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Risk factors associated with dog endoparasites infection spread in East Slovak Lowland

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Summary

This study aimed to determine the prevalence of endoparasites in dog feces from public places, investigate the occurrence of endoparasites in soil, and identify potential risk factors associated with the dog endoparasites infection spread in East Slovak Lowland - the region near the EU border with Ukraine. In a one-year study, a total of 803 dog fecal samples and 148 soil samples from public places were examined for the presence of endoparasite developmental stages. In general, 43.59 % of dog excrements were positive. Six different species of intestinal parasites were detected: *Toxocara canis* (22.65 %), *Trichuris vulpis* (14.07 %), family Ancylostomatidae (10.09 %), *Capillaria aerophilla* (5.60 %), *Ascaris* spp. (1.49 %). Altogether, 52.03 % of soil samples were positive for at least one endoparasites species. The occurrence of parasitic eggs in the soil was as follows: *Ascaris* spp. (37.16 %), *Toxocara* spp. (29.05 %), *Trichuris* spp. (29.05 %) %), family Ancylostomatidae (2.03 %) and *Toxascaris leonina* (2.03 %). The occurrence of endoparasites between the towns and villages varied significantly. The distribution of endoparasites in dogs and soil was affected by the population density, the minority people living in the studied territory, the number of people living in poor hygienic conditions with limited access to drinking water and the usage of sewerage. Persisting endoparasitic contamination of the environment in East Slovak Lowland forms a reservoir with zoonotic potential representing public and environmental health problems.

Keywords: public places; dogs; parasites; anthropogenic risk conditions; environment

Introduction

Public places (parks, playgrounds, sandpits, streets, sidewalks, and roadsides) play an important role in pet and human population welfare, where frequent traffic is observed. For this reason, many dog feces are present in various public places, which are of public health importance, and are of medical and veterinary concern. Papajová *et al.* (2014) stated that up to 70 % of dog feces in the Slovakian territory are found on lawns with dwelling houses nearby. A similar situation is also in other countries (Mizgajska, 2001; Rinaldi *et al.,* 2006; Dubná *et al*., 2007; Cinquepalmi *et al.,* 2013; Neves *et al.,* 2014; Ramos *et al.,* 2022). Batista *et al.* (2019) reported that an increase in soil contamination by helminth eggs correlates with the rise of the human and pet population, especially in urban areas where owners don't collect dog waste from public areas. It is important to note that while moving the lawns several times per year, the feces with endoparasites developmental stages are scattered around and thus contaminate new larger areas. (Papajová *et al.,* 2008; Milkovic *et al.,* 2009; Traversa *et al*., 2014; Sprenger *et al.,* 2014; Rubel *et al*., 2019). Dogs are the most fre-

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quently infected by protozoa *Giardia duodenalis, Cystoisospora* spp.; helminths *Toxocara* spp., *Trichuris vulpis, Ancylostoma* spp. and *Dipylidium caninum* (Mircean *et al.,* 2017; Saldanha-Elias *et al.,* 2019; Regidor-Cerrillo *et al.,* 2020; Šmigová *et al.*, 2021) of which many gastrointestinal parasites can also cause human infection (Souza *et al.,* 2023).

Human intestinal parasites are known to occur mainly where a lot of people are concentrated in a limited space and without access to drinking water and sewage system. In Slovakia, many such locations are located in the Košice region, including the East Slovak Lowland area (Ravasz, 2020). This area is ethnically mixed. Marginalized groups of inhabitants mostly live in poverty and have close contact with animals, which is associated with a lower hygiene standard and a large amount of environmental waste. Many studies have confirmed that in dogs living in the rural ecosystem and settlements, the incidence of parasites in Slovakia is higher than in the urban ecosystem due to lower hygiene standards and the higher presence of stray dogs. Jarošová *et al.* (2021) confirmed the high prevalence of endoparasites (up to 66.70 %) in stray dogs from Roma settlements. The most often diagnosed were eggs from the family Ancylostomatidae, eggs of *Toxocara canis*, *Taenia* spp. and *T. vulpis*. Papajová and Šoltys (2020) reported the highest prevalence of endoparasites in dogs from marginalized settlements (80.59 %), which was followed by dogs living in the countryside (40.09 %), and the lowest prevalence was found in dogs from the cities (25.45 %). Until now, little attention has been paid to the occurrence of intestinal parasitic diseases in the area of East Slovak Lowland. This region has gained importance, especially last year, because it is near the EU border with Ukraine and the strategic location of bigger cities nearby. There may be increased risk of spreding of endoparasites associated with migration. Because migrants arrive in this region regularly with their own pets which defecate into public places and it is possible, if the pets are infected, that they are contributing to endoparasites spread in public places. It is assumed that all pets transported between countries without proper veterinary control are potential carriers of endoparasites and other diseases.

