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Invited lecture

GAME MEAT SAFETY – WILD BOARS

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Abstract

Wild animal meat harvesting and processing is significantly different from classical livestock meat production and represents a challenge by itself. Implementation of the concept "from forest to fork" encompasses epidemiological difference between wild animals and livestock, influence of hunting ground ecology, type of hunting, field evisceration of hunted game, meat inspection after shooting or transport etc. The objective of this paper was to identify biological and chemical hazards important for wild boar meat safety. Several hazards were analysed: *Mycobacterium spp., Salmonella spp., Trichinella spp.,Toxoplasma spp*., *Alaria alata*, microelements, and heavy metals. Two criteria were established for hazards identification: evidence of shared pathogens presence in wild boar population in specific geografical region and evidence of hazardeous pathogens spread during handling, processing and consumption of wild boar´s meat. The research on the presence of food borne pathogens in wild boar´s meat is still scarce, while the *Trichinella spp.* life cycle is distinctly described and there are relevant data about epidemiology and natural reservoirs of the parasite in this part of the Europe, however little is known about other food borne pathogens in wild boar population. The presence of *Salmonella spp*., *Toxoplasma gondii*, *Alaria alata* is identified in wild boar population in our region. The analysis of the results of the microelements and heavy metals in wild boar meat has shown that no samples exceeded legally set limits, but our results suggest that pollution of the biosphere with chemical contaminants should be systematically monitored to identify potential increasing contamination tendencies. Programme for monitoring and control of game meat safety should include control measures for live animals, control measures during hunting and after shooting, guidelines for official meat inspection, control measures for carcass processing and surveillance of chemical residues.

Keywords: meat, wild boar, food borne pathogens, microelements, heavy metals

Introduction

Wild boar (*Sus scrofa*) distribution covers the entire continental Europe; it is missing in British Isles, Scandinavia and northern part of the European Russia. Wild boars in Serbia are native and very abundant big game species. Excessive and irresponsible hunting resulted in demographic decline in early 20th century. However, thanks to substantial changes of agricultural practice, reduced number of natural enemies as well as game feeding policies in last four decades the game population has recovered and is still increasing, not only in Serbia but also around Europe (Rippa et al., 2012).

Game meat production chain is substantially different from the conventional production of meat from domestic animals, thus presenting an unique challenge. The concept “from forest to fork” encompasses the effects of hunting ground ecosystems, type of hunting/shooting, evisceration in the field, meat inspection after slaughtering, transportation etc. The risk assessment regarding alimentary pathogens in wild boars implies elucidation of some basic issues, i.e., identifying the epidemiological difference between domestic and wild animals and determining the criteria for evaluating the safety of wild game meat.

When speaking of wild animals, the disease control measures encompass the control of the diseases that are transmissible from wild to domestic animals or directly to humans. Potential negative environmental impact of measures and actions taken to the purpose of disease control is a specific issue within the program of wildlife disease control.The opinion that complete eradication of infectious and zoonotic diseases among wild animal population is virtually impossible is nowadays widely accepted.

Domestic animals are raised for food and food production in controlled conditions, thus undergoing the range of measures for health status control and *ante* and *post mortem* examinations at slaughter. Contrary to that, game undergoes only a post mortem examination. In that respect, information on certain diseases or mortality rates is lacking as well as the veterinary treatment, specific laboratory testing, etc., thus restricting the proper evaluation of animals' health status and prevention of their entrance into the food chain.

The food safety criteria for assessing the safety of wild game meat are still lacking. They represent a complex food-safety issue, since the majority of criteria relies on the average daily food intake, which is highly specific in wild animals. The majority of the population consumes minor amounts of game meat, but small population consumes pretty large quantities of game meat. The average consumption of game meat among hunters' families is estimated to be some 4kg meat per person, i.e., family member (Ramanzin i sar., 2010).

The objective of this paper was to identify biological and chemical hazards of importance for wild boar meat safety. Moreover, control measures for particular hazards are described, too.

