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A survey of the current status of sylvatic trichinellosis in the Republic of Croatia

Tihomir Florijančić[1]*, Albert Marinculić[2], Boris Antunović[3], and Ivica Bošković[1]

[1]*Department of Zootechnics, Faculty of Agriculture, Josip Juraj Strossmayer University in Osijek, Osijek, Croatia*

[2]*Department for Parasitology and Invasive Diseases, Faculty of Veterinary Medicine, University of Zagreb, Zagreb, Croatia*

[3]*Department of Animal Husbandry, Faculty of Agriculture, Josip Juraj Strossmayer University in Osijek, Osijek, Croatia*

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ABSTRACT

In the Republic of Croatia both sylvatic and domestic infection cycles of *Trichinella* spreading are known. Domestic trichinellosis, caused by *Trichinella spiralis*, is a primary infection of domestic swine, and rats are the main carriers. The percentage of prevalence during the 1980-ies was under 0.10% and increased to 0.79% in the 1990-ies. That was the reason for the increased number of human infections. Eradication of domestic trichinellosis has been prescribed by following the regulations and systematic monitoring. Sylvatic trichinellosis exists latently, with *T. spiralis* and *T. britovi* as the main etiological factors. So far it is being proved in wild boar, bears and badgers and also in red foxes and wolves, wild animals, which enables the maintenance and dissemination of the infection in the wild on Croatian territory. The meat of wild boar, bear and badger consumed by people is a potential health risk. Studies indicate the constant presence of sylvatic *Trichinella* infection in the aforementioned wild animal species. They are a key factor in the circulation of sylvatic trichinellosis in natural ecosystems. No detailed data are available on sylvatic trichinellosis in Croatia. The prevalence of *Trichinella* spp. has only been studied in selected animal species and the intensity of infection or species composition were not determined. In order to identify areas with presence of the sylvatic trichinellosis, there is a great need to establish monitoring of hunted and dead carnivoric and omnivoric wild animals. This would make it possible to eradicate it, and to stop the transmission between wild and domestic animals.

Key words: sylvatic trichinellosis, epizootiology, game, wildlife

* Contact address:
Dr. Tihomir Florijančić, Chair for Agrarian Zoology, Department of Zootechnics, Faculty of Agriculture, Josip Juraj Strossmayer University in Osijek, Trg Svetog Trojstva 3, 31000 Osijek, Croatia, Phone: + 385 31 224 275; Fax: + 385 31 207 017; E-mail: flory@pfos.hr

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Introduction

Trichinellosis is an invading disease caused by the parasitic nematodes of the species *Trichinella*. This important zoonosis occurs mainly in wild carnivores with cannibalistic and scavenger behaviour (POZIO, 1999). *Trichinella* infection exists both in the sylvatic (i.e. in wildlife) and domestic cycle. Sylvatic trichinellosis is an infection of game and other wild animals living free in nature (i.e. red foxes, wolves, wild boar, bears, badgers etc.) and feeding on other animals and/or their carcasses is the key factors in transmission of the *Trichinella*. The domestic cycle involves infection and transmission by domestic swine and rodents, especially rats. The sylvatic cycle has been described in the majority of European countries, with the exception of Denmark and most insular countries (Ireland, Iceland and the United Kingdom), which can be considered *Trichinella*-free areas. However, islands in the Baltic and Barents Sea, connected with land by ice-cover, do have sylvatic trichinellosis (POZIO, 2001). Among the eleven known species of the nematode genus *Trichinella*, four of them have been identified in the sylvatic cycle in Europe.

T. spiralis is present in many countries, but the sylvatic cycle exists only concurrently with the domestic cycle, or it existed before. This species possesses the highest infectivity for domestic swine and wild boar (KAPEL, 2000). *T. spiralis* has not been found in wild animals in Estonia, Italy, Norway and Switzerland, while for Greece and Portugal, there is no data.

T. nativa is the main etiological agent of the sylvatic cycle in Arctic and sub-Arctic areas. In Europe it has been detected in Byelorussia, Estonia, Finland, Norway, Russia, Sweden and Ukraine. The southern distribution border is limited to the -5 ^{o}C isotherm in January. The largest natural reservoir of this species is the polar bear.

