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# Environmental contamination: influence of seasonality on the occurrence of geohelminths in a public area of Fortaleza, Ceará

Contaminação ambiental: influência da sazonalidade na ocorrência de geo-helmintos em área pública de Fortaleza, Ceará

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# ABSTRACT

**Introduction:** Contamination of public areas by infectious forms of zoonotic parasites is a serious public health problem worldwide. A high number of stray animals, seasonality and anthropization of the environment are important elements in the spread of these parasitic infections. **Objective:** The present study evaluated the influence of seasonality on soil contamination in a public area in the city of Fortaleza, Ceará, by two genera of geohelminths (*Toxocara* spp and *Ancylostoma* spp). **Method:** 480 soil samples were collected in a public area, corresponding to 240 samples during the dry season and 240 samples in the rainy season. They were processed and analyzed individually using the method of Rugai and Willis. **Results:** The results reveal a high rate of parasitic contamination in the analyzed soil samples (82.5%). The comparison of contamination in seasonal periods showed a significant difference (p < 0.05) between the percentage of samples contaminated during the dry season (96.7%) and the samples from the rainy season (68.3%). Microscopic analysis showed a predominance of eggs of *Toxocara* spp. and *Ancylostoma* spp. **Conclusions:** The results show a high parasitic contamination of soil, influenced by the rainfall indexes of the studied area.

KEYWORDS: Public Health; Zoonoses; Soil; Parasites; Dry and Rainy Seasons

# RESUMO

Introdução: A contaminação de locais públicos por formas infectantes de parasitos zoonóticos constitui um grave problema de saúde pública a nível mundial. Um elevado número de animais errantes, a sazonalidade e a antropização do ambiente são elementos importantes na disseminação destas infecções parasitárias. Objetivo: O presente estudo avaliou a influência da sazonalidade na contaminação do solo em uma área pública na cidade de Fortaleza, Ceará, por dois gêneros de geo-helmintos (Toxocara spp. e Ancylostoma spp.). Método: Um total de 480 amostras de solo foi coletado em uma área pública, correspondendo a 240 amostras durante a estação seca e 240 amostras na estação chuvosa na cidade de Fortaleza, as quais foram processadas e analisadas individualmente pelos métodos de Rugai e Willis. Resultados: Um alto índice de contaminação parasitária nas amostras de solo analisadas (82,5%) foi evidenciado. A comparação sazonal da contaminação mostrou uma diferença significativa (p < 0,05) entre a porcentagem de amostras contaminadas no período de estiagem (96,7%) e as amostras do período chuvoso (68,3%). Na análise microscópica, observou-se uma predominância de ovos de Toxocara spp. e Ancylostoma spp. Conclusões: Os resultados demostraram um grau elevado de contaminação parasitária do solo influenciada pelos índices pluviométricos da área estudada.

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#### **INTRODUCTION**

The high rates of environmental contamination by animal and human waste in urban centers shows the importance of these places in the spread of parasitic diseases. This type of contamination is a public health problem, particularly in tropical and subtropical regions of developing countries<sup>1,2</sup>. Public areas destined to recreational, sporting and educational activities have many environmental matrices of primary contact with the population (water, soil and sand), which need to be monitored periodically, since they can represent a risk to human health, mainly due to the population growth of stray dogs and cats throughout Brazil<sup>3</sup>.

The transmission of several zoonoses in urban areas is directly associated with the increase in the number of these animals. Zoonoses are characterized as all diseases and infections transmitted naturally between vertebrate animals and humans. Several studies have shown that these pathologies are among the group of neglected tropical diseases with a significant impact on public health<sup>4,5,6</sup>, especially for children, due to recreational activities, hygiene habits and appetite disorders, as geophagy, which result in the ingestion of eggs and/or cysts present in contaminated soil<sup>7,8,9</sup>.

In addition, factors as the soil pattern, high temperature and humidity, as well as the modifications produced by human activity, can influence directly or indirectly the parasite-host interaction, allowing the creation of favorable environments for the transmission of parasitic agents. In the case of geohelminths, this enables the development of eggs and larvae until the infectious stage<sup>3,10</sup>. However, the successful continuity of the evolutionary cycle of a parasite also depends on its ability to tolerate environmental changes through mechanisms that are inherent in each species<sup>11</sup>.

Infections caused by helminths, related to soil contamination, are morbidities associated with poor socioeconomic and sanitary conditions<sup>12,13</sup>. According to the World Health Organization, about 1.5 billion people are affected by intestinal parasitic infections. The severity of these diseases is mainly related to its chronic and insidious impact on health, impairing physical and cognitive development<sup>14</sup>.

