
PREVALENCE OF PARASITES WITH ZOONOTIC POTENTIAL IN SOIL FROM THE MAIN PUBLIC PARKS AND SQUARES IN CAXIAS DO SUL, RS, BRAZIL

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ABSTRACT

Soil contamination in parks and public squares caused by animal feces has been one of the main forms of parasitic contamination, becoming therefore a public health problem. The purpose of this study was to determine the prevalence of parasites in the soil in major parks and public squares in Caxias do Sul, RS, Brazil. Two samplings were performed at each location, the first in summer (February) and the second in winter (July) in 2015. Soil samples were collected in five spots in ten parks/public squares, per season and examined using the Hoffman, Pons & Janer (HPJ), Faust and Rugai, Mattos and Brisola methods. Among the 100 samples analyzed by HPJ and Rugai, Mattos and Brisola, 64% and 58% respectively, proved positive in summer and 28% and 60% in winter. The Faust method showed the same number of positive samples in both seasons (12% each). The following parasites were found: *Cystospora* sp oocysts (4%), *Entamoeba coli* cysts (1%), *Strongyloides stercoralis* suggestive larvae (0.3%), *Endolimax nana* cysts (1%) and *Ancylostoma* sp suggestive larvae (33.3%) with the highest prevalence. Both parks and squares presented parasites with zoonotic potential. Thus, it is necessary to highlight the importance of educational measures regarding the use of these locations as well as the cleaning of public areas with the removal of animal excrements.

KEY WORDS: Parasites; enteroparasites; public health; environment.

INTRODUCTION

The natural environment and mankind have been mutually attracted since earliest times. To survive, humans have always needed natural resources (Santin, 1993), and proof of this is in the increasing number of individuals who have practiced physical activities outdoors in recent years (Albino et al., 2010) in parks and public squares which are ideal for this kind of activity (Silveira & Silva, 2010). Physical exercise is a form of entertainment and is beneficial in reducing the adverse effects of stress from work, helping to improve self-esteem (Silveira & Silva, 2010).

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Received for publication: 12/6/2016. Reviewed: 17/3/2017. Accepted: 20/3/2017.

While physical activity is beneficial for the health of human beings, it can contribute to the development and spread of zoonotic diseases, given that individuals do not always take the necessary precautions to avoid exposure to microorganisms, such as skin contact with parasite contaminated soil. Poor environmental conditions and housing added to lack of hygiene, inadequate sanitation and basic levels of education, are some of the main factors contributing to the transmission of parasitic infections (Robertson et al., 2000; Anaruma, 2002; Traub et al., 2004; Pastório et al., 2009).

It is known that both dogs and cats, domestic or not, may defecate on streets, making these places a major focus for zoonosis transmission (Capuano & Rocha, 2005). This happens mainly because many owners are careless about collecting feces and the collection of this material by the public service may not be immediate. There are many organisms that parasitize these animals and when parasitic structures, such as cysts, oocysts, eggs and larvae are excreted in feces and dropped on the soil, they can infect a new host, be it animal or human (Pastório et al., 2009). Among the species that parasitize these animals, the most common are *Toxocara canis* or *Toxocara cati*, *Ancylostoma* sp, *Trichuris vulpis* or *Trichuris campanula* and *Giardia lamblia*. *Toxocara* sp and *Ancylostoma* sp etiological agents of visceral and cutaneous *Larva migrans* (Schantz, 1991; Robertson et al., 2000; Dunn et al., 2002).

Estimates show that parasites affect around 3.5 billion people worldwide, causing diseases in approximately 450 million. Children are the most affected (WHO, 2008; Neves et al., 2012) as their recreational activities are often directly linked to soil and sand, and because they are more in contact with animals. Children also often present disorders like geophagia and onychophagia, habits which favor the intake of eggs and/or cysts of various parasites contained in the soil (Glickman & Schantz, 1981; Capuano & Rocha, 2006; Chiodo, 2006; Motazedian et al., 2006; Gawor, 2008). Parasites may cause various physical disturbances to their hosts, such as inflammations, nutritional, neurological and cognitive problems, bleeding, difficulty in defecating and rectal prolapse. They may also affect fetal development, leading to physical and mental retardation, and even be the cause of death (Pessoa & Martins, 1978; Santos & Merlini, 2010).

