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SHORT COMMUNICATION

The prevalence of American liver fluke *Fascioloides magna* (Bassi 1875) in red deer from Croatian hunting grounds

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Abstract From October 2002 till April 2003, 194 feces samples and 28 liver samples of red deer shot in the Republic of Croatia territory were examined to determine the prevalence of *Fascioloides magna* in this game population. The majority of study samples were obtained from animals originating from the east part of Croatia, and only a minor proportion from animals originating from the central and west parts and littoral of Croatia. *F. magna* eggs were detected in 67/194 (34.53%) feces samples and *F. magna* adults in 8/28 (28.57%) liver samples. The majority of invaded red deer originated from the east part of Croatia, Baranya region, where *F. magna* eggs were found in 64/120 (53.33%) and *F. magna* adults in 8/15 (53.33%) animals. *F. magna* eggs were detected in only 3/74 (4.05%) animals originating from the other parts of Croatia.

Keywords *Fascioloides magna* · Prevalence · Red deer · Croatia

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Introduction

Fascioloides magna (Bassi 1875), a liver trematode, originates from North America. The North American white-tailed deer (*Odocoileus virginianus*) is the main natural definitive host of this parasite, from which it has spread to another 13 wild and domestic animal species. There are primary final hosts of *F. magna*, which include red deer (*Cervus elaphus*), white-tailed deer (*O. virginianus*), and fallow deer (*Dama dama*) and dead end hosts including wild and domestic cattle, sheep, and goat. Initial reports on the spread of *F. magna* in Europe appeared in 1875, when it was detected in an Italian hunting ground (Pybus 2001), followed by reports on the occurrence of *F. magna* in Germany (Salomon 1932), Czechoslovakia (Ullrich 1930), Poland (Ślusarsky 1955), Austria (Pfeiffer 1983), Slovakia (Rajský et al. 1994), Hungary (Majoros and Sztojkov 1994), and Croatia (Marinculić et al. 2002.; Janicki et al. 2005). *F. magna* is spreading from one site to other uninfected areas by natural migration of infected hosts or artificially via animal translocation (Pybus 1990). *F. magna* dissemination over Europe was rather slow and generally followed the course of the Danube basin (Erhardová-Kotrlá and Kotrlý 1968; Pybus 2001). In some hunting ground in the former Czechoslovakia, the game is invaded by 70–80 p.c. with *F. magna*, which made it the European region with the highest rate of *F. magna* infection (Erhardova 1961). Regional variation in the spread of *F. magna* and efficacy of the use of anthelmintics in red deer and other species of deer were investigated by (Chroust and Chroustová 2004) in Czech Republic, by (Rajský et al. 2002) in Slovakia, and by Ursprung et al. (2006) in Austria.

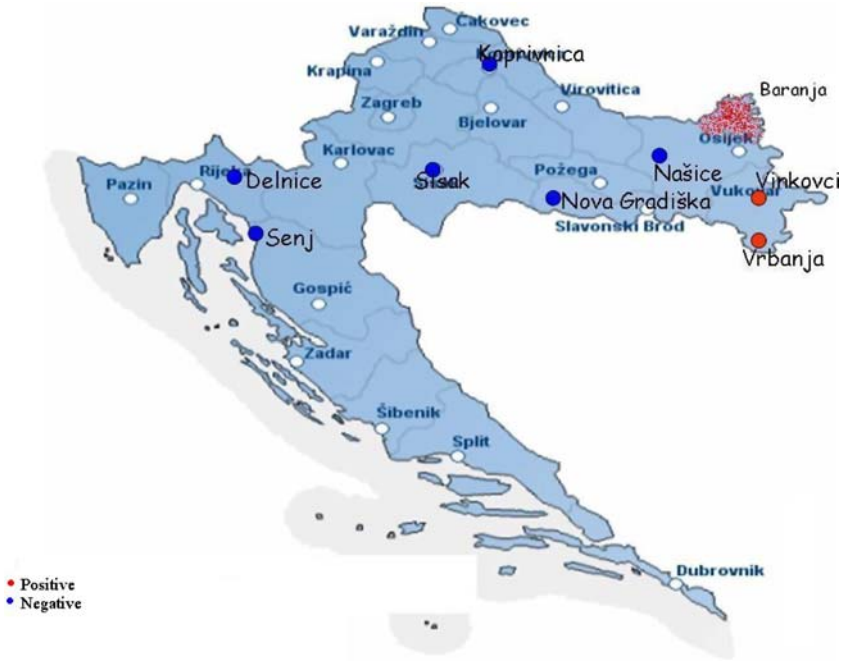

Fig. 1 Distribution of *F. magna* in Croatia (*red circles*, positive; *blue circles*, negative)

The aim of the present study was to assess the prevalence of *F. magna* in red deer in Croatia by the analysis of feces and liver samples obtained from red deer shot in different parts of Croatia.

Materials and methods

The entire territory of Croatia in its inland parts and littoral provides appropriate conditions as a habitat for deer. The red deer population in Croatia has been estimated at 9,641 (Trohar and Ozimec 2005). In Baranya, the east part of Croatia (sharing border with Hungary), the estimated number of red deer is about 2,000. Baranya is the leading area of hunting tourism in Croatia; therefore, the majority of study samples originated from this region.

