Contamination of soil with eggs of geohelminths in recreational areas in the Lublin region of Poland

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Abstract
Recreational areas cover parks, bicycle paths, lawns, urban squares, sports complexes, holiday camp areas, playgrounds for children, beaches, and even spontaneously used green field open spaces. While using recreational areas, people take with them accompanying animals (dogs, cats). These animals constitute the main source and reservoir of many dangerous zoonoses, including parasitoses caused by roundworms of the genus Toxocara and Trichuris. The objective of the presented study was determination of the level of contamination of soil with parasites’ developmental forms (eggs of Toxocara spp., Ascaris spp., Trichuris spp and Ancylostoma) in selected recreational areas in the Lublin Region of southeastern Poland. The material for the study was collected from beaches in recreational areas located around the Biale Lake near Wlodawa, the water reservoirs in Krasnobrod and Janów Lubelski. The studies were carried out from May-October 2010 during which time a total number of 215 samples of sand were collected. Eggs of Ascaris spp. and Trichuris spp. were detected based on the Polish Standard PN-Z-19000-4/2001. Beaches on the Biale Lake were the most contaminated. The eggs of intestinal parasites were isolated from 6 per 15 samples examined, which is 40%. In one sample, the presence of the eggs of 3 types of parasites were observed, while the eggs of Ancylostoma were not found in any of the samples. Attention should also be paid to the possibility of contamination of recreational areas with the eggs of intestinal parasites by wild animals. Recently in Poland, as well as in many other European countries, the phenomenon of synantropization of wild living animals has been observed, which most frequently concerned wild boar (Sus scrofa) and red fox (Vulpes vulpes).

Key words
contamination, soil, recreational area, eggs of geohelminths

While using recreational areas, people take with them accompanying animals (dogs, cats). These animals are the main source and reservoir of many dangerous zoonoses, including parasitoses caused by roundworms of the genus Toxocara and Trichuris. Attention should also be paid to the possibility of contamination of recreational areas with the eggs of intestinal parasites by wild living animals. Recently in Poland, as well as in many other European countries, the phenomenon of synantropization of wild living animals has been observed.

Species of large and medium-size mammals increasingly more often enter urbanized areas. Most frequently this concerns wild boars (Sus scrofa) and red foxes (Vulpes vulpes). The presence of wild living animals in areas inhabited by humans, apart from problems of a psychological nature (fear of attack, fear of collision), create a health risk for humans and their accompanying animals [1].

The eggs of roundworms Toxocara spp are the main cause of contamination of soil and sand in recreational areas in Poland. Worldwide, the infections in dogs range from 3–80%, and nearly 100% of puppies are born with this parasite. Toxocariasis is a serious clinical problem; in humans, it is associated with the occurrence of symptoms defined as visceral larva migrans – VLM or ocular larva migrans – OLM syndromes. This disease is most frequently observed in children [2].

The subsequent cause of contamination of recreational areas is the whipworm Trichuris vulpis. This parasite occurs in dogs, foxes and other carnivora, and is typical of older...
animals. In Poland, dog whipworm infection is found in 3-10% of the population, especially among children living in rural areas [3].

In addition, cases of infection with other dog and cat intestinal parasites non-specific for humans, such as Dipylidium caninum and Ancylostoma spp, are also relatively often noted [2].

Hookworms (Ancylostoma caninum), apart from roundworms, are the most frequent dogs parasites. In humans, these parasites may cause the diseases called cutaneous larva migrans – CLM syndrome, induced by the penetration of invasive larvae through the skin of the host. In the summer of 1994, in Berlin, Germany, scientists observed that an increase in temperature by 6°C resulted in a growing incidence of cutaneous larva migrans (CLM), a skin disease in humans caused by Ancylostoma caninum, a nematode parasite that infects dogs: “The study conducted of the increasing local temperatures and global warming may give rise to the emergence of CLM due to the ubiquitous presence of these zoonotic hookworms”[4].

The dog tapeworm (Dipylidium caninum) is a common parasite of cats and foxes as well as dogs, and parasitize in the small intestine. The extent of invasion depends on the availability of intermediate hosts (fleas, lice). Dipylidiasis in humans usually takes an asymptomatic course; in children under the age of 8 years it causes indigestion, loss of appetite, stomach pain, and even vomiting.

Studies of contamination of recreational areas with the eggs of goehelminths have been conducted in various regions of Poland: Wrocław, Łębork, Katowice, Elbląg Gdańsk, Poznań, Kraków and Warsaw. The sites of the study were primarily sand pits, playgrounds for children, and parks [2, 5, 6, 7, 8, 9, 10, 11, 12]. In the Lublin Region, investigation of the contamination of recreational areas (sand pits, playgrounds for children) with the eggs of the parasites were conducted by Gündlach [13], and Rzymowska [14].