Many government programs have been implemented in several countries in order to control these diseases caused by intestinal parasites. However, in developing countries, these initiatives are not very effective due to insufficient financial support for adequate sanitation, and the lack of participation of the population itself (WHO, 1987; Pedrazzani et al., 1989).

The purpose of this study was, therefore, to evaluate the prevalence of soil and sand contamination by parasitic structures from worms and protozoa in public parks and squares in Caxias do Sul, RS. Furthermore, a comparative study was performed on the prevalence of parasites within the period of one month in summer and one month in winter.

MATERIAL AND METHODS

To assess the prevalence of soil contamination by worms and protozoa, during February (summer) and July (winter) 2015, soil and sand sampling were performed in the Monteiro Lobato, Bandeira, Presidente Joao Pessoa and Dante Alighieri squares, in the Getulio Vargas, Cinquentenário and Lagoa parks, in the playgrounds around the Cinquentenário and Madureira neighborhoods and in the Botanical Gardens. These squares and parks are located in Caxias do Sul, Rio Grande do Sul, Brazil, and were chosen because they are frequently used by people for leisure and outdoor activities.

From each square and park (totaling 10 places), five soil samples were collected in the summer and five in the winter using gardening shovels. Each sample consisted of approximately 100g of soil and was collected at a depth of about 10 cm. The samples were placed in sterile plastic bags, labeled and kept in a cool box for transportation. The samples were analyzed in the Parasitology Laboratory of the Faculdade da Serra Gaúcha, in Caxias do Sul, RS, following the Hoffman, Pons and Janer (HPJ), Faust, and Rugai, Mattos and Brisola methods (De Carli, 2007). These methodologies were chosen due to the possibility of identifying specific types of parasites. The HPJ method is regularly used in laboratories and is based on spontaneous sedimentation, contributing to the visualization of helminth eggs/larvae and protozoan oocysts. The Faust method is used to verify the existence of cysts and oocysts from protozoa by centrifugal flotation in zinc sulfate, and the Rugai, Mattos and Brisola method is based on positive hydrotropism of nematode larvae (De Carli, 2007). In addition, these techniques are not expensive and are easily applied.

The samples were stained in duplicate using Lugol, covered with coverslips and observed under optical microscopy at 10x and 40x by two microscopists. This study was evaluated qualitatively, so every sample presenting at least one parasitic structure was considered positive, highlighting the evolutionary stage and the identification of the parasite.

Data were processed in the Statistical Package for Social Sciences (SPSS) version 19.0. The data, being categorical, were expressed in absolute frequencies and values. Station chi-square test was used for comparative analyzes, where a 5% significance level was adopted for statistical difference.

RESULTS

According to Table 1, within a total of 100 samples analyzed (50 per season) following the HPJ method in the first sampling (in summer), 64% (n = 32) were positive, while in the second sampling (in winter) 28% of the samples were positive (n = 14). According to the analysis performed using the Faust method, in the first and second samplings, 12% (n = 6) of the samples were

positive for each sampling. In the analysis performed with the Rugai, Mattos and Brisola method, in the first sampling 58% (n = 29) of the samples were positive and 60% (n = 30) in the second sampling. No statistical difference was observed comparing the samples per season of the year.

Table 1. Positive soil samples for parasites collected in February and July 2015 in public squares and parks in Caxias do Sul, RS, Brazil.

Method	1st sampling (n=50)	2nd sampling (n=50)
	February	July
Hoffman, Pons & Janer	32 (64%)	14 (28%)
Faust	6 (12%)	6 (12%)
Rugai, Mattos & Brisola	29 (58%)	30 (60%)

Table 2 shows the presence of two or more parasitic structures in the samples. It is possible to notice that 60% (n = 6) of the squares/parks presented polyparasitism in the first sampling analyzed with HPJ method, although in the second sampling this percentage decreased to 30% (n = 3). In relation to the Faust method, there was no presence of polyparasitism for the first sampling, and only 10% (n = 1) of the squares presented polyparasitism in the second sampling. For those samples analyzed using the Rugai, Mattos and Brisola method, neither of the samplings presented polyparasitism.

