

# The important zoonoses in the protected areas of the Tatra National Park (TANAP)<sup>1</sup>

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**ABSTRACT.** The northern part of Slovakia constitutes an important tourist and recreational area of the country. Protected localities of the Tatra National Park (TANAP) are characterised by specific ecological conditions. The high numbers of animals inhabiting protected areas of the TANAP and their potential encounters with tourists contribute to a risk of transmission of important parasitic zoonoses. The aim of presently reported study was to perform a long-lasting and detailed survey on the occurrence of zoonotic agents (in particular *Echinococcus multilocularis* and *Trichinella* spp.) with particular emphasis on the interactions between the environment, the host, and the parasite as well as the adaptation mechanisms in individual ecosystems. Within the pilot part of the study, which started in 2007, in total 397 samples of wild predators representing 10 species belonging to families Canidae, Felidae, Mustelidae and Ursidae were investigated. Helminthological necropsy (modified sedimentation and counting technique) and artificial digestion method were used for the parasites detection. Multiplex PCR approach has been used for species identification of *Trichinella* isolates obtained from infected animals. *E. multilocularis* was found in 42.7% of foxes and 1 raccoon dog. *Trichinella* larvae were present in 16.7% of foxes, 37.9% of martens, 33.3% of polecats, 1 bear and 1 lynx. All animals were infected by *T. britovi*.

**Key words:** zoonoses, TANAP, protected areas, helminths, echinococcosis, trichinellosis

## Introduction

The northern part of Slovakia constitutes an important tourist and recreational area of the country. In Slovakia, as well as in most of Europe, a significant increase of red fox (*Vulpes vulpes*) population arises due to sanitation of its population via anti-rabies vaccination during last decade. The growth of population goes along with spread into urban- and suburban localities, including recreational areas. In 2004 a windstorm affected the High Tatra Mountains that caused increase of annual temperatures when comparing with long-term average and effected significantly foraging and migration behaviour of wildlife. Migration of existing and immigration of new wildlife populations caused by damage or change of natural biotopes is expected. That implies the increase in game numbers followed by growth of carnivorous predator populations. The increase of biomass,

biodiversity and population density of small mammals has already been documented in areas impacted by windstorm. Those changes represent the risk of transmission and spread of parasitic infections, as they favour the circulation of many parasitic infections of zoonotic importance.

Alveolar echinococcosis (AE) is one of the most important parasitic zoonosis of Northern Hemisphere. The infection is caused by fox tapeworm *Echinococcus multilocularis*. Larval stage develops in intermediate hosts – small mammals, but occasionally also in human, where it causes a fatal disease. Longitudinal epizootiological study in red foxes pointed out the overall distribution of parasite and revealed profound geographical differences in the prevalence. In Northern Slovakia highly endemic foci occur with prevalence of 40–60% infected foxes. In these areas also first autochthonous human cases of AE were found [1,2].

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Table 1. Carnivore species infected with *Echinococcus multilocularis* and *Trichinella britovi* in TANAP area

Animal species	<i>E. multilocularis</i> Exam/posit	<i>E. multilocularis</i> prevalence	<i>Trichinella</i> spp. Exam/posit	<i>Trichinella</i> spp. prevalence
Red fox ( <i>Vulpes vulpes</i> )	328/140	42.7%	328/55	16.7%
Wolf ( <i>Canis lupus</i> )	0/0	–	6/0	0
Stone marten ( <i>Martes foina</i> )	3/0	0	3/1	33.3%
Pine marten ( <i>Martes martes</i> )	1/0	0	2/1	50%
European polecat ( <i>Mustela putorius</i> )	3/0	0	9/3	33.3%
Badger ( <i>Meles meles</i> )	1/0	0	2/0	0
European otter ( <i>Lutra lutra</i> )	0/0	–	4/0	0
Least weasel ( <i>Mustela nivalis</i> )	1/0	0	10/0	0
Stoat ( <i>Mustela erminea</i> )	0/0	–	5/0	0
Raccoon dog ( <i>Nyctereutes procyonoides</i> )	2/1	50%	2/0	0
Brown bear ( <i>Ursus arctos</i> )	0/0	–	2/1	50%
Lynx ( <i>Lynx lynx</i> )	0/0	–	2/1	50%

Trichinellosis is a zoonosis with cosmopolitan distribution and natural foci transmission patterns. In Slovakia, it occurs almost exclusively in sylvatic cycle with sporadic human outbreaks. The infection arises following the ingestion of meat harbouring infective larvae of parasite. Many human outbreaks occurred recently, caused by consumption of game meat or due to pork infected when pigs were fed with offal from game [3]. Within the long-term monitoring, trichinellosis was found widespread across Slovakia, the prevalence in red foxes increased from 4.9% in 2000 up to 20.1% in 2007 and new endemic foci emerged in recently [4].

Dynamic changeovers that are in progress in the TANAP areas impacted by windstorm are monitored by advanced network, which provides large-scale environmental data. The aim of our study was to use these data when studying the occurrence of zoonotic agents (*E. multilocularis* and *Trichinella* spp.), the host-parasite relationships and adaptation mechanisms in given ecosystem.

