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Environmental soil contamination by *Toxocara* species eggs in public places of Ilam, Iran

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Abstract

Introduction. The purpose of the study is to assess environmental contamination by *Toxocara* species eggs in public places in the city of Ilam, Ilam Province, southwest Iran

Materials and method. Between September 2018 and March 2019, 130 soil samples were collected from public places of 5 district municipalities of Ilam, southwest Iran. Soil samples were examined by microscopy following flotation method by sodium nitrate.

Results. Soil analysis showed that 5.88% of the soils stored, 52.54% from gardens, 29.42% from rubbish, and 11.72% from green spaces were contaminated with *Toxocara* spp. eggs. In total, 13.08 % of soil samples (17/130) were positive for *Toxocara* eggs ($P > 0.05$).

Conclusions. The findings revealed that care should be taken when using soil from gardens, green spaces and rubbish, and also should be seriously considered because of the potential issues of toxocariasis and also the risk to the public.

Key words

Toxocara eggs, soil, contamination, public places, Iran

INTRODUCTION

Toxocariasis is a zoonotic disease caused mainly by *Toxocara canis (T. canis)* and *Toxocara cati (T. cati)*, intestinal nematodes of dogs and cats, respectively. Transmission to humans occurs by ingestion of embryonated *Toxocara* spp. eggs in the soil, water and vegetables, or through contaminated hands and fomites, and/or eating the meat of paratenic hosts containing encapsulated larvae [1–5]. Most infections do not have any clinical symptoms, athough *Toxocara* larvae are released within the different tissues and organs and may cause dangerous clinical syndromes, including weight loss, fever with a cough and shortness of breath, generalized lymphadenopathy and hepatomegaly [6–8]. The diagnosis of human toxocariasis is mainly based on clinical symptoms, epidemiological and laboratory data, which include imagining features, peripheral blood eosinophil, total IgE level, and serological findings using Enzyme-linked immunosorbentassay (ELISA) and Western blotting (WB) [9]. In big cities, the soils of the public places, such as green spaces, can become an important source of parasites contamination because domestic and stray dogs and cats have the highest mobility in these areas. In Iran, previous studies have shown that the prevalence of *Toxocara* species eggs in soil samples from the public places varied from 2.3% in Qazvin to 63.3% in Khorram Abad [4].

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OBJECTIVE

Given the abundance of stray dogs and cats, and that toxocariasis could be a dangerous clinical complication in humans, the aim of the current research was to investigate the prevalence of *Toxocara* eggs in soils from the public places of 5 district municipalities in Ilam, southwest Iran.

MATERIALS AND METHOD

Study area. Ilam (33 38′14″ N and 46 25′21″ E) is the capital of Ilam province, Iran (Fig. 1). The climate of the city is moderate and the temperatures vary between -13.6 – 41.2 °C. The study was conducted between September 2018 and March 2019. The public places of 5 district municipalities in the city of Ilam were selected for sampling, regarding the size of the area and the mobility of the animals.

Samples collection. The study was performed on 130 soil samples collected in the public places of 5 municipality districts in the city of Ilam in order to recover *Toxocara* spp. eggs. A soil

