

THE IMPORTANCE OF TRICHINELLOSIS AND OTHER ZOOSES OF THE WILDLIFE IN THE WEST-BALKAN REGION

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Summary: In this contribution, reports on zoonoses in the West Balkan region are collated. Compared to data on *Trichinella* sp., reports on other parasitological, or bacterial and viral zoonoses are rather scarce. Effective control of zoonotic diseases will require more efforts in studying the role of wildlife as a reservoir for zoonoses. This includes epidemiological studies as well as ensuring effective meat inspection of game, implementation of safe game handling and evisceration techniques and safe disposal of offal. The latter requires consciousness and compliance of hunters, which has to be based on training and motivation.

Key words: game animal, zoonoses, West-Balkan, epidemiology

Introduction

Zoonoses are diseases that may be transmitted from animals (vertebrate) to humans under natural conditions. Humans may become infected via contact with or handling of animals (“occupational” diseases) or by consumption of contaminated food. [1]. Almost a half out of 1700 known pathogens affecting humans is estimated to be zoonotic [2].

Wild animals may act as a reservoir and permanent source from which domestic animals and humans may be infected and invaded [3]. Up to 335 pathogens have been estimated to be associated with emerging infectious diseases in the global human population between 1940 and 2004, the majority of them being associated with wildlife [2].

Outbreaks of wildlife zoonotic diseases were reported on the Balkan Peninsula during the last twenty years [4]. The wartime period (1992-1995) was associated with socio-economic, human demographic, behavioral, ecological and environmental changes that had significant impact on public health in general and on wildlife zoonotic diseases in particular [5].

In the West Balkan region, wild game meat has great economic (hunting), and nutritional significance -in cooking because of the high content of valuable protein and less fat content. Certainly, ensuring the safety of game meat has an important role in the entire system of food safety, particularly in terms of zoonoses. However, it is necessary to improve the current situation, because in terms of hunting tourism and the export of game meat we need to harmonize our veterinary and sanitary regulations with European Union member countries. It requires also a follow-up monitoring of wild game meat hygiene and zoonotic agents in wild game, including the development of risk analysis and control model. [44].

Viral agents in the wildlife in the West-Balkan region

Rabies

Rabies remains endemic in Southeast Europe including Romania, Bulgaria, B&H and Turkey [6]. With the exception of Turkey, the red fox (*Vulpes vulpes*) is the principal reservoir for this viral agent in Southeast Europe. Also, cases of rabies in dog (*Canis familiaris*) are regularly reported. In Croatia, rabies has been detected in wild animals (mostly foxes) and sporadically in domestic animals (dogs, cats). The last human case in Croatia was described in 1964. Sylvatic rabies is in enzootic form present in Serbia, while the urban rabies was eradicated during the eighties in last century. For the purpose of eradication of rabies from 2010. in Serbia is performed oral vaccination of wildlife against rabies within the regional program of eradication of contagious animal diseases, funded from the budget of the Republic of Serbia and the EU pre-accession funds (IPA funds) [41]. Throughout the region there is evidence of cross-border movement of rabies by both wildlife and canine vectors [7].

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Hepatitis E virus

Preliminary serological tests conducted in the Scientific Veterinary Institute in Novi Sad included the examination of 92 blood serum samples of wild boar, which had been collected in 2009, 2010 and 2011 from 15 hunting sites in Serbia. In 32 serum samples (34.8%), the presence of specific antibodies against Hepatitis E virus was detected. Laboratory testing was performed by non-commercial ELISA (in-house ELISA). Tests result showed that hepatitis E virus is present in wild boar population in Serbia and that the potential source of the infection for pigs and humans [8].

Bacterial agents in the wildlife in the West-Balkan region

Brucellosis

Brucellosis is a significant health problem among animals and humans in the Balkan Peninsula. In domestic animals from Croatia, the prevalence of *Brucella* sp. is low [9]. However, earlier surveys of brucellosis in domestic pigs and wild boars [10], revealed that *B. suis* biovar 2 is present in 1% of domestic pigs and in 7.5% of wild boars. This ratio is even higher (13.5% in domestic vs. 22.6% in wild boars) in areas where domestic pigs are free-ranging in the forest most of the year. *Brucella suis* biovar 3 in was isolated from horses in Croatia.