The present study aimed to determine the prevalence of endoparasites in dog feces from public places, investigate the occurrence of endoparasites in soil, and identify potential risk factors associated with the dog endoparasites infection spread in East Slovak Lowland.

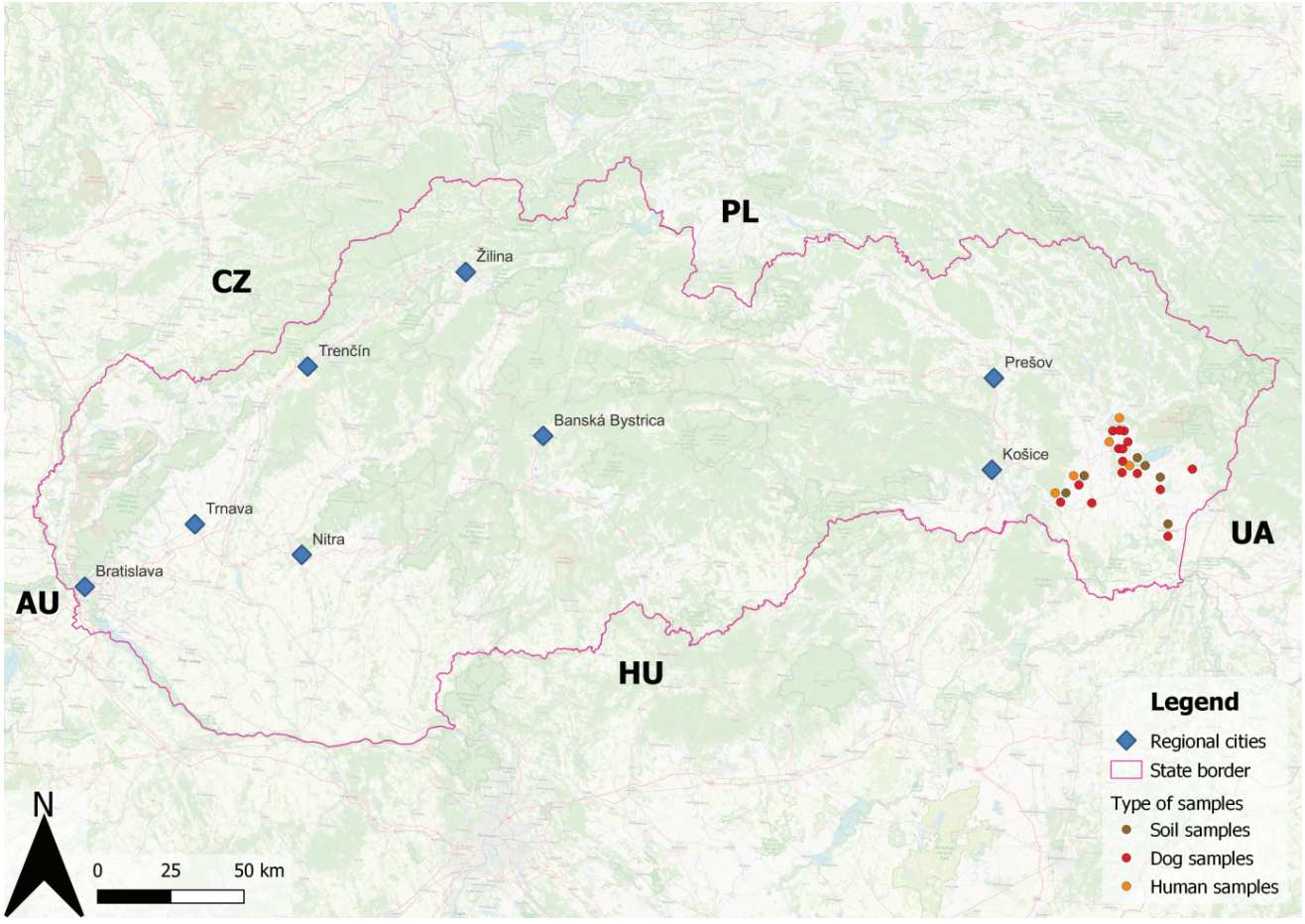

Fig. 1. The location of sample collection sites.