**MATERIALS AND METHODS**

**Collection of samples.** Material for the study (sand) was collected from beaches located in recreational areas in the Lublin Region, an area located in southeastern Poland. The study covered commonly available beaches located on the Biała Lake near Włodawa, and the water reservoirs in Krasnobród and Janów Lubelski. The beaches are administered by Commune Offices in Włodawa, Krasnobród, and Janów Lubelski, respectively. The studies were carried out from May - October 2010, during which a total number of 215 sand samples were collected. The samples were collected from 5 sites on a beach. Collective samples were used for parasitological studies prepared from 5 component samples, and 43 averaged sand samples were examined: 15 from the beaches on Białe Lake, 16 from beaches by the water reservoir in Krasnobród, and 12 from beaches by the water reservoir in Janów Lubelski. The samples were collected from the surface layer of sand, at a depth up to 3 cm, approximately 500 g each, into plastic bags.

**Detection of eggs.** Geohelminths’ eggs were isolated from 100 g sand samples with the use of the standardized flotation method according to the Polish Standard [15], and the flotation method by Quinn [16].

After delivery to the laboratory, the samples were dried at room temperature for 2-3 days. Dried samples were mixed and sieved through a 2 mm mesh to remove larger contaminants. Eggs of Ascaris spp. and Trichuris spp. were detected based on the Polish Standard PN-Z-19000-4/2001. 100 g soil samples were transferred to volumetric flasks containing 100 ml of 5% KOH or NaOH mixed and left for 1 h. After that time, the soil samples were vortexed for 10 min. The suspensions were transferred to centrifuge tubes, then centrifuged at 1,600 g for 2 min, after which the supernatant free of eggs was decanted. Saturated flotation solution NaNO₃ was added, the tubes were vortexed again, then centrifuged 3 times at 1,600 g for 2 min. After every centrifugation, 1 ml of the cover slip was transferred to a flask with 4-5 ml of water. These samples were filtrated with the use of a membrane vacuum pump. After that, the filter paper was transferred to a glass slide and examined under x10 magnification, and the number of Ascaris spp. and Trichuris spp. were counted.

Eggs of Toxocara spp. were detected with the use of the flotation method by Quinn [16]. Dried soil samples were sieved through a mesh. 100 g soil samples were transferred to volumetric flasks containing 100 ml of 0.0025% Tween 80, and vortexed for 60 sec. The suspensions were transferred to centrifuge tubes, then centrifuged at 2,600 g for 10 min., after which the supernatant free of eggs was decanted. Saturated flotation solution NaCl was added and the tubes were vortexed again, centrifuged at 2,600 g for 10 min., after which the solution was added to form a meniscus and a cover slip was overlaid. After 30 min., the cover slip was transferred to a glass slide, and examined under x 10 magnification and the number of Toxocara spp. eggs counted. No attempt was made to differentiate between eggs of T. catti and T. canis, A. suum and A. lumbricoides, T. trichiura and T. suis and T. vulpis. In each sample, the number of eggs and their genus were determined by microscope.

Eggs of Ancylostoma spp. were searched for by the flotation method according to Quinn et al. [16].

**RESULTS**

The studies conducted in 2010 showed a low level of contamination with the eggs of goehelminths on beaches in recreational areas in the Lublin Region. The total level of contamination was 18.6% (Tab. 1). Beaches on Białe Lake were the most contaminated: of the total number of 15 samples examined, the eggs of intestinal parasites were isolated from 6 samples (40%); in one sample, the presence of the eggs of 3 types of parasites was noted.

<table>
<thead>
<tr>
<th>No.</th>
<th>Site of study</th>
<th>No. of samples in which eggs were detected</th>
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<tbody>
<tr>
<td>1</td>
<td>Biała Lake</td>
<td>2</td>
</tr>
<tr>
<td>2</td>
<td>Water reservoir in Janów Lubelski</td>
<td>2</td>
</tr>
<tr>
<td>3</td>
<td>Water reservoir in Krasnobród</td>
<td>0</td>
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<td>Total</td>
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The eggs of *Toxocara* spp. were found in 16.6 % of sand samples from among 12 samples collected from beaches by the water reservoir in Janów Lubelski. In sand from beaches by the water reservoir in Krasnabrod, no eggs were detected of intestinal parasites. The eggs of *Ancylostoma* were not found in any of the samples.

**DISCUSSION**

In Poland, a constant increase in the size of the population of cats and dogs which become infested by parasites results in an increased contamination of the human natural environment (soil) with invasive forms of parasites. The level of contamination of the natural environment may be evidenced by the fact that a single female roundworm may produce about 200,000 eggs daily. Considering the presence of the thick egg capsules of *Toxocara*, they may survive in soil even up to 10 years, despite unfavourable environmental conditions, and do not lose their invasiveness.