## Materials and methods

The pilot part of the study started in 2007 and

animals shot during the hunting season or being victims of car accidents were sampled from the territory of the Tatra National Park. In total, 328 red foxes (*Vulpes vulpes*), 6 wolves (*Canis lupus*), 12 stone martens (*Martes foina*), 8 pine martens (*Martes martes*), 6 badgers (*Meles meles*), 9 european polecats (*Mustela putorius*), 4 european otters (*Lutra lutra*), 10 least weasels (*Mustela nivalis*), 5 stoats (*Mustela erminea*), 5 raccoon dogs (*Nyctereutes procyonoides*), 2 brown bears (*Ursus arctos*), and 2 lynxes (*Lynx lynx*) were investigated. For the presence of *Echinococcus multilocularis* small intestines were examined using sedimentation and counting technique (SCT) according to Raoul et al. [5]. Tapeworms were identified following morphological criteria according to Thompson [6]. Muscle samples (minimum of 10 g) were examined individually for presence of *Trichinella* spp. larvae by artificial HCl-pepsin digestion method according to Kapel and Gamble [7]. Larvae collected from infected animals were identified at species level by the multiplex polymerase chain reaction (PCR). Extraction of DNA and PCR amplification was done according to Pozio and La Rosa [8].

## Results

Summarized data from investigation for the presence of *E. multilocularis* and *Trichinella* spp. are presented in Table 1. From 328 red foxes originated in Tatra area, 42.7% were infected with *Echinococcus multilocularis*. Mean worm burden was 9 759 tapeworms with range from 1 to 245 000 and total biomass found in all infected foxes was 556 541 specimens. The most of the infected animals (37.15%) harboured 101–1 000 tapeworms, 18.6% of foxes were classified as belonging to a high worm burden class (1 001–10 000) and 6.4% of foxes were infected with more than 10 000 *E. multilocularis* worms. Also red foxes originated in (for visitors) the most attractive localities (Smokovce, Tatranská Lomnica and Ždiar) were found to be infected. One raccoon dog was also infected with *E. multilocularis*.

Concerning trichinellosis, 16.7% of red foxes, 37.9% of martens, 33.3% of polecats, 1 bear and 1 lynx harboured muscle larvae. Infected animals were more frequently found in the Low Tatra and the Pieniny National Parks in comparison to the High Tatra area. The intensity of infection was investigated and expressed as the number of larvae per gram of muscle tissue (lpg). The volume of infection varied slightly with the highest value of 51.3 lpg in Ždiar (aver. 2.1 lpg). The results of molecular analyses revealed exclusive presence of sylvatic species *Trichinella britovi*.

## Discussion and conclusions

In total 397 samples of wild predators representing 10 species belonging to families Canidae, Felidae, Mustelidae and Ursidae were investigated. The most numerous among examined predators were red foxes. According to our data, 42.7% of foxes and 1 raccoon dog were infected with *E. multilocularis*. This tapeworm was for the first time detected in red fox in 1995 in the north of Poland [9] and southern and western districts of the Czech Republic [10]. The first finding of *E. multilocularis* on the territory of Slovakia was recorded in 1999 [11]. A total prevalence of *E. multilocularis* determined during the period of 2000–2006 in Slovakia reached 31.1%. The results of long-term monitoring refer to the occurrence of two endemic areas situated in northwestern (Žilina and Trenčín region) and northeastern part of Slovakia (Prešov region), where also TANAP area

is included in, with the prevalence rates in several districts ranging between 40% and more than 60%. An important role in great differences in prevalence between northern and southern parts of the country play factors as fox population density, presence of intermediate hosts, and climatic conditions. The results of long-term monitoring provide the evidence that climatic conditions in endemic areas differ significantly from those in other parts of the country. Low mean annual air temperature, high annual precipitations, low mean annual temperature of soil surface and high humidity of the soil are characteristic for northern Slovakia. Cold and moist environment may be a key factor for the survival of the tapeworm eggs that are very sensitive for desiccation and high temperatures [12]. Our recent analyses showed significant correlation between prevalence of the tapeworm, mean annual precipitation values and population density of small mammals [2,13].

According to the epidemiological analyses, conducted within the zoonoses-surveillance program in the Slovak Republic, the prevalence of trichinellosis in wildlife of Slovakia varied in last 50 years. In 1954–1956 Mituch [14] examined free-living carnivores and found lynx to be most infected species with the prevalence value of 66.67%. Of other hosts, 33.3% of wolves, 27% of red foxes and 15.8% of wild cats were infected. In the Tatra National Park, the last monitoring on trichinellosis was performed in 1988–1989 by Mituch et al. [15]; 174 carnivores belonging to 9 species were examined, the parasite was absent in most of them except wolf and red fox, with prevalence rates 20.0% and 5.5%, respectively. However, results of our present study obtained more than one decade later are indicative of fluctuation trichinellosis among foxes that was found widespread across Slovakia and the prevalence increased from 4.9% in 2000 up to 21.5% in 2007 [4]. In Poland, 5.7% of foxes were found to be positive for trichinellosis in southern parts of Poland [16].

Our study revealed the intensity of infection with trichinellosis being highest in endemic mountainous areas of eastern a central Slovakia. It is generally considered that sylvatic trichinellosis affects carnivores with cannibalistic and scavenger behaviour and higher population density causes intense competition for food and thus results in more scavenging and cannibalism, which are the major routes of *Trichinella* transmission [17]. The maintenance and transmission mechanism are

conditioned by local food-chain and environments, making specific host accessible. Our investigation results revealing high number of infected carnivores from family Mustelidae (25%) are emphasize that in many areas of Europe the red fox is not the only wild predator involved in sylvatic cycle of *T. britovi*. This confirms the suggestion that mustelids should also be considered when assessing *Trichinella* epizootiology, because of their synanthropic behaviour that can represent a link between sylvatic and domestic cycle.

The high number of infected animals inhabiting protected areas of TANAP and their close proximity with tourists represents the transmission risk of important parasitic zoonoses. The presently reported findings pointed out the necessity to introduce such preventive and control measures that can minimize the risk of parasites spread and transmission to human. Feasible solution for reduction of *E. multilocularis* incidence in recreational localities is the application of baits containing antihelminthics drugs. Veterinary and sanitary educational programs for tourists and hunters are inevitable to maximize prevention and control of both, echinococcosis and trichinellosis.

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