Material and Methods

Characterization of studied localities

The study was performed in 5 towns, 7 villages, and 4 segregated settlements located in 3 districts (Trebišov, Michalovce and Sobrance) on the East Slovak Lowland. The East Slovak Lowland belongs to the geomorphological area of the Great Danube Basin territory located in the southern part of eastern Slovakia (Fig. 1). Climatically, the East Slovak Lowland ranks among the warm regions where the average annual highest temperatures are 27°C and the lowest temperatures range between -2 and -3°C. The average annual rainfall is 650 to 700 mm (https://www.geology.sk/geoinfoportal/mapovy-portal/geologicke-mapy). According to the 2021 Population and Housing Census in Slovakia (https://www.scitanie.sk/), the studied region is inhabited predominantly by the Slovak majority group (from 65.50 % to 87.88 %). The marginalized Roma population represents the third largest population group in this region (from 1.83 % to 5.50 %). The studied localities differ by hygiene standards, sanitary conditions, and socio-economic status of dog owners. Data analyzing drinking water accessibility, sewerages system availability, and sumps in all studied localities were obtained from Slovakia's 2021 Population and Housing Census (https://www.scitanie.sk/) and about marginalized communities from the Ravasz (2020).

Dog's excrement and soil examination

Dog excrement and soil samples were collected randomly from November 2021 to December 2022. All samples were stored without preservation at 4°C and immediately transported to the laboratory at the Institute of Parasitology of the Slovak Academy of Sciences in Košice for further parasitological examination. All analyses were performed within 24 – 48 h.

Totally 803 dog excrements were collected from around the dwellings and public places and examined for the presence of endoparasite developmental stages.

Coprological diagnostics were done by a flotation method with Shaeter's flotation solution of sucrose with a specific gravity of 1.27 $g.ml^{-1}$ and the Faust flotation solution with a specific gravity of 1.18 $g.ml^{-1}$.

Totally 148 soil samples were collected from public places and near human settlements to investigate the presence of parasites in the environment. Multiple soil samples of approximately 300

Table 1. Parasitological evaluation of dog's excrement from public places.

		N	P	OR	95% CI
Overall	Villages	137	32	0.36	0.23 – 0.55
	Towns*	624	284		1.79 – 4.19
	Settlements**	42	34	5.98	2.73 – 13.10
Toxocara canis	Villages	137	14	0.37	0.20 – 0.66
	Towns*	624	146		1.49 – 4.80
	Settlements**	42	22	4.13	2.20 – 7.75
f. Ancylostomatidae	Villages	137	5	0.27	0.10 – 0.68
	Towns*	624	76		1.45 – 9.23
	Settlements**	42	0	0	–
Toxascaris leonina	Villages	137	1	0.20	0.02 – 1.50
	Towns*	624	2		0.66 – 37.19
	Settlements**	42	8	7.54	3.14 – 18.10
Trichuris vulpis	Villages	137	12	0.60	0.31 – 1.13
	Towns*	624	86		0,88 – 3,14
	Settlements**	42	15	3.75	1.93 – 7.31
***Ascaris* spp.**	Villages	137	4	6.22	1.37 – 28.14
	Towns*	624	3		0.03 – 0.72
	Settlements**	42	5	14.55	4.40 – 48.04
Capillaria aerophilla	Villages	137	4	0.43	0.15 – 1.24
	Towns*	624	40		0.80 – 6.45
	Settlements**	42	1	0.411	0.05 – 2.95

n - number of examined samples; p - number of positive samples; f. - family; 95 % CI - 95 % Confidence Interval of OR; OR - Odds Ratio; *- reference group for calculating OR; **- reference group for calculating the OR in settlements were towns and villages are combined; "-"- unable to calculate due to lack of data

to 500 g were taken from a single site at a surface level (5 cm). Pooled sand samples were then used for parasitological examination. The samples were surveyed according to Kazacos (1983). Endoparasite's developmental stages were determined based on morphological characteristics by light microscopy (Soulsby, 1982; Mircean *et al.,* 2011).
The research related to animals complied with all the relevant national regulations and institutional policies for the care and use of animals. Therefore, no additional authorization was required.