The contamination of soil with various species of parasites may constitute a potential source of parasitic diseases in humans. To-date in Poland, the greatest attention has been paid to studies of parasitological situation in urban areas. The examinations covered sand pits, playgrounds for children, urban recreational areas, lawns, squares and yards in various cities, such as Elbląg, Kraków, Lublin, Poznań, Puławy, Warsaw and Wrocław. The parasites most frequently found in the above-mentioned areas were roundworms of *Toxocara* species. The observed level of contamination of yards with the eggs of *Toxocara* spp. was as follows: Warsaw – 11.8%, Poznań – 27%, and in Kraków as much as 61.9% [9, 10, 12]. A comparable contamination with the eggs of roundworms of *Toxocara* species was noted for squares and lawns located in Poznań (8%), Kraków (28%), Warsaw (26.1%), Lublin and Puławy (22%) [9, 10, 12, 13].

Sand pits were also considerably contaminated with the eggs of *Toxocara*, and were clearly dominant in sand samples from the areas of Łębork (28%), Warszawa (5.4%), Kraków (11.5%), as well as Lublin and Puławy (31.6%). Recent studies carried out in 2010 in the recreational areas and sand pits in Lublin confirmed high contamination of these places on the level of 23.3 % [5, 10, 12, 13, 14].

The eggs of *Toxocara* spp. occurred considerably more rarely in rural than urban areas. In the villages of Grodkowice and Łązkowice, in close proximity to Kraków, the level of soil contamination was 16% [10], in the Poznań Region – 2% [9], and in villages in the Lublin Region – 35.5% [13]. This may be associated with a lower population density of cats and dogs in the rural areas, and the tradition to chain the dogs in backyards. The latest studies conducted in the Łódź area in 2011 have confirmed a high prevalence of the eggs of *Toxocara* spp. in yards – from 20–100% [17].

There are few reports concerning the helminthological contamination of beaches in Poland. Studies of the largest beaches of the Olsztyn lakes conducted in 2009 (Lakes Ukiel and Kortowskie) indicated that they create risk for people who spend leisure time there. The eggs of *Toxocara* spp. were present in 7.5% of the total number of samples examined [18].

Studies concerning the contamination of the environment with the eggs of intestinal parasites are conducted worldwide. The monitoring covers parks, playgrounds, school areas, municipal beaches, centers and suburbs of large cities, as well as farms, recreational centres, private properties, and areas around apartment building complexes. It is estimated that the contamination of soil with *Toxocara* eggs worldwide ranges from 1%–92% [19].

Eggs of *Toxocara* spp. were found in 20.4% of parks in Prague, Czech Republic, in 11.9% of sand pits and 45% of household gardens [20]. In the city of Shiraz in Iran, the level of contamination of municipal squares and playgrounds for children with the eggs of helminths was 21.4%, including 12.4% of contamination with the eggs of *Toxocara* spp. [21]. In Khorram Abad, also in Iran, 63.3% of municipal squares were contaminated with the eggs of *Toxocara* spp. [22]. Studies conducted in Turkey during the period from July 2007 - June 2008 confirmed that the contamination of unfenced public parks was approx. 64%, whereas in fenced parks where cats and dogs were not allowed, no contamination was observed [25]. Studies carried out in recreational areas in the Lublin region showed a low degree contamination of these areas with geohelminths’ eggs. The eggs of *Toxocara* spp. were detected only on the beaches of 2 lakes. The Białe Lake is the largest lake in the Lublin Region, with the highest intensity of tourism during the summer season. The water reservoir in Janów Lubelski is situated within the administrative borders of the city, and is also often visited by the inhabitants and holidaymakers, often accompanied by dogs. While considering the sources of toxocariasis in humans, cats must not be forgotten as potential disseminators of the eggs (*Toxocara catti*). Disseminators of *Toxocara* eggs are also foxes, the population of which in Poland increased from over 50,000 in 1990 to 220,000 in 2006 [23]. The threat is created especially by foxes inhabiting urbanized areas. The results of epidemiological studies and an increasing number of annually diagnosed cases of toxocariasis indicate the occurrence of a considerable risk of this disease in Poland, and confirm the opinions of specialists that this is the most prevalent parasitosis [24].

The results obtained in the presented study confirm the observations by other researchers and necessitate the following preventive measures:

1. The need for regular deworming of dogs and cats should be popularized.
2. The protection of beaches against contamination with geohelminths’ eggs is important.
3. The contamination of recreational areas require the undertaking of educational actions by the authorities of cities and communes to make society aware of the risk.

**REFERENCES**

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