Tuberculosis

Long term control programs have almost eradicated *Mycobacterium bovis* in domestic cattle in Western and Central Europe. Yet, in the West Balkans, the situation is different. In South Bačka region (North Serbia) three tuberculous foci were detected on the territory of Žabalj, Novi Sad and Titel municipality. The first 11 reactors were detected in the year 2004. The number of estimated tuberculin reactors was 320, located in six settlements and 37 husbandries. The percent of infected animals in some herds ranged from 11.10 to 59.18%. An epizootiological evaluation revealed that the high herd prevalence and high animal incidence of bovine TBC in South Bačka region was associated with the following factors: lack of TBC diagnosis in pastured animals in swampy areas for a long period, grazing areas are often flooded resulting in bad quality fodder and cattle malnutrition, presence of other domestic animals on the pastures, co-mingling of animals from different herds is frequent, common water through, presence of different wild animal species on pasture (as a potential reservoir of the disease), uncontrolled movement and illegal trade of infected animals, natural breeding as well as overpopulation in the sheds during winter housing [43].

However, rather high prevalences of these bacteria in wild boars (*sus scrofa*) [11] wild red deer (*Cervus elaphus*), roe deer (*Capreolus capreolus*) and foxes [12] and captive (zoo) animals [13] indicate that this disease is maybe under control, but not completely extinguished. Reports exist on sporadic occurrence of tuberculosis in domestic [10], wild zoo animals [14] and humans in Croatia and neighbouring countries, i.e. *Mycobacterium* sp. was present in the environment [9] and in wild animals.

Leptospirosis

Leptospirosis is a zoonosis enzootic in Croatia and B&H [15]. This is a widely spread disease with frequent epidemic occurrence, especially among miners in B&H and foresters in Croatia [16] *Leptospira* infection was also found among European brown bears (*Ursus arctos*) in Croatia, with the serovars: *australis*, *sejroe*, *canicola* and *icterohaemorrhagiae*. A strong correlation between serovars in bears and serovars previously isolated from small mammals in Croatia was noted [17]. Different serovars of *Leptospira* spp. were also documented in red foxes (*Vulpes vulpes*), [18] red deer, roe deer, wild boars and bears [19]. These studies confirmed that in the lowland parts of Croatia leptospirosis is constantly present in wild animals, while it is sporadic in mountainous areas of Croatia.

Zoonotic parasites in the wildlife in the West-Balkan region

Echinococcosis

Echinococcosis remains serious public health problem in Southeast Europe, with the former republics of Yugoslavia reporting one of the highest prevalence rates [20, 21]. Although a decrease in incidence has been observed in some endemic areas during the last decades, new foci of animal echinococcosis emerged. Echinococcosis is zoonosis transmitted by dogs in livestock-raising areas and accidentally affects humans. There are reports from Croatia that predilection sites of hydatid cysts are in the liver (78%), followed by lungs (17%), and less frequently, spleen, kidneys, heart, bone marrow and central nervous system [20, 21].

Larval alariosis

Alaria alata, mesoecariae have been detected in muscle, glandular and fatty tissues of wild boar in various European countries [22, 23]. There are indications that this is a potential zoonotic pathogen, as humans could be infested through consumption of undercooked meat. Jaksic et al. (2002) reported a prevalence of 1.8% in 210 wild boars shot in Croatia.

Trichinellosis

Trichinellosis is an invading disease caused by nematodes of the species *Trichinella*. This important zoonosis occurs mainly in wild carnivores with cannibalistic and scavenger behaviour [25]. *Trichinella* infection cycles can be established both in the wildlife (i.e. sylvatic; involving red foxes, wolves, wild boar, bears, badgers) and in farmed animal populations (i.e. domestic). Feeding on other animals and/or their carcasses is the key factors in transmission of the parasite. Among the eleven known species of the genus *Trichinella*, four 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 [26]. *T. spiralis* has not been found in wild animals in Estonia, Italy, Norway and Switzerland, while for Greece and Portugal, there is no data.