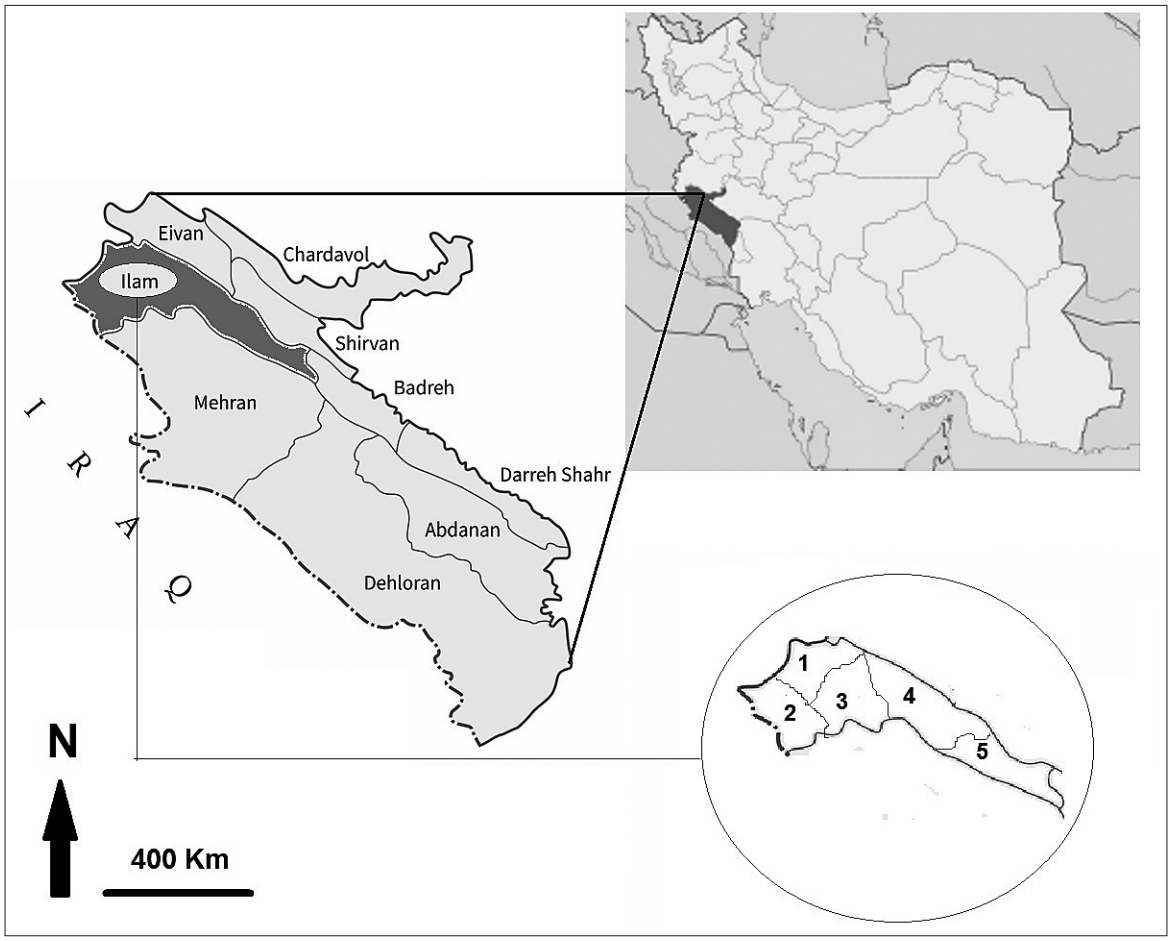

Figure 1. Regions in Ilam district from which soil samples were examined for *Toxocara* eggs

sample of 150–200 g per 4 sq m area was collected at a depth of 3 cm. This resulted in 19–28 samples per specified area. The soil samples from the same site were thoroughly mixed and stored in sealed and labeled polythene bags, and taken to a laboratory for recovery of *Toxocara* eggs.

Detection of eggs. For eggs detection, soil samples were examined according to the method described by Zibaei et al. [2]. The tests were carried out in triplicate. Briefly, soil samples (dried at room temperature and sifted through a 150 µm mesh sieve) were placed in flat-bottomed flasks and 5% NaOH (Merck, Germany) was added to separate the eggs from the soil particles. The contents of the flasks were stirred and allowed to settle for 1 h, before being shaken for 20 min at 100 rpm. After this vigorous mixing, they were placed in test tubes and centrifuged for 5 min at 1,500 rpm, following which the supernatants were removed from the test tubes and replaced with tap water.

The mixture was centrifuged again for 5 min at 1,500 rpm. After spinning, the H_2O was discarded, a saturated $NaNO_3$ solution was added to the pellet and centrifuged for 5 min at 1,500 rpm. The tubes were transferred to tripods and saturated $NaNO_3$ solution was added with a pipette to form a convex meniscus. A coverslip was placed over each sample, and after 30 min they were placed on a microscope slide. The preparations were evaluated at magnifications of x400 and x1,000 under a light microscope for the presence of *Toxocara* spp. eggs. *Toxocara* species eggs were identified only at the genus level due to remarkable morphological similarities.

Statistical analysis. All epidemiological and laboratory data was tested for their association with toxocariasis. Chi-square test and Fisher's exact test were used for categorical data. A *P*-value less than 0.05 was considered statistically significant.

RESULTS

Parasite eggs were found in 13.08% (17/130) of samples. Among the samples studied, the number of *Toxocara* eggs recovered varied from 1–9 with a mean of 4 eggs per 100 g of soil samples.