Data analyses and map construction
Statistical analysis of the parasitological results were performed with STATISTICA (version 8.0; StatSoft, TIBCO Software; Palo Alto, California, USA) software for data analysis and MS Excel (Office 365; Microsoft; Redmond, Washington, USA). All data were first described by basic descriptive statistics (mean and standard deviation). Next, the odds ratio (OR)-and correlation coefficient (r) were calculated, which were further converted into a percentage value (Markechová *et al*., 2011). Afterward, the distribution of parasites was displayed using cartogram maps utilizing basic features of the Quantum Geographic Information System (QGIS 3.16, Hannover, Germany).

Results

The results on the prevalence of endoparasites in 803 dog feces from public places are shown in Table 1. Endoparasite developmental forms were found in 350 dog fecal samples, with an overall prevalence of 43.59 %. The most frequently observed endoparasites species were *T. canis* eggs (22.65 %), followed by *Trichuris vulpis* eggs (14.07 %) and eggs of the family Ancylostomatidae (10.09 %). The infection prevalence rates with *Toxascaris leonina*, *Ascaris* spp. and *Capillaria aerophilla* were low and varied from 1.37 % to 5.60 %. No protozoan cysts or oocysts were found in the dog feces collected from public places in East Slovak Lowland. When the endoparasites prevalence was analyzed according to ecosystems (villages, towns, settlements), the results showed that dogs excrements collected from public places in segregated settlements (80.95 %) had higher infection rates than those in towns (45.51 %) and villages (23.36 %; Table 1). The highest occurrence