T. britovi has been detected in wild animals in the temperate zones in Europe (many countries including Croatia) and Asia. The northern distribution border is limited to the -6 ^{o}C isotherm in January. The main reservoirs of this species are red foxes and racoon dogs, but can be other carnivores (wolves, bears, weasels etc.).

T. pseudospiralis is non-encapsulated species, detected in carnivores, carnivore birds, marsupials, rodents, swine, and rarely in human. It occurs in many natural areas of Australia, Northern America and Asia. In Europe it has been found in Finland, France, Italy, Russia, Spain, Sweden, Russia, Lithuania and the Slovak Republic (POZIO, 1998; HURNÍKOVÁ et al., 2005). *T. spiralis* and *T. pseudospiralis* are characterized by cosmopolitan distribution, while the other species are limited to particular zoogeographic areas, mainly outside Europe.

In Croatia, until 1991 (the beginning of the Homeland War), trichinellosis occurred sporadically, mainly in the eastern part of the country. During the 1990-ies it became one of the most serious public health problems (MARINCULIĆ et al., 2001). Most people were

infected by pork, while some were infected by uncooked game meat, wild boar, badgers or bears (BEUS, 1999). Since both *Trichinella* infection cycles are known from the past in Croatia, and wild animals are a natural reservoir of this serious parasitic nematode, the aim of our work was to analyse the epizootiology of the sylvatic cycle in particular animal species and to assess the potential risk for infecting humans.

Materials and methods

This paper considers the prevalence of trichinelosis in five animal species: wild boar (*Sus scrofa* L.), brown bears (*Ursus arctos* L.), badgers (*Meles meles* L.), red foxes (*Vulpes vulpes* L.) and wolves (*Canis lupus* L.) over a twenty-seven year period (1977 – 2004). Examination samples of muscle tissue are taken from hunted or perished animals throughout the entire Croatian territory. Examinations were made by trichinelloscopy and artificial digestion methods (GAMBLE et al., 2000) in authorised laboratories, while genetic identification was performed by multiplex polymerase chain reaction (PCR) (POZIO and LA ROSA, 2003).

Results and discussion

Wild boar (*Sus scrofa* L.), according to the Hunting Act (ANONYMOUS, 2005), is categorized as big game. It is prevalent at almost all the territory of the Republic of Croatia with the exception of certain Adriatic islands, where sometimes appears by natural migration, swimming over the sea or by human introduction. The number of wild boar increased considerably in the last decade of the 20th Century. Wild boar are omnivorous and thus it can be easy infected by *Trichinella* spp., and the meat of this game, used for human consumption, can be a potential source of infection in humans. The most common pathogen of trichinellosis in pigs and wild boars is *T. spiralis*, although in Croatia infection of *T. britovi* has been determined, and also mixed infections in wild boars (MARINCULIĆ et al., 2001). Whereas the human infection occurs through eating inappropriately thermally processed meat, the largest danger is in areas where processing and preparing meat products are done on a traditional basis (for example sausages, paprika-flavoured sausage – kulen, etc). These products are most common in eastern Croatia, where meat products are home made, and prepared from pork meat to which wild boar meat is added as a flavour enhancer. On the other hand, because of the significant prevalence of trichinellosis in pigs and humans in the middle of the nineties, the eastern part of Croatia was proclaimed as a trichinellosis endemic area. The new epizootiological situation resulted in the adoption of new legal regulations which regulate certain measures for the repression and prevention of trichinellosis (ANONYMOUS 1998; 1999). This involved the obligatory inspection of pork meat for slather for domestic purposes and also game meat for human consumption (Council Directive 92/45/EEC).

Trichinellosis in wild boars in Croatia was well-known many years before, as confirmed by studies made by BATIS et al. (1977) who determined 2.4% prevalence of trichinellosis. Studies made at the Croatian Veterinary Institute in a five year period (1986–1990) on 4359 samples of wild boar meat, proved that nematode *Trichinella* spp. was present in 28 samples (0.64%) (ČOVIĆ and VINKOVIĆ, 1998). VUČEMILO et al. (1998) announced that prevalence of trichinellosis in wild boars in eastern Croatia in 1995 was 5.8%, and in 1996 5.7%. TUCAK et al. (2000) determined that in the common open hunting grounds in the Osječko-baranjska County, the prevalence of trichinellosis in wild boars was 0.34%. VUČEMILO et al. (2001) determined the same prevalence (0.35%) in the state hunting ground Podunavlje-Podravlje in the Baranja region. This data prove the constant presence of the nematode *Trichinella* spp. and confirms that wild boar is a latent reservoir of this parasite in the Republic of Croatia, especially in the areas of Slavonia and Baranja. We have to have this fact in mind when by consuming wild boar meat and because of the potential contact of domestic pigs with wild animals through traditional (ecological) breeding methods.