Table 2. Prevalence of polyparasitism in the samples collected in February and July 2015 in public squares and parks in Caxias do Sul, RS, Brazil.

Method	1st sampling (n=50)	2nd sampling (n=50)
Hoffman, Pons & Janer	6 (60%)	3 (30%)
Faust	-	1 (10%)
Rugai, Mattos & Brisola	-	-

Among the 300 analyses (100 samples per method) shown in Table 3, in 61% (n = 183) parasitic structures were not observed. In 39% (n = 117) of the positive samples, 33.3% (n = 100) contained the presence of larvae suggestive of *Ancylostomidea*, 4% (n = 12) *Cystospora sp* oocysts, 1% (n=3) *Entamoeba coli* cysts, 0.3% (n = 1) larvae suggestive of *Strongyloides sp* and 0.3% (n = 1) *Endolimax nana* cysts.

Table 3. Prevalence of parasites in soil from public squares and parks in Caxias do Sul, RS, Brazil in 2015, adding positivity to all methods performed.

Result	N	%
Negative	183	61
Suggestive of <i>Ancylostoma</i> sp larvae	100	33.3
<i>Cystospora</i> sp oocysts	12	4
<i>Entamoeba coli</i> cysts	3	1
Suggestive of <i>Strongyloides</i> sp larvae	1	0.3
<i>Endolimax nana</i> cysts	1	0.3
Total number of analyses	300	100

After 300 analyses, this study detected that all the parks and squares were contaminated by some form of parasitic structure. Table 4 shows the frequency of positivity for each square/park along with the species found. The most parasitized place was João Pessoa Square, with a positivity of 5.3%, while Lagoa Park was the least parasitized site, with a positivity of 2.6%.

Table 4. Frequency of positivity (%) for isolated parasites in soil/sand from public squares and parks in Caxias do Sul, RS, in February and July 2015, adding positivity to all methods performed (n=300 analyses).

Sampling sites	Larvae (suggestive)		Cysts		Oocysts
	<i>Ancylostoma</i> sp	<i>Strongyloides</i> sp	<i>Entamoeba coli</i>	<i>Endolimax nana</i>	<i>Cystospora</i> sp
Squares					
Bandeira	3	-	0.3	-	-
João Pessoa	4	-	0.3	0.3	0.7
Monteiro Lobato	2.7	-	0.3	-	1.7
Dante Alighieri	3.3	-	-	-	-
Parks					
Getúlio Vargas	4	0.3	-	-	-
Cinquentenário	4.3	-	-	-	-
Lagoa	2.3	-	-	-	0.3
Jardim Botânico	3	-	-	-	0.3
Parquinho Cinquentenário	3.3	-	-	-	0.3
Parquinho Madureira	3.3	-	-	-	0.7

DISCUSSION

This study demonstrated that all analyzed parks and squares were potential transmitters of parasites to individuals who use them for any recreational purposes. Among the parks and squares analyzed, 39% were contaminated by at least one parasitic stage. Among the positive samples the following parasitic structures were identified: larvae suggestive of *Ancylostoma* sp and of *Strongyloides* sp, *Cystospora* sp oocysts, *Entamoeba coli* and *Endolimax nana* cysts. In studies carried out in the southern region of Brazil 52% parasitic structures were observed by Mascarenhas and da Silva (2016) in soil from recreation areas in schools and 40% by Pritsch and Frighetto (2016) in public squares. *Ancylostoma* sp was the only worm in common with ours in both studies.

