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Soil Contamination With Eggs of *Toxocara* Species in Public Parks of Karaj, Iran

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Abstract

Background: Human toxocariasis is one of the zoonotic helminth diseases that is usually occurred with exposure to contaminated soil. Both *Toxocara canis* and *Toxocara cati* are considered the causative agents of *Toxocara* infection.

Objectives: This survey was intended to provide data on the *Toxocara* species eggs contamination in soil samples in the public parks of Karaj, Iran.

Materials and Methods: This study was carried out among 200 soil samples collected from 12 public parks between August and September 2016 to examine the soil contamination with *Toxocara* species eggs. Soil samples were tested for the presence of *Toxocara* eggs using sucrose flotation method.

Results: Prevalence of *Toxocara* species eggs in soil samples collected from public parks was 36.4%. The highest number of eggs recovered from 200 g of soil was 20. A total of 200 eggs were recovered and 7.6% were fully developed to embryonated egg stages. The contamination rate in the third region in 4 studied areas was higher than the other regions. A similar tendency was observed in park areas, so that parks higher than 5000 m^2 were highly contaminated.

Conclusion: According to the results of this study, soils of the public parks in Karaj are one of the main risk factors for human toxocariasis.

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Background

Toxocariasis is one of the zoonotic helminth diseases that is caused by the nematode larvae of *Toxocara canis* and *Toxocara cati*.[1] *Toxocara* infection has a worldwide distribution, and actually is an important health problem in developing countries. It is one of the most prevalent gastrointestinal parasites in dogs and cats worldwide. The seroprevalence found in humans reflects the frequency of contact with the parasite in a contaminated environment.[2] Human and animals are infected by eating contaminated foods with embryonated *Toxocara* spp. eggs specially paratenic host meat containing parasite larvae or when contacted with contaminated soil. This contact or ingestion may be without any clinical signs, even the parasite can involve the visceral tissues by larvae migration (VLM) that causes serious injury and finally dysfunction in vital organs (i.e., heart, liver, muscles etc), also has ocular syndrome (OLM) with symptoms such as blurred vision, double vision and in end stage leads to blindness.[3,4]

The highest risk for humans is represented by canine and feline fecal contamination of the environment, where *Toxocara* spp. eggs are developed and may remain infectious in the soil over a long time at different conditions. In the life cycle of toxocariasis and its transmission, the soil significant role is undeniable. *Toxocara* eggs need to stand in soil for 4-6 weeks to be embryonated and infectious.[5] Soil of public places, like parks, can become an important source of parasitic contamination, because of the highest traffic of dogs and cats, due to the increment in the domestic animal population in big cities.[7] In many provinces of Argentina, it has been found an association between a higher environmental prevalence of *Toxocara* spp. in the periphery of urban centers, due to a greater household's precarization and poor sanitary conditions. In regions where the prevalence was higher in urban centers, it coincided with a higher population density, higher density of fecal contamination in public spaces, and an urban demographic structure with similar socioeconomic conditions. Most studies in Argentina showed a relationship between human seroprevalence and soil contamination, and to a lesser extent, with the prevalence found in dogs.[6,7] Therefore, a survey was performed to estimate the degree of contaminated soil by

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Toxocara spp. eggs in public parks of Karaj, Iran.

Objectives

The current survey intended to assess the data on the *Toxocara* species eggs contamination in soil samples in the public parks of Karaj, Iran.

Materials and Methods

Study Regions

Karaj (35°49′57″, 50°59′29″) is capital of Alborz province, Iran, with a population of around 3 million, and is the fourth-largest city in Iran (after Tehran, Mashhad, and Isfahan). The climate of Karaj is to some extent cooler than Tehran's, and it receives 260 mm of rain annually. This area has yearly temperature of 15.1°C. This study was conducted between August and September 2016. Twelve public parks in the city of Karaj according to the size of the area, the traffic and the municipalities were selected for soil sampling.