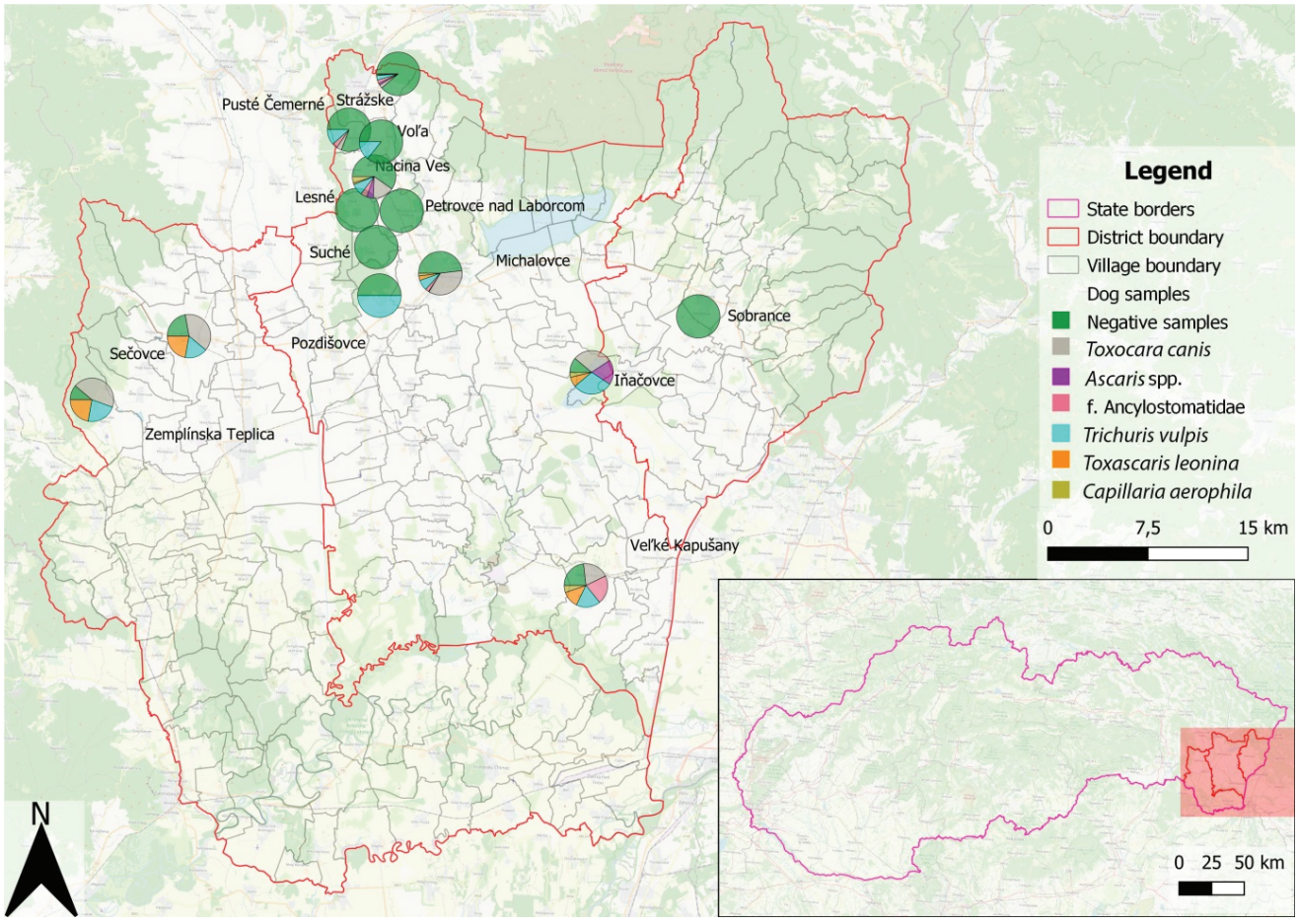

Fig. 2. The spatial distribution of the endoparasites in the dogs' excrements from public places.

Table 2. Results of parasitological examination in the soil.

		N	P	OR	95% CI
Overall	Towns*	50	6		0.015 – 0.11
	Villages	98	71	23.66	8.49 – 65.94
***Toxocara* spp.**	Towns*	50	1		0.003 – 0.20
	Villages	98	42	36.75	4.87 – 277.01
f. Ancylostomatidae	Towns*	50	3	–	
	Villages	98	0	–	
Toxascaris leonina	Towns*	50	2		0.035 – 45.69
	Villages	98	1	0.24	0.02 – 2.79
***Trichuris* spp.**	Towns*	50	1		0.003 – 0.20
	Villages	98	42	36.75	4.87 – 277.01
***Ascaris* spp.**	Towns*	50	0	–	
	Villages	98	55	–	

n - number of examined samples; p - number of positive samples; f. - family; 95 % CI - 95 % Confidence Interval of OR; OR - Odds Ratio; *- reference group for calculating the OR; "-"- unable to calculate due to lack of data

of *T. canis* (52.38 %), *T. vulpis* (35.71 %), *T. leonina* (19.05 %), and *Ascaris* spp. (11.9 %) was found in dog excrements from segregated settlements. *Ascaris* spp. eggs occasionally occurred also in feces collected from towns and villages (Table 1), and these findings are alarming. The eggs of *T. vulpis* and eggs from the family Ancylostomatidae were the most prevalent in dog excrements collected from the town's public places (Table 1). The spatial distribution of parasitic species in the dog's excrements from public places in selected towns, villages and segregated settlements is summarised in Figure 2.

Overall, 77 of 148 soil samples from public places in the East Slovak Lowland (52.03 %) were positive by microscopic examination for endoparasite developmental stages. The occurrence of endoparasites in soil according to the ecosystems is summarised in Table 2. The prevalence of endoparasites in soil from the towns was 12 %, while the prevalence in soil from the villages was 72.45 % (Table 2). During the studied period, the soil's most frequent developmental forms of the endoparasites were eggs of *Ascaris* spp. (37.16 %), *Toxocara* spp. (29.05 %) and *Trichuris* spp. (29.05 %). The eggs of *T. leonina* and Ancylostomatidae occasionally occurred (Table 2).

In 50 examined soil samples from the surveyed towns, 4 different taxa of parasites were detected (Table 2). Typically, the eggs of the Ancylostomatidae family (6.00 %), eggs of *T. leonina* (4.00 %), *Toxocara* spp. eggs (2.00 %) and *Trichuris* spp. eggs (2.00 %) were present. In the villages, the eggs of *Ascaris* spp. (56.12 %) and *Trichuris* spp. (42.86 %) as well as the *Toxocara* spp. (42.86 %) were the most frequent in the 98 examined soil samples. As shown in Table 2, the eggs of *T. leonina* occurred in samples occasionally. The spatial distribution of parasitic species in the soil from the public places in selected towns and villages is displayed in Figure 3.

In total, we investigated 9 various factors (soil type, number of inhabitants, number of Roma, the availability of drinking water in villages and settlements, the accessibility to sewage systems in villages and settlements, the presence of sumps in villages and settlements to identify the potential of risk factors associated with the dog endoparasites infection spread in East Slovak Lowland (Table 3). The target line was established and considered significant if the factor has a correlation share above 10 % (Fig. 4).