In the majority of Southeast European countries cases of trichinellosis among the human and animal populations were described in the late 19th or early 20th century [27], also among wildlife [28]. Today, the prevalence of trichinellosis between the Balkans and bordering countries is different. A high prevalence of trichinellosis in domestic animals and humans has been reported in Bulgaria, Serbia and Montenegro, Romania and Croatia [27; 29; 30] and a moderate prevalence was found in B&H. In B&H, Serbia as well in Croatia, however, the re-emergence of trichinellosis did not only happen due to political and social changes, but also due to the war that took place in these countries during the last years of the 20th century [30, 31].

In order to reduce the incidence of trichinellosis in animals on Vojvodina territory (northern Serbia), Ministry of Agriculture, Forestry and Water Management of Serbia in late nineties provided funding for rodent control programs in the villages with the highest percentage of pigs found with trichinosis. As a general rule, deratization should be performed systematically twice a year: in spring and autumn. It must include all settlements with its direct surroundings ie. animal cemeteries, garbage dumps and canals. Action of deratization should be conducted professionally and work permanently. However, as the budget for that purpose was limited, deratization in some areas only partially executed. Thus, for example in villages Čalma and Veliki Radinci (municipality Sremska Mitrovica) after done this incomplete deratization has been a significant increase of trichinellosis in slaughtered pigs, from 0.09% in 1996. year to 0.27% in 1999. year, which is 3 times higher [42].

In the Republic of Croatia, both *T. spiralis* and *T. britovi* have been detected in wildlife. Recently, a control programme has strongly reduced prevalence in domestic swine from 3.6% to 0.0001%. However, between 30 and 50 human infections are reported every year.

In Croatia, *T. spiralis* and *T. britovi* have been detected in wild boar (*Sus scrofa* L.), as single as well as mixed infections [32]. Human infection may occur through eating inappropriately thermally processed meat, or through traditionally processed raw-fermented meats (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. This prompted for the adoption of new legal regulations which regulate certain measures for the control and prevention of trichinellosis. This involved the obligatory inspection of pork meat for slaughter for domestic purposes and also game meat for human consumption. Jaksic et al. (2002) could not detect the parasite in samples from 210 wild boars hunted in Croatia in 2001. Among 119 samples of wild boars originating from the Croatian region of Imotska krajina in 1995-2003, 2 were found positive [33]. Vučemilo et al. (2001) determined the prevalence of 0.35% in the state hunting ground Podunavlje-Podravlje in the Baranja region. These data indicate that wild boar is a latent reservoir of this parasite in the Republic of Croatia.

Brown bears (*Ursus arctos* L.) are the biggest game in biotope of Croatia. The number of bears is in the range of 400 to 600 units, and it is one of the rare stable populations in Europe.

Bears are originally carnivorous, but will also supplement their diet with plant food. Sources of animal protein may be invertebrates, but also corpses of big animals. In previous years [35], the prevalence of trichinellosis in bears was 7.04%. After the implementation of the legal obligation of examining meat and all epizootiologically important groups of game for trichinellosis in Croatia, only one bear shot in the Karlovačka County area in 2003 was found positive for *Trichinella* sp.

The badger (*Meles meles* L.) is small game prevalent from lowland to mountain regions in Croatia with the exception of the Adriatic islands. This omnivorous species is used for human consumption (mostly in northwest and eastern Croatia), and, thus, it is a potential source of human trichinellosis. Until now, there have been no recorded cases of trichinellosis originating from badger meat, most probably because meat is heat processed before consumption. Studies on badgers killed in traffic revealed that the parasite is prevalent in badgers 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 [36].

The parasite has also been recovered from wolves in Dalmatia in the period 1996 and 2007, where about 31% of 67 animals were positive for either *Trichinella britovi* (19 out of 21) or *Trichinella spiralis*. The presence of the latter species unexpected, since only *T. britovi* was previously reported in that region [37, 38].

