Original papers

Prevalence of *Toxocara* eggs in the soil of public parks of Khorramshahr city, southwest Iran

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ABSTRACT. Human toxocarosis is a chronic zoonotic disease, which creates various syndromes in humans such as visceral larva migrans, ocular larva migrans, and neurological toxocarosis. This disease is commonly contracted through ingestion of eggs from the *Toxocara* species. The eggs are transmitted to humans through contact with soil that contaminated with feces of infected animals by *Toxocara* species worm. This study, investigated the soil from various parks and public areas of Khorramshahr city southwest of Iran for *Toxocara* egg contamination. One-hundred and fifty soil samples were collected from 21 parks and green public areas from April to July 2018 and examined by the modified floatation method using saturated sucrose solution. The *Toxocara* eggs were isolated in 27 (18%) out of 150 samples collected. Based on Chi-square test, none of the factors: location of sampling (in city and the suburb), the presence of dogs and cats and the size of the sampling location; was not significant difference by soil contamination by *Toxocara* eggs. The presence of *Toxocara* eggs in Khorramshahr parks soil and green public areas is one of the main risk factors for catch the toxocarosis. This is important to develop preventive measures in the city and suburb areas to reduce the Environmental contaminations and thus reduce the incidence of disease in humans.

Keywords: zoonotic infectious diseases, Toxocara eggs, soil contamination, public health, soil pollutants

Introduction

Human toxocarosis is an important zoonotic infection that causes health problems in many parts of the world, especially in areas with a tropical climate [1,2]. The causative agents of the disease are *Toxocara canis* and *Toxocara cati* worms. The eggs of these worms are excreted through the stools of infected dogs and cats and lead to contamination of the soil in public areas [3]. Infections in human initiate by the contact with the infected dogs and cats, contaminated soil and consumption of contaminated vegetables. Once ingested, eggs hatch in the intestine and the larvae enter the circulatory system and transfer to other organs of the body and cause human toxocarosis (HT). In the human body, there are different manifestations of this disease such as visceral larva migrans (VLM), ocular larva migrans (OLM), neurological toxocarosis (NT) and covert toxocarosis (CT)[4]. Most of the cases are seen in children between the ages of 1 and 5 years old and most of them are asymptomatic. However, fever hypereosinophilia, liver symptoms, cerebral, pulmonary and ocular symptoms, and even mental illness have been linked to the disease. In some cases, there may not be any specific complications associated with the disease [5].

The reports of toxocarosis in different regions of Iran have been recently studied [6]. Parks and playgrounds were identified as the most important places for the transmission of this disease to humans, especially to children. There are several reports of dogs and cats infected with *Toxocara* in Iran [6]. As the infected stray dogs and cats enter the parks and public places easily, their stool can contaminate the soil and increase the risk of human toxocarosis. Many studies have been done on soil contamination of parks and public places with *Toxocara* eggs in Iran [6–19]. The aim of this study was to investigate and determine the presence of *Toxocara* spp. eggs in the soil of parks and green public areas in the city of Khorramshahr.

Materials and Methods

Study description and area. This research is a cross sectional study on soil contamination of parks and green public areas by Toxocara egg in the city of Khorramshahr southwestern Iran done between April to July 2018. The city of Khorramshahr (30°26'12.3"N, 48°11'00.8"E) is located at the end of the southwestern part of Iran and in the Khuzestan province, among the rivers of Karoon, Arvand and Bahmaneshir. The area of the city is 67.5 square kilometers and the average height is 5 meters above sea level, with a warm and humid climate. The soil of this city is alluvial with an alkaline pH. According to the latest national census (in 2016), Khorramshahr has 171 000 inhabitants. According to information received from the municipality, there are 20 parks in Khorramshahr. The total area of these parks is about 279200 m^2 .

