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## *Echinococcus multilocularis* in the red fox (*Vulpes vulpes*) in Slovenia

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**Abstract** Using the parasitological washing out method, we examined the intestines of 428 red foxes (*Vulpes vulpes*) for the presence of *Echinococcus multilocularis* (Leuckart 1863) and found that the overall prevalence was 2.6% (confidence interval 95% 1.3–4.5%). This is the first extended research reporting on the presence of *E. multilocularis* in the Slovenian fox population.

**Keywords** Zoonosis · Tapeworm · *Echinococcus* · Fox

### Introduction

Human alveolar echinococcosis (AE) represents a zoonotic infection which is usually lethal if left untreated. It is caused by the fox tapeworm, *Echinococcus multilocularis* (Leuckart 1863) (Eckert and Deplazes 2004). Its life circle in Europe is predominantly sylvatic, involving the red fox as the main definitive host (Romig et al. 2006). The presence of *E. multilocularis* in red foxes has been detected in many European countries, including Slovenia's neigh-

bours Austria, Hungary, and Italy, but not Croatia (Manfredi et al. 2002; Vervaeke et al. 2005; Bagrade et al. 2008; Borecka et al. 2008; Romig 2009).

Other canids such as wolves, raccoon dogs, coyotes, and wild cats can also be definitive hosts of *E. multilocularis* (Eckert and Deplazes 2004; Kern et al. 2004). Several species of small rodents that serve as intermediate hosts become infected by ingesting the tapeworm eggs (Smith et al. 2003; Kern et al. 2004). Domestic dogs and cats can also act as the definitive hosts of *E. multilocularis* and may therefore be the most important source of infection for people (Deplazes et al. 1999; Gottstein et al. 2001; Seimenis 2003).

Transmission to humans requires ingestion of the parasite eggs in contaminated food, water, soil, or animal fur (Kern et al. 2004; Borecka et al. 2008).

Typical histological lesions on liver and lungs caused by *E. multilocularis* were reported for the first time in Slovenian cattle in 1966 (Šenk and Brglez 1966). Brglez and Kryštufek (1984) also confirmed the presence of the larval form of this tapeworm in the abdominal cavity and liver of a rodent *Apodemus flavicollis* (Melchior 1834), while Logar et al. (2007) in the years 2001–2005 reported a 0.7% prevalence of AE in humans.

Foxes have been present in Slovenia for centuries and inhabit the entire country with an annual catch of approximately 8,000. Despite hunting, the population density of red fox in Slovenia has, due to successful fox vaccination against rabies, increased during the last decade.

The aim of the presented work was to evaluate the prevalence of *E. multilocularis* in Slovenia, since no systematic studies on the fox have been conducted in this area before.

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## Material and methods

### Locations and sampling

During hunting seasons between 2002 and 2005, the carcasses of 428 foxes were collected as road casualties, or as a part of the oral rabies vaccination control programme throughout Slovenia. Samples were collected from four different regions: Central Slovenia ( $n=160$ ), South East Slovenia ( $n=86$ ), East Slovenia ( $n=109$ ), and West Slovenia ( $n=73$ ). Carcasses were labelled and stored in individual plastic bags at 4°C with the necropsy performed within 24 h. Before parasitological examination, the intestine was isolated, removed from the abdominal cavity, and stored for at least 4 days at -80°C.

### Parasitological investigation

The presence of *E. multilocularis* was diagnosed using the technique described by Hofer et al. (2000). The adult parasites were preserved in 70% ethanol. Their identification was based on the microscopic investigation, examining the size and morphology of *E. multilocularis* (Soulsby 1968; Boch and Schneidawind 1988; Ballek 1991).

## Results

A total of 428 fox intestines were collected from different hunting areas in Slovenia between the years 2002 and 2005. Adult tapeworms were discovered in 11 foxes (2.6%). The highest prevalence 7% (6/86) was detected in South east Slovenia, 1.3% (2/160) in Central Slovenia, 1.8% (2/109) in East Slovenia, and 1.4% (1/73) in West Slovenia. Five animals were infected with more than 1,000 adult worms; in the other six animals, two to 22 adult tapeworms were respectively confirmed. The confidence intervals (CI) were calculated for each region as well as for Slovenia as a whole (Table 1).