The highest prevalence noted was the genus *Ancylostoma* sp (33.3%), corroborating the study conducted by Miranda et al. (2015), where the presence of this parasite was observed in all samples, and Nunes et al. (2000), who after analyzing sand samples from recreational areas from schools in Araçatuba, SP, Brazil, through the centrifugal flotation and Baermann methods, verified the presence 46.4% of this kind of larvae. Scaini et al. (2003) analyzed 237 samples of soil in the city of Balneario Casino, RS in 2002, using Willis flotation method, and found 169 (71.3%) positive samples for *Ancylostoma* sp. In a study carried out by Ribeiro et al. (2013), a frequency of 85% was found for these larvae in squares in the city of Belo Horizonte, using Caldwell & Caldwell and HPJ methods.

According to Labruna et al. (2006) the genus *Ancylostoma* is the most common in dogs and cats, confirming the high prevalence of positive samples in our study, as the number of these animals in public areas is high. Coelho et al. (2011) found *Ancylostoma* sp eggs in 87.8% of the fecal samples collected from dogs and in 94.2% of the fecal samples collected from cats using the Willis-Mollay method in Andradina, São Paulo.

This helminth, which is the etiologic agent for cutaneous *Larva migrans*, is mainly transmitted through active penetration into the skin by direct contact of individuals with contaminated soil, usually through the hands or feet. This confirms the high probability of contamination in public areas (Neves & Massara, 2009; Pastório et al., 2009).

Concerning the methods used, after analyzing all the samples, the Rugai, Mattos Brisola and HPJ methods were found to be the most effective because of the number of positive samples for *Ancylostoma* sp larvae. In the samples analyzed using the Faust method, the positive percentage was lower. This is probably due to the low prevalence of protozoa in the samples, or to the low specificity of this method for analyzing this kind of material.

In a study conducted by Taparo et al. (2006), in which methods for parasitological analysis used to identify helminth eggs and protozoan oocysts in

dogs were compared, the Mollay-Willis method and spontaneous sedimentation were the most effective, while the Faust method was the least efficient. In another study conducted by Ross et al. (2011), which verified the prevalence of eggs, larvae, cysts and oocysts of parasites in public squares in the city of Cruz Alta, RS, using the Willis-Mollay, Faust and HPJ methods, 26 positive samples were found among those analyzed with the HPJ method, while the Faust and the Willis-Mollay presented 6 and 4 positive samples, respectively. In the same study, a larger variety of parasitic genera was observed when compared to this study, given that the 34 positive samples (n=100) contained: *Ascaris* sp eggs; *Ancylostoma* sp eggs; *Trichuris* sp eggs; *Dipylidium caninum* eggs; *Toxoplasma gondii* oocysts; *Sarcocystis* sp oocysts; *Giardia* sp cysts; *Toxocara canis* eggs; and *Gnathostoma* sp eggs. Neves and Massara (2009), using spontaneous sedimentation and the Faust method, observed *Toxocara* sp, which was not found in this study, and/or cysts of *Entamoeba* sp (which we also found) in 89.6% of the samples.

Although recent studies (Cavagnolli et al., 2015; Camello et al., 2016; Rech et al., 2016) have shown that the prevalence of parasitic diseases in children (highest risk group) is not high in our city and nearby municipalities, care must be taken since there are parasites in the soil as shown in this study. Given that these sites are often used for recreational activities and sports, and that people are unaware of the risk they are exposed to, preventive measures should be applied. The large amount of garbage that is often seen on streets, as well as stray animals and feces from these animals, or even from humans, besides other factors, contributing to the transmission of diseases, are a public health issue, as they are directly related to poor hygiene and insufficient education, as well as unhealthy environmental conditions and the population's lack of information (Robertson et al., 2000; Anaruma, 2002; Traub et al., 2004; Pastório et al., 2009).

Hopefully, these results will stimulate the proper offices to take actions regarding decontamination of these places, thus ensuring the health of the population. We also reinforce the importance of pet owners collecting their animals' feces and periodically consulting the veterinarian for treatment. This will avoid inflammation, nutritional, neurological (physical and mental retardation) and cognitive problems, among other consequences of parasitic infections. Control and prevention of zoonotic parasitic diseases avoid public expenditure for future treatments.

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