Hazard identification and control options

Our identification of hazards of importance for wild boar meat relied on two main criteria. Is there evidence for hazard transmission during handling, preparing and consuming of wild boar meat and is there evidence of the presence of the pathogen among the game population in our region? The following hazards were categorized:

- High-priority hazards: *Mycobacterium spp*., *Salmonella spp*., *Trichinella spp*. *Toxoplasma gondii*
- Low-priority hazards: chemical hazards, *Alaria alata*
- Hazards of undefined priority, i.e. hazards that still lack sufficient data *Campylobacter spp. Y. enterocolitica*, pathogenic verotoxic *E. coli*

The guidelines of Codex Alimentarius Commission (CAC, 2005) put the emphasis on hygiene and inspection surveillance of hunted game at the primary stage of meat production chain (including transport) as the critical points for game meat control. The recommendations are implemented through Directive Regulations (EC) 853/2004 and 854/2004.

The measures are basically distributed into two groups:

- identification of the diseases and all major changes by visual inspection (in case of apparent pathological changes, severe contamination from the environment or suspected specific biohazard, additional laboratory testing is recommended)
- application of practical skills and knowledge aimed at preventing the spread or increase of biohazards (such as *Salmonella spp*.) on/in edible tissues

Mycobacterium bovis

M. tuberculosis is considered the most important biological hazard in large game. Among the wildlife population of Europe, three animals are considered major hosts of tuberculosis: badger, wild boar and deer from the subfamily *Cervinae.* Wild boar can act as a direct source of human

tuberculosis infection, as well as the reservoir of infection in domestic animals (Gortazari i sar, 2008). Transmission of tuberculosis to humans occurs mainly by inhaling infectious aerosols, consuming raw milk and, though somewhat less frequently, by consuming meat products originating from infected animals. Some literature reports described potential infection routes from wild boars to humans, including contact via the raw meat (Ashford et al., 2001).

Some 90% of known-infected wild boars with tuberculosis manifest calcified granulomatous lesions in the mandibular lymph nodes. Moreover, 60% of these animals have generalized tuberculosis, i.e., lesions in multiple organs, predominantly in the head, thorax and abdomen. However, even generalised tuberculosis in wild boar rarely causes visible loss of body condition (Martín-Hernando et al., 2007). Hence, tuberculosis in wild boars is not manifested by specific clinical picture and only the pathoanatomical examination can reveal suspect disease.

Wild boar tuberculosis has been reported in the last decades in at least 10 European countries: Bulgaria, Croatia, France, Germany, Hungary, Poland, Portugal, Slovakia, Spain and the UK (Gortázar et al., 2012). Wild boar experience much higher levels of exposure than deer (Vicente et al., 2006). *M. bovis* prevalence in wild boar ranged from 46 to 52 % in three different surveys in the Iberian Peninsula (Gortázar et al., 2008), where this host is considered the main carrier of wildlife tuberculosis and a key factor in cattle tuberculosis eradication (Naranjo et al., 2008). In the region of South Bačka, three endemic foci of tuberculosis were recorded located in municipalities of Žabalj, Novi Sad and Titel. The percentage of infected animals in individual herds ranged between 11.10% and 59.18% (Pušić et al., 2007; Pušić et al., 2009; Pušić et al., 2013). So far, relevant data on the presence of tuberculosis in wild boars in this region are not available and the research is ongoing. The occurrence of tuberculosis in cattle was recorded in the village of Kovilj (the territory of Novi Sad municipality), which suggests potential contacts between wild and domestic animals and thus presence of tuberculosis in wild game.

Eradication programmes in domestic animals rely on annual diagnostic examination of cattle using the method of intradermal tuberculinization reaction, computed records and animal tracking, testing positive herds and those who were in close contact with them at short intervals, and mandatory slaughter of positive animals (Pušić et al., 2008; Pušić et al., 2009a,b,c). When speaking of wild animals, the surveillance is practically impossible, so meat inspection is the only potential current source of information and measure for control of tuberculosis.