Studies carried out in Brasil<sup>15,16,8,13</sup> and in other parts of the world, like Cuba<sup>6</sup>, Poland<sup>18</sup>, India<sup>18</sup> and Chile<sup>5</sup>, have demonstrated that *Toxocara* spp. and *Ancylostoma* spp. are the main geohelminths found in the soil and in stool samples from public spaces. Thus, studies in the field of environmental parasitology have enabled the identification of important biomarkers in the assessment of the health conditions of public areas, as well as the correlation between seasonal variations and the continuity of the parasitic evolution cycle. These studies work as an important tool, both for the epidemiological understanding of these infections and for the implementation of integrated health education programs.

Therefore, the aim of the present study was to evaluate the influence of seasonality on the soil contamination by *Toxocara* spp. and *Ancylostoma* spp. in a public area of Fortaleza, state of Ceará (CE), Brazil.

### **METHODOLOGY**

This study is classified as a cross-sectional study with a quantitative and experimental approach. It was conducted in a public area of Fortaleza (Brazil), during the dry season, from August to December 2015, and the rainy season, from January to July 2016.

Fortaleza is located on the central coast of the state of Ceará, at an altitude of 16 m above sea level ( $03^{\circ}43'06''S$ ;  $38^{\circ}32'34''W$ ), with an estimated population of 2,643,247, semi-humid tropical climate with temperature averages of 26.3°C and irregular annual rainfall<sup>19</sup>. The predominant soil type is red-yellow latosol with humidity around 73.0% to 82.5%<sup>20</sup>.

The collection points were selected based on the ecological niche occupied by the population of stray animals in the area. Thus, four collection points were determined and at each point an area of  $320 \text{ m}^2$  was established, divided into four quadrants of  $80 \text{ m} \times 80 \text{ m}$ . The sampling points were defined by the zigzag collection methodology, as described by González-Cárceres et al.<sup>21</sup>.

From each collection point, 120 soil samples were collected. Therefore, we had 240 samples collected in the dry season and 240 in the rainy season, completing 480 samples throughout the study period. The collections were carried out on alternate days, in the morning, with the aid of an aluminum labyrinth placed approximately 3 cm deep in relation to the surface, with a minimum difference of 5 m away from any suspected fecal contamination. Approximately 300 g of soil from each collection point were removed, packed in properly identified plastic containers, transported under refrigeration at  $4^{\circ}$ C to the Parasitology Laboratory at the State University of Ceará.

Soil aliquots were individually processed to search for light helminth eggs, using the fluctuation method in hypersaturated sugar solution (d = 1.24)<sup>22</sup>. Briefly, approximately 14 g of soil were homogenized in 60 mL of saturated sugar solution. The resulting suspension was sieved and a 5 ml aliquot was transferred to glass tubes. After the meniscus was formed, a glass cover slip was placed over the tube, and after 5 min, the cover slip was carefully removed and placed on a microscope slide to investigate the parasites stages in each sample. The material was analyzed in triplicate under optical microscope at 100x and 400x magnification.

For the detection and identification of nematode larvae, we used the Rugai, Mattos and Brisola<sup>23</sup> technique, which consists of wrapping 100 g of soil sample over pieces of folded gauze (30 cm X 30 cm), forming a small "bundle" immersed in a sedimentation cup, containing water heated to  $45^{\circ}$ C in sufficient quantity to



enable direct contact of the water with the sample. After 1 h, the gauze containing the sample was removed and the material was sedimented again for 1 h. After this period, 3 mL of the sediment were distributed in a watch glass and left to rest for 5 min, the time it takes for the larvae to settle in the center of the watch glass. The identification of the collected larvae was done according to the morphometry parameters proposed by Yamaguti<sup>24</sup>. The slides were analyzed under optical microscope at 100x magnification.

The data obtained from the soil analysis were tabulated using Microsoft Excel 2010 software, and the results are presented in the form of graphs. The statistical analysis was performed using the GraphPad Prism version 3.0. The chi-square test (c2) in contingency tables (2 X 2) with Yates's correction was used to compare the percentages of parasitic contamination found in the different seasonal periods. Values of p <0.05 were considered statistically significant.

## **RESULTS AND DISCUSSION**

Of a total of 480 soil samples analyzed from a public area in the city of Fortaleza, 82.5% presented contamination by developmental stages of parasites, corroborating the studies by Capella et al.<sup>25</sup> that found an index of 100.0% contamination by parasitic forms in soil samples from a community in the municipality of Pelotas, state of Rio Grande do Sul. Similarly, Pedrosa et al.<sup>10</sup> demonstrated that 94.4% of soil samples obtained from the beaches of Fortaleza were contaminated by parasites of medical importance.