Discussion

Our study aimed at the occurrence of endoparasites in dog excrements and soil in public places in East Slovak Lowland and the risk factors associated with dog endoparasites infection spread in these localities. Such a study was needed because of the lack of recent data on this topic in a region close to the Schengen border. Concerning increased migration, there is a more increased chance of introducing parasitic diseases into places historically without endoparasites occurrence. The migration can also negatively impact the epizootological and epidemiological situation, and parasitic diseases spread in localities close to the border. Dog feces in public places thus may represent an important source of endoparasites for dogs, dog owners and the community.

As we mentioned, East Slovak Lowland's region is ethnically mixed. Marginalized groups of inhabitants are represented by Roma people who mostly live in poverty in close contact with animals without veterinary care in segregated settlements. Those dogs move freely in public spaces, not only in settlements but also in towns and villages and can negatively impact the spread of endoparasites. These animals can also come into contact with wildlife, contributing to the widespread parasite spread. Regarding population density and habitat distribution for the spread of parasitic infections, the rusty fox, a state-wide animal, plays the most prominent

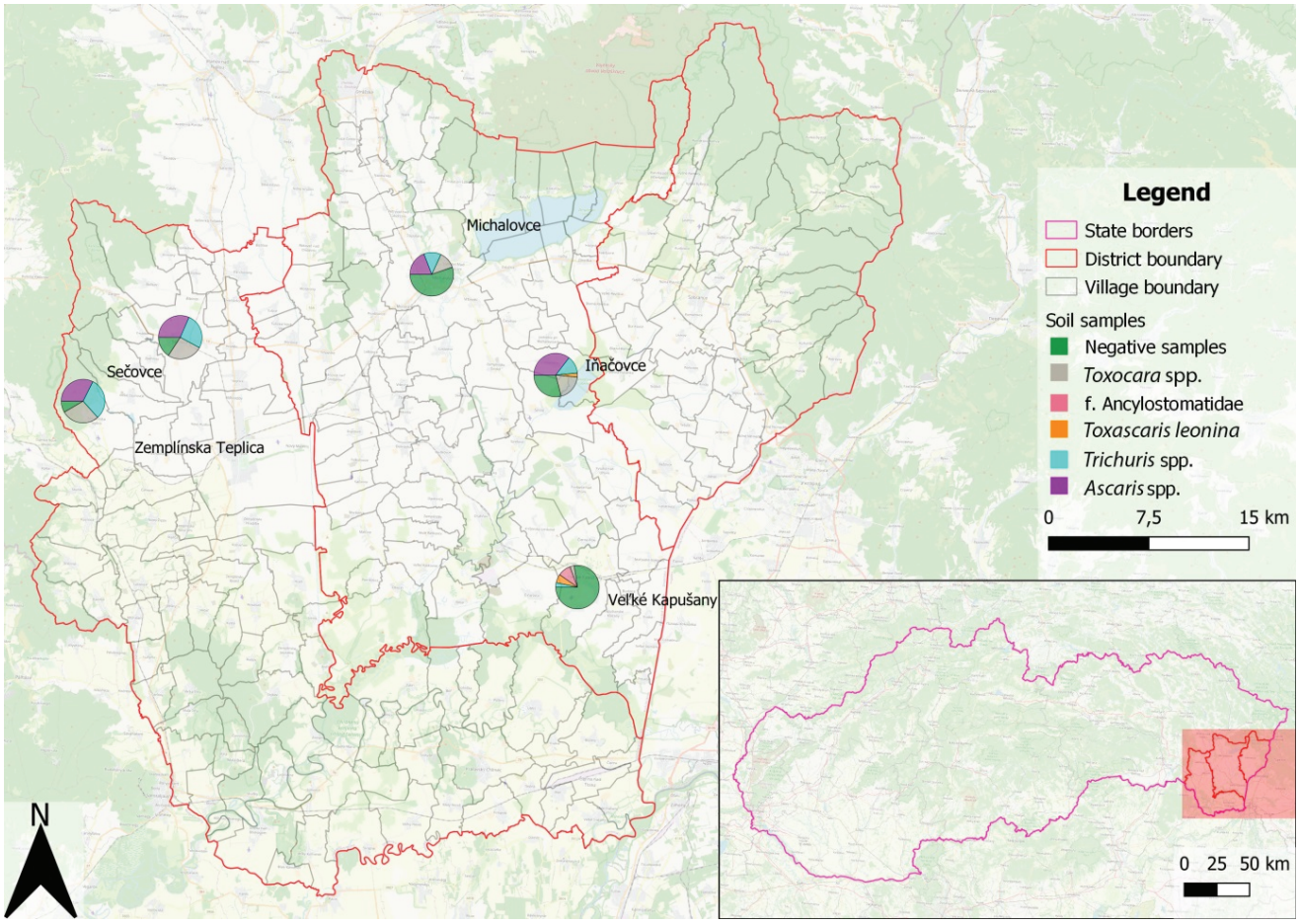

Fig. 3 The spatial distribution of the endoparasites in the soil from the public places.