Trichinella spp

Trichinellosis is endemic in almost all European countries, affecting also the regions of Srem and river valeys of Danube, Drina and Kolubara in Serbia (Petrović et al.,2012a). The analysis of recorded trichinellosis epidemics in Vojvodina in the period 2001-2011 identified domestic pig as the main reservoire of Trichinella. The infection is commonly acquired by consuming raw or undercooked meat infected with living Trichinella larvae. Documented cases of human infections after consuming wild boar meat were reported in Serbia (Urošević et al., 2013). In Europe, wild animals are considered the main reservoir of Trichinella, which makes the eradication of the infection impossible in spite of its pretty low prevalence in wild game (Rafter et al., 2005).

According to Petrović et al. (2012 a, b, c), high incidence of trichinellosis was established in the territory of Vojvodina among several animal species such as jackals (7.89%), foxes (4.76%) and wild boars (0.53%). In countries in which trichinellosis of domestic animals has been fully eradicated, such as Danemark, the prevalence of sylvatic trichinellosis is extremely low (0.001%) (Enemark et al., 2000). Moreover, an average infection rate in carnivores in Vojvodina (3 larvae/10g) is significantly higher than that recorded in Danemark (1 larva/10g). In some cases, extremely high infection rates has been established in wild boars in Vojvodina region, ranging even up to 1100 larvae/g (Petrovic et al., 2013a, b). High incidence of sylvatic trichinellosis in some

geographic regions poses substantial risk of infection spreading to domestic pigs grazed in sylvatic habitats. The potential transmission routes of *Trichinella* spp. in pigs include canibalism, ingestion of synanthropic and sylvatic animals as well as of the faeces of pigs that have been infected some 1-2 days earlier (Petrović et al., 2014).
According to Petrovic et al. (2014), life cycle of *T. spiralis* in Vojvodina region includes circulation from domestic pigs to wild boars and *vice versa,* which is assotiated with specific behaviour of this animal species. Wild boars are very tolerant to the presence of humans, often commingling with domestic pigs on common pastures and have access to laystall and food waste. Improper disposal of pig carcasses and offals in the field, is the greatest risk factor for trichinellosis maintenance and spread within pig population.

The diaphragms of all killed wild boars must be examined by artificial digestion method, for estimating the presence of *Trihinela spp* larvae. This is the most effective measure for the control of meat safety when speaking of this pathogen. However, it should be emphasized that appropriate disposal of meat originating from infected animals represents an important step in preventing further spreading of trichinellosis.

Salmonella spp.

Salmonella has long been recognised as an important zoonotic pathogen of economic significance in animals and humans. Human salmonellosis is usually characterised by the acute onset of fever, abdominal pain, nausea and sometimes vomiting. Symptoms are often mild and most infections are self-limiting, lasting a few days. The common reservoir of *Salmonella* is the intestinal tract of a wide range of domestic and wild animals, which may result in a variety of foodstuffs of both animal and plant origin becoming contaminated with faecal organisms either directly or indirectly (EFSA, 2013).

Finding of *Salmonella spp* in wild boars has traditionally been associated with *S. Typhimurium;* however, in the past few decades, the spectrum of serotypes isolated from carcasses, tonsils, faeces and lymph nodes is much more diverse. Great differences in the prevalence of *Salmonella spp*. in game were reported between individual species (e.g., the rates are higher in wild boars than in ruminants) as well as between particular regions (e.g., higher prevalence rates were recorded in southern countries of the EU) (Table 1). According to data reported by EU MSs in the framework of the Zoonoses Directive (2003/99/EC) in 2004–2011, 1.1 % of deer, 11.1 % of reindeer, 18.3 % of wild boar, 1.8 % of ostrich and 2 % of rabbit faecal samples were positive for this organism. *Salmonella* was therefore shortlisted for risk ranking (EFSA, 2013)