The relationship between the location and contamination of soil sample is shown in Table 1. There was no significant

Table 1. Recovery of eggs of *Toxocara* spp. from soil samples at each site

Soils	Soil samples (No.[¥])					Total No. (%)	*Toxocara* spp. eggs No. (%)	P-value[£]
	Areas							
	Mustafa Khomeini	Talaghani	Imam Khomeini	Kosar	Ghaem			
Garden	10	12	18	7	3	50 (38.46)	9 (52.94)	0.435
Stored	6	1	5	2	1	15 (11.54)	1 (5.88)	
Green space	5	4	10	2	2	23 (17.70)	2 (11.76)	
Rubbish	9	10	15	5	3	42 (32.30)	5 (29.42)	

[¥]Number
[£]No statistically significant difference in prevalence of the parasite among soil samples ($P > 0.05$)

Table 2. Prevalence of *Toxocara* spp. eggs in soils of different areas in Ilam

Areas	No.[¥] of Samples	*Toxocara* spp. eggs No. (%)	Prevalence Ratio	*P*-value[£]
Mustafa Khomeini	30	4 (23.53)	1	0.857
Talaghani	27	3 (17.65)	0.9	
Imam Khomeini	48	6 (35.29)	0.9	
Kosar	16	3 (17.65)	1.5	
Ghaem	9	1 (5.88)	0.8	
Total	130 (100)	17 (100)		

[¥] Number
[£] No statistically significant difference in prevalence of the parasite among regions ($P > 0.05$)

difference between the contamination rates and sampling site ($P > 0.05$). Table 2 shows the distribution of *Toxocara* species eggs in the soil samples of the specified sites regarding the soil areas examined. The highest rate of contamination was 32.29% in the Imam Khomeini area, compared to other areas investigated.

Toxocara eggs recovered from soil samples were developed into unembryonated and embryonated eggs. Overall, 17 *Toxocara* eggs were recovered, of which 10 (85.82%) were fully embryonated.

DISCUSSION

Infectious diseases caused by soil-transmitted helminths (STHs) are important diseases of humans which affect about one-third of the world's population. From the public health perspective, soil examination is an effective substitute for faecal examination in epidemiological surveys of STH infection [10–16]. Earthworms may be play a significant role in transmitting the eggs in soils [17], as well as flies [18, 19]. Soil contaminated by eggs of the *Toxocara* species have been shown to be one of the main infection sources of toxocariasis [20]. The prevalence of *Toxocara* spp. eggs has been found in in soil samples in Spain (64–67%) [20, 21], Brazil (62%) [22], China (55.7–77.9%) [23], Iraq (50.0%) [24], and Poland (4.5–50.0%) [25, 26]. In opposition to original thought, it has been shown that toxocariasis is highly prevalent in human life, and prevention of the transmission of the parasite to humans is necessary because of the different clinical (mild to severe) clinical complications.

The presented study is the first to identify and estimate soil contamination of *Toxocara* eggs in Ilam. However, studies have been conducted on the prevalence of human toxocariasis among different individuals, including children, pregnant women and diabetic patients in this region [27, 28]. The current study describes soil contamination with *Toxocara* spp. eggs from public places in 5 municipalities districts of the city of Ilam in Iran. The results of the study show that the contamination of soil samples with *Toxocara* spp. eggs was 13.08%. The prevalence of *Toxocara* spp. ova in soil samples from public parks and other sites in Iran varied from 16.0% in Shiraz [29] up to 18.0% in Kermanshah [30], Isfahan 28.6% [31], Abadan 29.2% [32], Tehran 38.0% [33], Ahvaz 38% [34], and 63.3% in Khorram Abad [35].

The sampling period occurred in different weather conditions over a year. However, studies in the cities of Urmia, Tabriz, Ardabil, Amol and Mashhad in Iran showed a lower prevalence than the present study (3.2–7.7%) [3, 36]. For development of *Toxocara* eggs in the soil, oxygen and humidity are required; however, the observed prevalence could also be significantly different if these environmental conditions are present. In a meta-analysis study on the prevalence of *Toxocara* species eggs in soil samples from 200-o 2016 in Iran, there was no significant correlation between *Toxocara* egg (16.0%) and soil sample size [37]. As in the current study, soil contaminations by *Toxocara* spp eggs were found mostly in gardens [37]. In a similar study, the contamination rate of soil with *Toxocara* eggs in gardens has been reported in Mexico City [38].

CONCLUSIONS

The results of the present study show that the some public places in Ilam were contaminated with the faeces of animals and *Toxocara* eggs, indicating that control measures, as well as the education of the public, is needed for protection from zoonoses diseases.

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