Sylvatic and domestic cycles of trichinellosis are present also in Bosnia-Herzegovina. Human trichinellosis is usually caused by the consumption of pork from wild boars as well as from pigs. In the last 14 years more than 51 outbreaks (775 infected people) occurred, with some fatal cases [29].

So far, the red fox has been the main reservoir of sylvatic trichinellosis in Serbia, however, the increasingly important role of jackals must be noted. In one investigation in year 2011, twelve jackals were examined, all of them belonging to the genus of golden jackal (*Canis aureus*). Diaphragms were tested using artificial digestion (Commission Regulation – EC, No 2075/2005). There was relatively high prevalence of trichinellosis in jackals (8,33%) on the territory of Vojvodina with a high degree of infestation (3 larvae/g) which is much higher than in countries considered to be trichinellosis free. Jackals, foxes and rats belong to synanthropic animals and present a link between sylvatic trichinellosis and domestic animals. [45].

In Serbia, during the period 2009-2010, muscle samples were collected at Branicevo and Podunavlje area (in East Serbia, near Danube river and Romania border) from 167 wild animals; 94 wild boars (*Sus scrofa*). Muscle samples were analyzed by artificial digestion. Genotyping was performed by multiplex PCR. *Trichinella* spp. infection was detected in 11 (11,7%) wild boars. *Trichinella spiralis* and *Trichinella britovi* were only two species identified in the isolates as single or mixed infection. *Trichinella britovi* was identified in 31% of isolates, and *T. spiralis* in 53%, and mixed infection in 16% of samples [39]. It should be in mind the importance of legislation related to suppression of trichinellosis. According to the strategy of Serbia's accession to the European Union, and the commitment to harmonization of regulations related to human and animal health and health safety of food of animal origin, there is space for correction and supplement of legislation relating to swine trichinellosis [46].

Conclusion

Unfortunately, no recent (i.e. from the last 10 years) literature data could be found on the following diseases or agents in wild animals in Serbia, Bosnia & Herzegovina (B & H) and Croatia: Listeriosis, Q-fever, Aujeszky's disease, Salmonellosis, Cysticercosis. Also, there are no published data about these seroprevalence in exposed humans (hunters, farmers, veterinarians) against the respective infectious agents. Compared to data on *Trichinella* sp., reports on other parasitological, or bacterial and viral zoonoses are rather scarce.

Regarding the rabies in Serbia, the ultimate goal of control of the rabies in wild animals using the oral vaccination is progressive reduction of occurrence nature reservoirs of the rabies virus to its complete eradication and achieving the status of a country free from rabies. The results obtained after three oral vaccination campaigns (autumn 2010, spring 2011, autumn 2011) were correlated with results from other countries, especially those shorter time implemented oral vaccination (Bulgaria). Number of rabies cases in domestic and wild animals in Serbia is in the progressive decline as a result of effective implementation of oral vaccination, but the ultimate success requires long-term, sustained efforts and cross-border cooperation with neighboring countries, with continuous monitoring of the implementation and surveillance of rabies. Thus, disposal of oral vaccination in neighboring countries can affect the results of eradication program of rabies in wild animals in Serbia. There is a constant threat of a rabies in border areas due to the direct exposure of animals that inhabit areas where vaccination is carried out (and where the number of foxes is increasing because of immunization against the rabies) and animals will come from areas where rabies is endemic phenomenon, and where not conducted oral vaccination. In addition, the total duration of vaccination of wild animals in Serbia will also depend on effectiveness of oral vaccination carried out by other countries in the region, and which have this program started after Serbia (Croatia, B & H, Macedonia, Montenegro), or have not yet started (Albania) and surveillance of rabies in the wider Balkan region.

Effective control of zoonotic diseases will require more efforts in studying the role of wildlife as a reservoir for zoonoses. This includes epidemiological studies as well as ensuring effective meat inspection of game, implementation of safe game handling and evisceration techniques and safe disposal of offal. The latter requires consciousness and compliance of hunters, which has to be based on training and motivation.

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