Recovery of Soil

To estimate *Toxocara* spp. eggs, superficial soil samples at a depth of 3 to 5 cm were taken from areas identified in the parks (each sample of approximately 10 g every 50 steps). A sample of 500 g soil was packaged separately from parks in labeled and sealed bags. The soil samples were thoroughly mixed and stored in plastic bags and taken to the Parasitology Laboratory of School of Paramedicine, Alborz University of Medical Sciences, for recovery of *Toxocara* eggs.

Detection of Helminth Eggs

The soil samples were dried overnight at 37°C, then filtrated using a 150 µm mesh sieve. Almost 2 g of the powdery sand was placed in a tube mixed with 0.5% tween 20 solution and was shaked and centrifuged at 1500 rpm for 10 minutes. The supernatant was sucked off. The precipitate was re-suspended in 8 mL water and was again centrifuged under the same condition. The supernatant was removed and sediment was suspended in sucrose solution with a specific gravity of 1.200 to 1 cm from the top. The suspension was mixed well on vortex and centrifuged at 1500 rpm for 10 minutes. Then tubes containing the suspension were filled to the brim with additional sucrose solution and covered with a clean coverslip. After a final centrifugation of the tube together with coverslip at 600 rpm for 5 minutes, the coverslip was transferred to a glass slide and tested at the magnifications of ×40 and ×100 for *Toxocara* eggs. The ova found in the current study were counted and classified according to their developmental stage, namely, monocell, 2 to pre-embryonated and embryonated.

Statistical Analysis

Data analysis of the relationship between contamination rates and changes in environmental factors were evaluated by chi-square test. Probability (*P*) values of less than 0.05 were considered to be statistically significant ($P < 0.05$).

Results

A total of 200 soil samples were collected from Karaj city and examined for *Toxocara* species eggs. The parasite eggs were recovered from 4 (36.4%) out of 12 public parks. Among the samples studied, the number of *Toxocara* eggs recovered varied from 3 to 20 with a mean of 12 eggs per 200 g of soil samples.

The relationship between increasing the park size and contamination of soil sample is shown in Table 1. There was a significant difference between increasing the contamination rates and park size ($P < 0.05$). Table 2 shows the distribution of *Toxocara* spp. eggs in the soil samples of public parks due to park areas that were examined. The highest contamination rate was in public parks in the third region (42.5%) of 4 regions studied.

Toxocara eggs that were recovered from soil samples were developed into unembryonated and embryonated eggs. Overall, 184 *Toxocara* eggs were recovered, of which 14 (7.6%) were fully embryonated (Figure 1).

Discussion

In the current study, the environmental contamination with *Toxocara* eggs was investigated for the first time in Karaj city. The recovery of *Toxocara* eggs from soil samples depends on several important environmental factors including sunlight, temperature, humidity, air quality, the site of sampling, and the laboratory techniques employed.[7] *Toxocara* species eggs require a period of time under appropriate environmental conditions to become infectious to definitive and paratenic hosts.[8]

The results of the study showed that the soil contamination of public parks with *Toxocara* spp. eggs was 36.4% in Karaj, Iran. The prevalence of *Toxocara* spp. ova in soil samples from public parks or places in Iran varied from 2.3% in Qazvin up to 38.0% in Tehran and 63.3% in Khorram Abad.[5,9,10] These findings are similar

Table 1. Relationship Between Contamination of Soil Sample With *Toxocara* spp. Eggs and Park Size of Karaj, Iran

Condition	Soil Sample		*P* Value[a]
	Examined (No.)	Positive (%)	
Less than 5000 m^2	102	11.1	$P < 0.05$
More than 5000 m^2	98	25.3	
Total	200	36.4	

[a] Chi-square test.