Sample size. Using similar articles [17] and sample size formula, the number of samples for this research was calculated. For this purpose z, p and d were equal to 1.96, 0.06 and 0.04, respectively. As a result the minimum sample size obtained 135. That is for sure 150 sample was considered for this study. To calculate the sample number of each park, the ratio of the area of each park to the total of the parks was calculated and the number of samples was obtained from each park.

Recovery of soil. For the purpose of this study, the data and information regarding the parks, public areas and children's playgrounds were received from the municipality of Khorramshahr. A total of

150 soil samples were collected from 20 different locations from April to July 2018. One-hundred grams' soil was collected from the depth of 3 cm at each marked location, packed in zipper bags and transferred to the research laboratory of the Abadan Faculty of Medical Sciences.

Detection of Toxocara eggs. The collected samples were initially dried overnight at room temperature and the coarse particles were crushed, then filtered by multi-layer fabric gases and tap water. The filtered material of each sample centrifuged at 488 G for 5 minutes. The supernatant was removed and the sediment was separated from the tube using a vortex and simultaneously, the sucrose saturation solution (1.2 g/cm³) was added slowly to fill 80% of the tube. The tubes were centrifuged at 252 G for a further 20 minutes. Following this, the tubes were completely filled with sucrose solution and a Coverslip was placed on the tube and rested 1 hour. After this resting period, the coverslips were collected from the tube and placed on a slide and examined by optical microscope with a magnification of 100 and 400× (Lens 10 and 40) using a zigzag method.

The presence of dog and cat. The presence of dogs and cats was assessed in several ways: observation of dog and cat in the park and thier stool and footprints in the soil.

Statistical analysis. The sampling location (in the city and the suburb), the presence or absence of dogs and cats in the contaminated parks and observation of *Toxocara* eggs in the soil and also the contaminated parks and the size of them, were analyzed using SPSS 16 software and Chi-square test with a significant level of 0.05.

Results

The 150 soil specimens were collected from 21 different parks and green public areas. Of these 150 specimens, 27 (18.0%) sample were contaminated with *Toxocara* eggs. Another contamination was also noted such as: *Taenia* spp. eggs found in seven samples (4.6%), hookworms egg found in three samples (2%), *Trichuris* spp. egg noted in one sample (0.6%) and nematode larvae noted in 11 samples (7.3%).

The *Toxocara* eggs were observed in 11 sampling areas. In other words, 52.0% of the public parks and green areas in Khorramshahr city were contaminated with *Toxocara* eggs. It is worth noting that none of the *Toxocara* eggs identified were

A11		Positive (%)	Negative (%)	Total (%)	Dlus
All samples		27(18)	123(82)	150(100)	P-value
Location	In city	11(7)	60(40)	71(47)	0.449
	Suburb	16(11)	63(42)	79(53)	
Presence of dogs and cats	yes	23(15)	94(63)	117(78)	0.320
	No	4(3)	29(19)	33(22)	

Table 1. *Toxocara* eggs recovered from soil samples of parks and public areas of Khorramshahr city according to the location and presence of dog and cat

embryonated.

In statistical analysis between sampling location (in the city and the suburb) and observation of *Toxocara* eggs in soil, There was no significant difference between them (P = 0.32). There was also no significant difference between the presence or absence of dogs and cats in the contaminated parks with *Toxocara* eggs. (P = 0.449) (Table 1). Finally, there was no significant difference noted between the contaminated parks and the size of the areas. According to the Table 2, p-values obtained for 10000, 5000 and 2000 m² were 0.621, 0.983 and 0.311, respectively.

Discussion

The isolation process of *Toxocara* eggs from soil can be achieved by different sized sieves [12,14,18,20,21]. Collecting *Toxocara* spp. eggs using a sieve in this process increases the amount of water and is time consuming, therefore in this study, a multi-layer fabric mesh was used instead of a sieve. Coarse soil, rocks, branch, leaves and roots of plants did not pass through multilayer fabric mesh. On the other hand, fine particles of soil that contain eggs and nematodes larvae crossed the filter faster and in a shorter period of time. The advantages of this method were a reduction in testing time and a reduction in the amount of water and containers used to separate the particles and allow more organic particles (such as worm larvae) to enter floatation stage.