role. In addition to rabies, it is also an important reservoir for many parasitic diseases, including zoonoses such as *T. canis* and *Echinococcus multilocularis*, the etiological agent of alveolar echinococcosis in humans (Miterpáková *et al*., 2003). Foxes move close to human settlements, leaving their excrements in the surrounding environment and contributing to the parasitic spread, leading to contamination of overlapping sylvatic and synanthropic cycles.

This study revealed a 43.59 % prevalence of endoparasites in dog excrements collected from public places. In feces, only parasites that are common in Slovakia have occurred where the eggs of *T. canis, T. vulpis, T. leonina, Ascaris* spp., *C. aerophilla* and eggs of the family Ancylostomatidae were found in positive samples. Our results correspond with the works of Antolová *et al.* (2004) and Szabová *et al.* (2007). A previous study from Slovak Republic

Table 3. The establishment of correlation coefficients between risk factors and examined samples.

		Correlation coefficient	
		Soil samples	**Dog samples**
Number of inhabitants		-0.764196263	0.962282232
Number of Roma		-0.382587722	0.57934496
Presence of drinking water	village	-0.773563092	0.276271504
	settlement	-0.600215911	0.31220805
Presence of sewage	village	-0.337528609	0.334179395
	settlement	0.423367867	-0.088357355
Presence of sumps	village	0.2645727	-0.241945122
	settlement	-0.248234706	-0.180872313

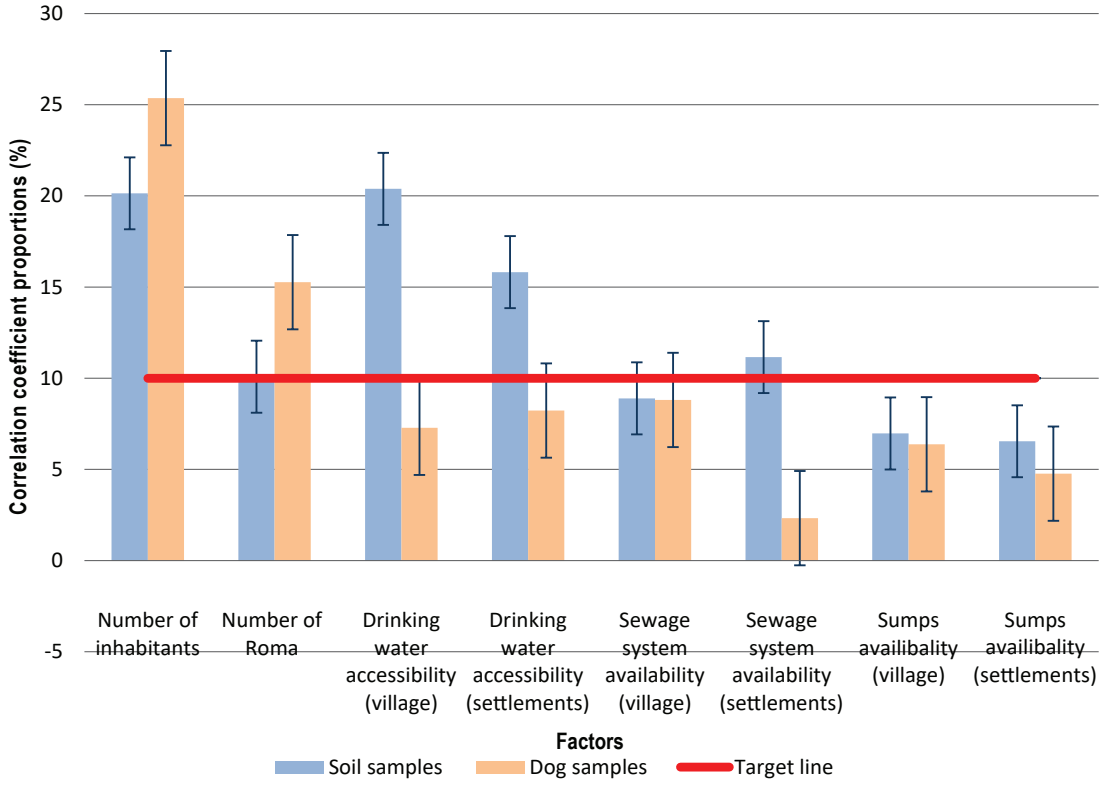

Fig. 4. Comparison of correlation coefficients for each factor in East Slovak Lowland.
Error bars represent SE (Standard Errors).