In the seasonality analysis, the results have shown that 96.7% (232) of the soil samples obtained during the dry season and 68.3% (164) of the samples obtained during the rainy season showed parasitic contamination (Figure 1). The correlation between rainfall and the percentage of positivity of the samples has shown that the region's rainfall indexes negatively influence the contamination with parasites (p <0.05), suggesting a leaching effect on the soil due to the action of rainwater, which can carry these parasites stages away from their original site.

The result obtained differs considerably from that found by Rêgo et al.<sup>13</sup>, who demonstrated a high parasitic load in the soil of public squares in the municipality of Macapá, state of Amapá, mainly in the winter, which corresponds to the rainy season in that region. On the other hand, results similar to those observed in this study were described by Figueiredo et al.<sup>26</sup>, who found higher percentages of helminths in sandbox samples from public schools of Uruguaiana, state of Rio Grande do Sul, during spring (36.7%) and summer (45.7%). The variation observed in different regions, regarding the seasonality-parasitic contamination of the soil, can be attributed to several factors, which include: (i) climatic parameters like humidity, temperature and soil oxygenation; (ii) socioeconomic and geographical issues; (iii) basic sanitation conditions; (iv) health education, among others<sup>27,28</sup>.

Regarding the relationship between the parasitic species found and seasonality, we found a high rate of *Ancylostoma* spp. and Toxocara spp. eggs (96.7% and 93.8%, respectively) and a lower occurrence of Ancylostoma spp. larvae (39.6%) in soil samples during the dry season (p <0.05) (Figures 2 and 3). The result obtained in this study demonstrated a high percentage of parasitic contamination when compared to that described by Brilhante, Nunes and Dorval<sup>29</sup>, who detected contamination rates of 28.0% for *Toxocara* spp. and 26.0% for hookworms in soil samples from a fishing community in Bonito, state of Mato Grosso do Sul.

On the other hand, the study carried out by Nooraldeen<sup>4</sup> in the city of Erbil, Iraq, has shown 50% contamination rate by *Toxocara* spp. eggs and 25% of *Ancylostoma* spp. in the soil of squares and public parks. The difference found between the percentage of species obtained in the present study and the data in the literature can be explained by the favorable environmental conditions in the region for the development of free-living stages of these genera.

According to Lopes et al.<sup>30</sup>, regions of tropical and subtropical climate, with high temperatures and adequate humidity, have ideal conditions for the development of the biological cycle and the spread of these parasites. In addition, the type of soil also acts as a decisive factor for the survival of helminth eggs.

Lima et al.<sup>31</sup>, in a study carried out in the municipality of Moreno, state of Pernambuco, found a higher frequency of eggs from the Ascarididae family, including *Toxocara* spp. eggs, especially in the quadrants with clayey soil. Thus, the climatic aspects mentioned above are characteristic of the present study's region, which has an average annual temperature of 26.6 °C, high relative humidity (73.0% to 82.5%) due to the maritime influence and the high evaporation rate, and red-yellow latosol type soil, which presents a clay texture, strong acidity and low natural fertility<sup>20</sup>.

Therefore, analyses of soil samples from a public area in the city of Fortaleza have shown that parasitic contamination in the region can be affected by seasonal changes. This observation can contribute to the implementation of effective measures to



**Figure 1.** Parasitic prevalence in samples of public area soil from the city of Fortaleza, Brazil, in dry and rainy seasons.





Figure 2. Distribution of parasitic stages of Ancylostoma spp. and Toxocara spp. in soil samples from a public area in the city of Fortaleza, Brazil, in the dry and rainy seasons.



Figure 3. Parasitic forms detected in soil samples from a public area in the city of Fortaleza, Brazil. (A) Ancylostoma spp. larvae; Ancyclostoma spp. egg; (c) Toxocara spp.

improve public health in the municipality, thus reducing the risk of zoonoses being acquired by the population.

## **CONCLUSIONS**

This study has shown a potential risk of transmission of zoonoses to the population, especially in the dry season, when a high percentage of geohelminth eggs and larvae were found. Additionally, the study reaffirms: (i) the importance of soil as an important vehicle for disease transmission, both for humans and animals; (ii) the population's awareness of the role of stray and domestic dogs and cats as carriers of anthropozoonoses; (iii) periodic monitoring by health authorities as important initiatives to control the quality of the soil used by the population and (iv) environmental education as a tool to improve the quality of life and the health profile of the population.

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#### Author's Contribution

Melo MVC - Conception, planning and review of the paper. Oliveira TR - Acquisition, analysis, interpretation of data and writing of the paper. Rodríguez-Malaga SM - Analysis, interpretation of data and review of the paper. Cruz DRS, Jonas JM - Sample acquisition and analysis. All authors approved the final draft of the paper.

#### **Conflicts of Interest**

The authors report that there is no potential conflict of interest with peers and institutions, politicians or financiers in this study.



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