In the current study, the soil contamination rate of *Toxocara* eggs was 18.0% in Khorramshahr city. The rate of contamination in various cities in Iran was as follows: 36.4% in Tabriz and Karaj [7,8], 29.2% in Abadan [9], 28.6% in Isfahan [10], 22.2% in Khorram Abad [11], 13.5% in Kermanshah [12], 10.3% in Khaf [13], 10% in Tehran [14], 7.8% in Urmia [15], 7.7% in Mashhad [13], 7% in Ardabil [16], 6.3% in Shiraz [17], 5.8% in Qazvin [18] and 4% in Jahrom [19]. Several studies in various countries have recently reported a vareity of soil contamination rate of *Toxocara* eggs including 63.2% in Portugal [22], 24.5 in Iraq [23], 23.3% in Russia [24], 17.2 in Greece [25], 14.9% Poland [26] and 12.5% in Egypt [27].

A reason for the level of Toxocara eggs in the

Table 2. The relationship between the size of the parks and identified Toxocara eggs

Parks size	number of	f samples		P-value	
	examined	positive	— Positive rate (%)		
More than 10000 m ²	118	21	17.8	0.621	
less than 10000 m ²	32	6	18.8		
More than 5000 m ²	122	22	18.0	0.983	
less than 5000 m ²	28	5	17.9		
More than 2000 m ²	134	26	19.4	0.211	
less than 2000 m ²	16	1	6.2	0.311	

soil of Khorramshahr city may be related to the hot weather in this area. Some studies have shown that the number of *Toxocara* egg in the warm seasons is lower than the cold season [8,23,28,29]. All of the observed Toxocara eggs in this study were unembryonated. However, in a similar study in the city of Abadan (10 km away Khorramshahr), some of isolated Toxocara eggs were embryonated [9]. Considering that, the sampling in the city of Abadan was carried out during the cold season whilst this study was completed during the warm season. Also in other study in Germany, the total observed eggs and the embryonated eggs in cold season was more than the warm season [29]. Recent studies have shown that increasing temperature and lowering moisture would reduce Toxocara egg development [30,31]. Therefore, it can be concluded that the weather conditions of summer in the area caused to prevent the Toxocara egg to develop embryos.

If the embryonated eggs of *Toxocara canis* and *Toxocara cati* enter in the human body, they are more likely to cause human toxocarosis. This disease develops in various forms such as visceral larva migrans, ocular larva migrans and neurological toxocarosis [6]. There are various reports of human toxocarosis in owners of domestic cats and dogs (20.43%) [32], in the people with eosinophilia (67%, 19% and 11.7%) [33–35], in school and preschool children (25.6%, 29.46%, 37.3% and 6.3%) [36–39], and in pregnant women and newborns (7.4% and 20%) [40,41].

Infected dogs and cats can easily disseminate *Toxocara* eggs in to the environment [42–46]. Therefore, it is acceptable to believe there may be a correlation between the presence of cats and dogs in the area and the rates of *Toxocara* eggs [9]. However, in this study there was no sensible relationship identified between the above factors (Table 1). In addition, according to the Table 2, there was no significant relationship between the size of parks and *Toxocara* eggs observation. This relationship was reported to be significant in other studies [7,47].

Contaminated soil by *Toxocara* spp. eggs may increase the prevalence of toxocarosis for human especially in children since their high exposure with contaminated soil. To reduce soil contamination preventative measures should be developed for urban park structures, the preparation of wasteland in urban areas, the management of wastes and the control of stray dogs and cats in the city and suburb. Changes in these areas can lead to a significant reduction in soil contamination with *Toxocara* and reduce the risk of transmission of pathogens to humans.

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Competing interests. The authors declare that they have no competing interests.

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