showed that 12 different species of endoparasites infected 34.90 % of household dogs: *G. duodenalis*, *Cystoisospora* spp., *Sarcocystis* spp., *Hammondia*/*Neospora*-like eggs, *Angiostrongylus vasorum* larvae, *Capillaria aerophila, Crenosoma vulpis, Toxocara* spp., *Toxascaris leonina*, *T. vulpis*, *Strongyloides stercoralis,* and eggs from the family Ancylostomatidae (Šmigová *et al.,* 2021). In western Slovakia, a similar study that monitored the occurrence of parasitosis in dogs from cities was conducted by Ondriska *et al.* (2013), where the *G. duodenalis* (17.8 %) and *T. canis* (16.5 %) were most frequently detected in dogs. Szabová *et al.* 2007 found the *G. duodenalis* oocysts only in the dog excrements from shelters in the cities of Trebišov, Košice and Zvolen. The overall prevalence was 1.60 %. Nevertheless, *Cryptosporidium* spp. was not detected in the dog droppings. In 2014 a higher prevalence of giardiasis (33.30 %) was confirmed from Košice shelters by Štrkolcova *et al.* (2014). The same authors found that the prevalence of giardiasis in Roma settlements near Medzev city was 4.20 %. Pošiváková *et al.* (2014) performed an extensive study within the Košice urban region. The giardiasis prevalence in this territory ranged from 5.90 % to 18.40 %. In contrast to the mentioned authors, no protozoan cysts or oocysts were confirmed microscopically in our survey. It may be caused by the fact that we have examined dogs' excrements from public places originating from different age groups, various dogs' living environments, and excrements could not be collected immediately after the defecation. Nevertheless, we should pay attention to the importance of the endoparasitic protozoan infections transmission between dogs and humans, which is indicated by the existence of a zoonotic cycle in the environment,

Papajová and Šoltys (2020) found a higher incidence of endoparasites in dogs from rural than urban locations. Our research confirmed a higher incidence of endoparasite eggs in the feces of dogs from the towns than in dogs from the villages. This may have been due to the fact that public places in the villages are not so often used for wreaths, and only a few dog owners in the villages had their animals examined. The results of the statistical analysis show that in the segregated settlements, the ratio of the chances of infection in dogs is higher than in the towns and villages. We have not confirmed that in an urban environment where improvements in hygienic conditions exist and endoparasites have been eliminated due to a lack of hosts. Similarly, Rudohradská *et al.* (2011) confirmed a high parasitic occurrence (73.80 %) in dog excrements collected from segregated Roma settlements in Slovakia. In the same way, Pipiková *et al.* (2017) reported differences in the endoparasites occurrence in dog excrements from localities with different hygiene standards. In the villages with a higher standard of living, parasitic infections in dogs were confirmed in 19.44 % of examined samples. Meanwhile, in villages with low hy-

gienic standards where the Roma people live, endoparasites were detected in 71.65 % of samples. The most prevalent were eggs from the Ancylostomatidae family, followed by eggs of *Ascaris* spp. and *T. canis*. The occurrence of naturally human parasites in dog feces points to the dog's role as a mechanical spreader of parasitic germs in the environment (Traub *et al.,* 2002; Shalaby *et al.,* 2010).

This study showed that the distribution of endoparasites in dogs was affected by the population density and the Roma people living in the studied territory. The statistical results show that number of inhabitants and people living in poor hygienic conditions with limited access to drinking water and usage of sewerage play an essential role in the endoparasites spread in the soil.

We conclude that no new endoparasites species pathogenic to humans were found in the dog's excrements and soil samples from public places. Our study confirms that persisting endoparasitic contamination of the public places in East Slovak Lowland may be a reservoir of endoparasites with zoonotic potential, thus representing a public and environmental health problem.

Conflict of Interest

The authors declare no conflict of interest.

Acknowledgment

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