:12270734.

Santana BB, Silva TLB, Ramos RAN, Alves LC, Carvalho GA. Evaluation of different parasitological techniques for diagnosing intestinal parasites in dogs. *Open J Vet Med* 2015; 5(02): 19-24. http://dx.doi.org/10.4236/ojvm.2015.52003.

Santarém VA, Giuffrida R, Zanin GA. Larva *migrans* cutânea: ocorrência de casos humanos e identificação de larvas de *Ancylostoma* spp. em parque público do município de Taciba, São Paulo. *Rev Soc Bras Med Trop* 2004; 37(2): 179-181. http://dx.doi.org/10.1590/S0037-86822004000200014. PMid:15094907.

Santos PC, Telmo PL, Lehmann LM, Lorenzi C, Hirsch C, Mattos GT, et al. Frequency of *Toxocara* spp. antibodies in umbilical cords of newborns attended at the University Hospital in Southern Brazil and factors associated with infection. *Acta Trop* 2017; 170: 43-47. http://dx.doi.org/10.1016/j.actatropica.2017.02.003. PMid:28188768.

Scaini CJ, Toledo RN, Lovatel T, Dionello MA, Gatti FA, Susin L, et al. Contaminação ambiental por ovos e larvas de helmintos em fezes de cães na área central do Balneário Cassino, Rio Grande do Sul. *Rev Soc Bras Med Trop* 2003; 36(5): 617-619. http://dx.doi.org/10.1590/S0037-86822003000500013. PMid:14576878.

Schoenardie ER, Scaini CJ, Brod CS, Pepe MS, Villela MM, McBride AJ, et al. Seroprevalence of *Toxocara* infection in children from southern Brazil. *J Parasitol* 2013; 99(3): 537-539. http://dx.doi.org/10.1645/GE-3182. PMid:23738711.

Sviben M, Cavlek TV, Missoni EM, Galinovic GM. Seroprevalence of *Toxocara canis* infection among asymptomatic children with eosinophilia in Croatia. *J Helminthol* 2009; 83(4): 369-371. http://dx.doi.org/10.1017/S0022149X09381213. PMid:19460194.

Táparo CV, Perri SHV, Serrano ACM, Ishizaki MN, Costa TP, Amarante AFT, et al. Comparação entre técnicas coproparasitológicas no diagnóstico de ovos de helmintos e oocistos de protozoários em cães. *Rev Bras Parasitol Vet* 2006; 15(1): 1-5. PMid:16646994.

Traversa D. Are we paying too much attention to cardio-pulmonary nematodes and neglecting old-fashioned worms like *Trichuris vulpis? Parasit Vectors* 2011; 4(1): 32. http://dx.doi.org/10.1186/1756-3305-4-32. PMid:21385441.

Villela MM, Pepe MS, Ferraz ML, Morais NCM, Araújo AB, Ruas JL, et al. Nota: contaminação ambiental da orla da Laguna dos Patos (Pelotas, RS, Brasil), por parasitos com potencial zoonótico. *Vittalle* 2009; 21(2): 69-74.

Willis HH. A simple levitation method for the detection of hookworm ova. *Med J Aust* 1921; 2(18): 375-376. http://dx.doi.org/10.5694/j.1326-5377.1921.tb60654.x.