Investigation of wild game pathogens in our country were mainly aimed at wild birds and other enteropathogens (Stojanov et al., 2012; Velhner et al., 2012). The prevalence of salmonellas on wild boar carcasses is relatively low, i.e. below 10% (unpublished data). Wild boars are more frequently the carriers of *Salmonella spp*. than wild ruminants. Thus, an inadequate evisceration and/or bad shot (e.g. shooting wound in abdominal region) increases the risk of meat contamination with *Salmonella spp*. (Wisniewski, 2001).

Table 1. Prevalence of *Salmonella* spp. in wild boars in some European countries (Paulsen et al., 2012)

Animal species and sample	Country	Number of samples	Number of positive samples
Wild boars, faeces	Italy	2365	441 (18.7%)
	Portugal	77	17 (22.1%)
	Switzerland	73	4 (5.5%)

There are few practical options for eradication of *Salmonella* and other zoonotic diseases in wildlife, with the possible exception of vaccination or culling in geographically isolated areas. Regulating animal density (to avoid crowding and overabundance), can contribute to disease control; also, when game is eviscerated on the spot, the offal should be removed from wildlife (Gortazar et al., 2006). However, there are no records indicating that such measures would have successfully eradicated *Salmonella* in the wild.

In actual scientiffic literature there are no published recommendations (at least to our knowledge) on *Salmonella spp*., control measures in live wild boars. Therefore it is important to control hygiene during hunting, evisceration, bleeding and cleaning of carcasses, cooling and transportation, coupled with sampling of processed carcasses for *Salmonella spp*. presence.

Toxoplasma gondii

T. gondii infection is common in animals and humans. *T. gondii* is an obligate intracellular protozoan parasite. Nearly all warm-blooded animals can act as intermediate hosts, and almost all animals may be carriers of tissue cysts of this parasite. However, the parasite matures only in domestic and wild cats, which are the definitive hosts (EFSA, 2013). In pregnant women, the parasite can cause congenital infections resulting in abortion, stillbirth, mortality and hydrocephalus in newborns. The parasite can also cause severe disease in immune compromised individuals such as organ graft recipients and individuals with AIDS or cancer (EFSA, 2013). In immune-competent individuals, 80–90 % of cases of *T. gondii* infection are asymptomatic and the majority of the remainder result in only mild, self-limiting symptoms.

Common infection routes for wild boars include ingestion of oocysts from the environment, ingestion of infected rodents and birds or cannibalism (Tenter et al., 2000). An interesting observation was reported by Dubey et al (1992) suggesting significantly higher seroprevalence of *Toxoplasma* among pigs raised outdoor than in those reared in conventional settings. The natural production system is conducive for exposure of the pigs to various known risk factors for *Toxoplasma* infestations such as cats (the definitive host) and other species that can be harbouring cysts in their musculature as compared to the indoor conventional production system. In addition, the outdoor environment with open access to soil, vegetation, and moisture allows viable environments for *Toxoplasma* oocysts. The similar trend of higher seroprevalence of *Toxoplasma* and *Trichinella* in outdoor-reared swine was reported in the Netherlands (van der Giessen et al., 2007). The frequency distribution of *Toxoplasma* seroprevalence reported in Dubey et al (1992) study ranged from 0.38% in intensive production to 5.62% in outdoor-reared system.

T. gondii is common in hunted wild boars in EU, where the seroprevalence has been reported to vary between 8% and 38% (Lutz, 1997; Gauss et al., 2005; Antolova et al., 2007). The seroprevalence in farmed wild boar has been reported to be 33 % (EFSA, 2007). Given the high incidence in wild boar, this organism is significant hazard for wild boar meat safety.