Brown bears (*Ursus arctos* L.) are the biggest game in our biotope. They inhabit a total of 15700 km^2 but live permanently in 10200 km^2. This area is mostly covered by woods and located in Lika and Gorski Kotar in the Dinarides (HUBER, 2004). The number of bears is between 400 and 600 units, and is one of the rare stable populations in Europe. Today in Croatia, hunt management of bears is outside protected areas; the annual shoot quota is around 10% of the population. FRKOVIĆ (2002) quotes that in a 10 year period (1990-1999) the total mortality of bears was 273 units, whereof 138 units were shot in the hunting period, and the other half were killed in various other ways (illegal hunting, traffic and train accidents, effects of war, etc.). Although bears are originally carnivorous, in terms of body structure, about 95% of them supplement their eating habits with plant food, and some of their animal proteins are composed mostly of invertebrates and corpses of big animals. This is, probably, the main reason why in the five year period from 1986-1990 in the Croatian Veterinary Institute in Zagreb 28 bears were examined of which only 2 were diagnosed with *Trichinella* infection (7.14%) (ČOVIĆ and VINKOVIĆ, 1998). In previous years (BATIS et al., 1977), the prevalence of trichinellosis in bears was 7.04%. After the implementation of the legal obligation of examining meat and all epizootiologicaly important groups of game for trichinellosis, according to the Ministry of Agriculture, Forestry and Water Management of the Republic of Croatia only one bear was determined to have *Trichinella* infection when shot in the Karlovačka County area in 2003. This case confirms the persistent danger for human health from consumption of bear meat even in areas declared trichinellosis endemic, and this should be valorised in the epizootiological situation assessment.

The badger (*Meles meles* L.) is small game prevalent from lowland to mountain regions in Croatia with the exception of the Adriatic islands. In terms of its eating habits it is an omnivore, and because of the use of its meat for human consumption (mostly in northwest and eastern Croatia) it is a potential source of trichinellosis. Until now, there have been no recorded cases of trichinellosis originating from badger meat, most probably because of how it is prepared i.e. good thermal processing. At the Croatian Veterinary Institute in Zagreb, in the period from 1986 to 1990, by testing the tissue of two badgers, no larvae of *Trichinella* spp. were found ČOVIĆ and VINKOVIĆ (1998). BATIS et al. (1977) determined a prevalence of trichinellosis in badgers of 0.9%. Literature does not quote any other data of *Trichinella* infection in badgers in Croatia, but in our own examination (badgers killed in traffic) we identified very strong infections in Slavonia, in the area around Đakovo city and Belišće city. Since the badger is a natural reservoir of the *Trichinella* spp., and especially the meat of this animal is a culinary speciality, permanent monitoring of this illness is necessary.

Red foxes (*Vulpes vulpes* L.) are small game prevalent throughout Croatia and also on some Adriatic islands (Krk, Rab). It can rarely be found at altitudes above 700 m because of the lack of its basic food (JANICKI, 2004). Regarding feeding, it is very adaptable, preferring food of animal origin (feather game, domestic poultry, eggs, small rodents and so on), where this is lacking, foxes will even eat plants. As regards trichinellosis, the fox has not any particularly importance regarding human health because its meat is not for consumption, but in epizootiology it is an important link in the spread of this disease, especially due to its feeding habits, whereas it can be invaded by eating contaminated animals or their corpses. On the other hand, its corpse can be a source of infection for other game, as well as for domestic animals. In Western Europe the fox is the important indicator animal for the epizootiology of sylvatic trichinellosis (VAN KNAPEN, 1998). BATIS et al. (1977) say that of 7000 examined foxes for trichinellosis in Croatia, 896 of them were positive (12.8%). KOVAČ et al. (2001) examined the prevalence of trichinellosis in foxes in three Counties in the endemic area in eastern Croatia (Vukovarsko-srijemska, Osiječko-baranjska and Brodsko-posavska), while of 1652 examined samples of fox muscle tissue, 82 were positive (4.9%). Most of them were found in the Vukovarsko-srijemska County (11.1%). This data confirms that the fox has an important role in the sylvatic cycle in maintaining and spreading of trichinellosis.