Table 2. *Toxocara* spp. Eggs Recovered From Soil Samples of Public Parks From 4 Regions of Karaj, Iran, and Their Developmental Stage

Regions	Eggs Recovered (No.)	Eggs Developed (No.)	
		Unembryonated	Embryonated
1	18	16	2
2	36	32	4
3	85	78	7
4	45	44	1
Total	184	170	14

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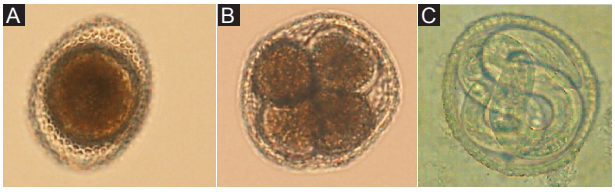

Figure 1. *Toxocara* spp. Eggs at Different Developmental Stages, Recovered From Soil Samples of Public Parks of Karaj, Iran. (A) One cell, (B) More than one cell (unembryonated), and (C) Embryonated.

to those reported by Cardillo et al in a historical review of toxocariasis in Argentina. Prevalence of *Toxocara* spp. in soil samples from different provinces showed an average of 10.0% (median of 12.1%), a minimum of 0.8% corresponding to the province of Santa Fe and a maximum of 35.1% corresponding to Patagonia provinces. The climatic condition of Patagonia is tempered to cold and appears to be similar to that reported in Karaj, with rainfall below 300 mm/y and an annual temperature of 8°C to 17.9°C.[7] The contamination rates of soil with *Toxocara* eggs in public parks and children playgrounds have been reported to be 5.71% in soil samples in Thailand, 38-53% in Poland, 53.3% in Brazil, 62.5% in Turkey, 64% in Italy, and 67% in Spain.[11-16] Some factors may have contributed to this inconsistency, such as climatic and environmental conditions.

In this investigation, soil contamination rate was related to the extent of park. The public parks more than 5000 m^2 were highly contaminated with *Toxocara* spp. eggs. It might be related to the defecation habits of dogs and cats on greater public parks. Zibaei et al reported widespread contamination of public parks with *Toxocara* eggs that had an area more than ten-thousand square meters.[5] The presence of many green areas in bigger parks may be the right places for defecation of dogs and cats.

We found *Toxocara* spp. eggs in 83% of soil samples with feces on the surface that shows relationship between the degree of contamination by *Toxocara* eggs and by feces. Dubana et al reported 51.4% of *Toxocara* ova recovered from the soil with feces in public places in Czech Republic.[17] It is important to note that contamination of soil with feces may increase the risk of infection by other helminth eggs or bacteria.

In this study, 7.6% of *Toxocara* eggs recovered from soil samples were embryonated. The report of survey by Uga et al showed that 85.8% of all *Toxocara* eggs that had been recovered from soil were embryonated,[18] whereas another study revealed that 42.1% of the soil samples were fully embryonated.[5] This difference was observed probably because of seasonal changes. The low percentage of development of eggs on August and September arose from high temperature. Moreover, it can be concluded that the differences in the results found in this work were directly related to the environmental conditions present in the studied regions. Generally, moisture is essential for encouraging the development and maintaining egg viability while temperature is responsible for the rate of embryonation. Regions with high thermal amplitude, low moisture, strong sunlight, and soils with little vegetation provide adverse circumstances for the viability of the eggs of *Toxocara* species. Nevertheless, in Argentina, the climate does not seem to interfere with the transmission of toxocariasis; since similar environmental prevalences have been found between the provinces of Patagonia and the provinces of the center and north of the country, with a notable difference in climatic amplitude. And studies on the persistence of soil contamination did not show significant differences depending on the months of the season.

To conclude, the results of the present study revealed the contamination of soil samples of public parks with *Toxocara* spp. eggs. On the basis of these data, the authors hope that increasing the hygienic control on dogs' and cats' defecation, to prevent environmental contamination, and community training programs could support the concept of responsible pet ownership.

Authors' Contributions

Study concept, design and study supervision: MZ; Sampling, processing and performing the conventional procedures: SB; Sampling: ARK; Advice provision: NC.

Ethical Approval

Not applicable.

Conflict of Interest Disclosures

The authors declare that they have no conflict of interests.

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