Toxoplasmosis in animal commonly takes an asymptomatic course, which makes the diagnostics in living animals impossible. To detect *T. gondii* infection in livestock, serologic assays can be applied. With the exception of cattle, the presence of antibodies and tissue cysts is assumed to correlate well. Meat or tissue samples can be tested by bioassay or PCR. *T. gondii* oocysts cannot be differentiated from *Hammondia* or *Neospora* oocysts morphologically, thus molecular techniques need to be applied. If infected meat is consumed without prior freezing or proper heating (core temperature over 67 °C) *T. gondii* can be transmitted. Salting, fermenting, drying, and smoking also reduce tissue cyst viability, but the exact conditions needed to inactivate *T. gondii* are less well-established (Opsteegh et al., 2013).

Alaria alata

Alaria alata is a trematode parasite, and the transmission of this parasite occurs when humans eat undercooked game or frog meat infected with the mesocercarial stage of this parasite. The epidemiology of *Alaria* infection is not well-understood (Moehl et al., 2009a). The reported cases of human larval alariosis are most likely due to mesocercariae from *Alaria* species other than *A. americana*, but primates can be infested by *A. americana* (Moehl et al., 2009b). A study in Germany (Riehn et al., 2012) found a high prevalence of *A. alata* in wild boar (11.5%). Although specific methods for targeted detection of *A. alata* are available, its diagnosis is feasible during the official *Trichinella* inspection in the competent veterinary inspection offices. Recent studies conducted in the eastern parts of Austria indicated an overall prevalence of *A. alata* mesocercariae in wild boar of 2%, when lean muscle (*M. masseter)* tissue was tested (Sailer et al., 2012) or 6.7%, when a muscle – fat tissue mixed sample was tested (Paulsen et al., 2012).

Jakšić et al. (2002) and Grosse and Wüste (2006) pointed out that the parasite represents a potential source of infection for both humans and animals, and that consumption of wild boar meat can be an important factor in the epidemiology of this zoonosis (Moehl et al., 2009b). However, to date, there has been no report on human alariosis cases due to consumption of wild boar meat and thus *Aaria alata* was ranked as hazard with low priority.

Chemical hazards

EFSA (2013) ranked chemical residues and contaminants on the basis of bioaccumulation, toxicological profile and likelihood of occurrence, and they took into account the findings from the NRCPs for the period 2005–2010. The ranking results were as following:

- No substances were classified in the high potential concern category for game.
- Within the category of medium potential concern for farmed game is cadmium.
- All other substances listed in Council Directive 96/23/EC was ranked as being of low or negligible potential concern
- Potentially higher exposure of consumers to these substances from game meat takes place only incidentally, as a result of mistakes or non-compliance with known and regulated procedures.

The analysis of the results of the microelements and heavy metals in wild boar meat in Serbia (non published data), has shown that no samples exceeded legal limits, but our results suggest that pollution of the biosphere with chemical contaminants should be systematically monitored to identify potential increasing contamination tendencies.

Conclusion

The absence of characteristic clinical picture is common to all aforementioned hazards, thus a thorough post mortem examination for the presence of tuberculosis and artificial digestion to confirm the presence of *Trichinella* but also *Alarai alata* is of outmost importance. However, some hazards are not detectable by these procedures (*Salmonella, Toxoplasma*, chemical hazards) and designing and implementation of appropriate hazard control programs is necessary.

The presence of alimentary pathogens and contamination of boar meat with chemical hazards in Serbia has not yet been fully elucidated. While relevant data on *Trichinella spp* are available and the mechanisms of the maintenance of its life cycle in our region are well known, the data on other hazards are still unclear. A number of ongoing research-scientific projects in our country address the prevalence of selected hazards among wildlife population with an aim of obtaining

scientifically-based risk analysis as a corner-stone for implementing appropriate programmes and measures for the control of alimentary hazards. The programme for control and monitoring of wild boar meat should encompass control measures for live animals, control measures during and after hunting, guidelines for veterinary inspection of meat, control measures for carcass processing and monitoring over chemical residues.

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