The wolf (*Canis lupus* L.), according to the regulations in Croatia, has not been included as game since 1994 (ANONYMOUS, 2005), and is protected species since 1995 (ANONYMOUS, 1995). Today wolves inhabit an area of 17468 km^2, mostly in the Dinarides area (Lika and Gorski Kotar) and Dalmatia. The total population is estimated as 100-150 units (KUSAK, 2004). As a distinct carnivore, they feed mostly on large mammals, but also

any other animal they catch. BECK et al. (2004) researched over a nine year period (1996-2004) the prevalence of trichinellosis in wolves in Croatia and of 33 examined samples, 9 were infected (27%). Using the multiplex polymerase chain reaction (PCR) method, the *T. britovi* type was determined in all wolves. From a epizootiological point of view, it is important to say that besides positive wolves shot in the area of Lika and Dalmatia, one wolf positive for trichinellosis was also shot near Zagreb. Like the fox, the wolf is not a direct source of infection by the *Trichinella* spp. parasite, but since this predator has no natural enemy; its corpse could be a possible source of contamination for other animals.

Conclusion

Although no systematic studies have been performed on trichinellosis prevalence in free-living animals in Croatia, the occurrence of *Trichinella* parasites has been demonstrated in several animal species, potentially representing a risk for human infections. To obtain more profound knowledge on the overall prevalence of sylvatic trichinellosis and to ascertain *Trichinella* species distribution, it is necessary to conduct parasitological monitoring of trichinellosis in hunted and dead carnivores and omnivores, by examining muscle tissues in accredited veterinary organisations. The basic goals will be the determination of endemic areas as well as designing effective prevention programs. No less important is the education of hunters regarding the means of infection and its consequences, and maintaining sanitary order in hunting grounds, safe removal of shot and dead game, and also their parts.

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FLORIJANČIĆ, T., A. MARINCULIĆ, B. ANTUNOVIĆ, I. BOŠKOVIĆ: Istraživanje silvatične trihineloze u Republici Hrvatskoj. Vet. arhiv 76, S1-S8, 2006.

SAŽETAK

U Republici Hrvatskoj opisana su oba epizootiološka ciklusa širenja trihineloze, silvatični i ruralni. U ruralnom ciklusu, glavni je etiološki čimbenik *Trichinella spiralis*, a njeni (pre)nositelji domaće svinje i štakori. Prevalencija, koja je u svinja tijekom 1980-ih godina iznosila 0,10%, tijekom 1990-ih se povećala na 0,79%, što je rezultiralo povećanjem broja oboljelih ljudi. Njezino suzbijanje regulirano je zakonskim propisima i sustavnim nadzorom. Silvatična trihineloza prisutna je u latentnom obliku i znatno ranije, a njezini su glavni uzročnici *T. spiralis* i *T. britovi*. Do danas je silvatična trihineloza dokazana u divlje svinje, medvjeda i jazavca, čije meso služi kao ljudska hrana i kao takvo predstavlja potencijalnu opasnost za njihovo zdravlje. Invazija je također dokazana u lisice i vuka, životinjskih vrsta koje su iznimno značajne u epizootiološkom smislu za održavanje i širenje trihineloze u prirodi. Istraživanja govore o njezinoj stalnoj prisutnosti u navedenih divljih mesoždera odnosno sveždera, što se smatra ključnim čimbenikom za održavanje silvatične trihineloze u prirodnim ekosustavima u Republici Hrvatskoj. Silvatična trihineloza do sada nije sustavno istraživana, nego samo djelomično, po pojedinim životinjskim vrstama, pri čemu je uglavnom utvrđivana tek učestalost oblića *Trichinella* spp., bez određivanja jačine invazije, vrsne determinacije i dr. Radi otkrivanja žarišta trihineloze u divljih životinja, prijeko je potrebno organizirati i sustavno provoditi parazitološki nadzor nad odstrijeljenim i uginulim mesoždera i sveždera. Ovime bi se moglo pridonijeti njezinom iskorjenjivanju, kao i sprječavanju širenja uzročnika između divljih i domaćih životinja.

Ključne riječi: silvatična trihineloza, *Trichinella* spp., epizootiologija, divljač