## Discussion

Red foxes are an important game species in Slovenia, with the hunting bag of around 8,000 foxes per year. According

to the statistical data (Statistical Office of the Republic of Slovenia 2008), the hunting-related reduction of foxes in Slovenia has been increasing gradually in the past few years. However, despite that fact fox population is still growing. The increase in the fox population is probably a consequence of initiation of the antirabies vaccination programme (Chautan et al. 2000; Eckert et al. 2000; Gloor et al. 2001) that has been implemented in Slovenia since 1995 (Hostnik et al. 2006). An increase in the fox population has been reported across the European countries (Eckert et al. 2000; Manfredi et al. 2002; Sréter et al. 2004; Borecka et al. 2008).

Foxes have a significant role in spreading the *E. multilocularis* as the definitive hosts. Due to the unknown situation of *E. multilocularis* in foxes in Slovenia, the monitoring programme has been implemented over the past 4 years. The diagnostic procedures using scraping, sedimentation, and counting techniques are the most reliable methods, but they can be carried out only post-mortem, which may cause some trouble in collecting the samples. It also demands safety precautions (Deplazes et al. 2003).

Parasitological investigation in our study confirmed the presence of adult *E. multilocularis* in 11/428 (2.6%) foxes in Slovenia within four regions. The prevalence was lower in comparison to Switzerland (44.3%) (Hofer et al. 2000), Poland (20.1%) (Borecka et al. 2008), Slovak Republic (10.6%) (Letková et al. 2006), Belarus (7.5%) (Shimalov and Shimalov 2002), Estonia (29.4%) (Moks et al. 2005), Germany, Southern Bavaria (80%) (König et al. 2005), and Northern Germany (average provincial prevalence 19.7%) (Berke et al. 2008), while in Belgium a 1.8% prevalence was confirmed (Vervaeke et al. 2005).

The data from neighbouring countries (Austria, Italy, and Hungary) revealed the presence of *E. multilocularis* (Duscher et al. 2006; Casulli et al. 2005; Sréter et al. 2004), while in Croatia (Rajković-Janje et al. 2002), the presence of *E. multilocularis* was not reported. According to the latest records, the known range of this parasite has extended across Europe (Romig 2009); therefore, the monitoring programmes in regions of former Yugoslavia could contribute valuable data on the presence of this parasite.

In our research, five foxes were heavily infected with more than 1,000 adult tapeworms, four of them originating

**Table 1** Prevalence of *Echinococcus multilocularis* in foxes in Slovenia

Region	Examined foxes	Positive	Estimated prevalence (%)	95% CI exact
Central Slovenia	160	2	1.3	0.00152–0.04442
South East Slovenia	86	6	7	0.02603–0.14569
East Slovenia	109	2	1.8	0.00223–0.06471
West Slovenia	73	1	1.4	0.00035–0.07398
Slovenia	428	11	2.9	0.0129–0.04552

from South East Slovenia, while in six foxes the presence of only two to 22 adults was confirmed. The distribution of positive diagnosed cases was expected, as the majority of animals originated from the area with the highest population density. As was previously reported (Shimalov and Shimalov 2002; Sréter et al. 2004; Moks et al. 2005; Takumi et al. 2008), the number of tapeworms in our research varies significantly among the individual animals.

This is the first report on *E. multilocularis* in the territory of former Yugoslavia and even in South Eastern Europe. Up to now, the parasite was not discovered in red foxes in Slovenia, but AE has already been reported in cattle (Šenk and Brglez 1966) and in rodents *A. flavicollis* (Brglez and Kryštufek 1984). In the years 2001–2005, Logar et al. (2007) reported a 0.7% prevalence of AE in humans in Slovenia and suggested that dogs and cats might be a possible source of this infection (Romig et al. 1999; Romig 2003; Thompson et al. 2006). The general role of domestic cats and dogs in the epidemiology of the infection has not yet been studied in Slovenia; additional studies on this topic are therefore required. Since other European countries have already tested the presence of *E. multilocularis* in red fox, similar results were expected in our research. *E. multilocularis* was recently confirmed also in the Baltic states, in most of Belgium, in the south and west of The Netherlands, northern Italy, eastern Austria, and in northern Hungary (Romig 2009). Although with low prevalence, the presence of *E. multilocularis* might present a potential threat for AE in humans. In all the countries where the parasite shows the tendency of spreading, a continuous monitoring programme should be considered.

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