Twenty-eight liver samples and 194 feces samples obtained from red deer shot at nine grounds during hunting season from October 2002 till April 2003 were analyzed to assess the occurrence and spread of *F. magna* in red deer in Croatia. The greatest proportion of study samples were from the red deer shot in Baranya (east Croatia), characterized by underground water and marsh-land favoring the development of the intermediate hosts and at the same time serving as a deer habitat. The rest of samples were collected from red deer in the hunting grounds around Vinkovci and Vrbanja, both near Baranya; in the central part of Croatia around the towns of Nova Gradiška, Našice, Sisak, and Koprivnica; in Gorski Kotar around Delnice; and in the coastal area around Senj. The number of study feces and liver samples from particular locations (hunting grounds) ranged from 1 to 120, including various age groups. The entire study area has continental climate characterized by cold winter and warm summer. All rectum feces samples and liver samples obtained from shot red deer were labeled with number, date, and place of shooting. All liver and feces samples originated from different animals.

The classic method of sedimentation was used to detect and determine *F. magna* eggs in feces, and the usual method of parasite isolation to identify *F. magna* adults in liver samples. The data thus obtained were processed by

Table 1 The spread of *Fascioloides* (*F.*) *magna* in Croatia hunting ground according to red deer feces and liver analyses

Area	Number of red deer invaded with *F. magna*					
	Feces*			Liver		
	n^a	$+^b$	%^{c}	n^a	$+^d$	%^{c}
Baranya	120	64	53.33	15	8	53.33
Vrbanja	21	1	4.76	–	–	–
Vinkovci	16	2	12.5	3	0	0
Nova Gradiška	6	0	0	7	0	0
Našice	12	0	0	1	0	0
Koprivnica	5	0	0	–	–	
Sisak	–	–	–	1	0	0
Delnice	14	0	0	–	–	
Senj	–	–	–	1	0	0
Total	194	67	34.53	28	8	28.57

* $p<0.001$
[a] Number of samples analysed
[b] Number of samples containing *F. magna* eggs
[c] Percentage of *F.-magna*-positive samples
[d] Number of samples containing *F. magna* adults

methods of descriptive statistics. Statistical analysis of differences in the parasite *F. magna* prevalence between different geographical areas was done by use of Fisher's exact test.

Results

F. magna eggs were detected in 67/194 (34.53%) feces samples, and *F. magna* adults in 8/28 (28.57%) liver samples (Table 1, Fig. 1). The highest proportion of feces samples positive for *F. magna* eggs were recorded in red deer originating from Baranya (64/120; 53.33%). Among 74 feces samples of the red deer shot in other areas, *F. magna* eggs were detected in three samples, including 2/16 samples originating from the Vinkovci surroundings and 1/21 sample from Vrbanja. Differences in the prevalence of *F. magna* between different areas were statistically significant (χ^2=49.9444, p<0.001). *F. magna* adults were only found in 8/15 (55.33%) liver samples of the red deer shot in Baranya. The analysis of feces samples revealed 23/70 (32.85%) and 44/124 (35.48%) invaded animals in 2002 and 2003, respectively. In addition to *F. magna* eggs, eggs of the trematode *Paramphistomum* sp. were identified in 65/194 (33.50%) red deer feces samples.

Discussion

In Croatia, the occurrence of the large American fluke *F. magna* (Bassi 1875) in red deer (*C. elaphus*) was first suspected in 1999 and initially confirmed in 2000 (Marinculić et al. 2002).

In the present study, *F. magna* eggs were found in one third of feces samples and *F. magna* adults in one third of liver samples, indicating a high rate of red deer invasion with *F. magna* (Bassi 1875). Coproscopic studies revealed about a half of *F.-magna*-positive animals to originate from east Croatia (Baranya) and only occasionally from the other two hunting grounds near Baranya. *F. magna* adults were also exclusively detected in the red deer from Baranya (Table 1). These results pointed to great variation in the rate of invasion between the east part and other regions of Croatia. Similar findings on invasion with *F. magna* have been reported from Czech Republic, Slovakia, and Austria, with prevalence ranging from 0% to 95% in various regions (Chroust and Chroustová 2004; Rajský et al. 2002; Ursprung et al. 2006). Marshland, mildly alkaline areas reticulated with shallow warm streams, favor the development of snails as intermediate hosts while being the favorite habitat of cervids; therefore, *F. magna* is by far more commonly found in these areas than at high uplands (Pybus 2001). These conditions are characteristic of Baranya, where a high rate of red deer invasion in contrast to other parts of Croatia was recorded, suggesting the area to be a focus from which the infection may have spread to other parts of Croatia. The timing and route of *F. magna* transfer to the area of Baranya are difficult to estimate. Erhardová-Kotrlá and Kotrlý (1968) believe that the interval between the arrival of new animals in a new habitat and large-scale invasion may take decades due to the fluke adaptation to the overall life conditions. In our case, the parasites were probably transferred by natural migration of infected deer from the neighboring Hungary (Marinculić et al. 2002). The presence of conditions favoring the development of snails as intermediary hosts in the area was indicated by the finding of the trematode *Paramphistomum* sp. eggs in the red deer from Baranya and Vrbanja whereas intermediate host for the trematode *F. magna* has not been identified in Croatia to date. In European countries, *F. magna* was found to have adapted to different snail species, the most appropriate host being *Galba truncatula* (Erhardová-Kotrlá 1971).

The experience from former Czechoslovakia (Erhardová-Kotrlá and Kotrlý 1968) shows the long-term presence of *F. magna* to have decimated the cervid population in the region. Among wild animals, roe deer is highly vulnerable as aberrant host (Pybus 2001), where the presence of only four *F. magna* flukes may cause death (Erhardová 1961). In enzootic areas, wild animals serve as a reservoir of *F. magna* for domestic animals, which may cause great economic losses, especially in cattle (Balbo et al. 1989), sheep, and goat industry (Pybus 2001).

According to Braun et al. (1995), the method of sedimentation has a sensitivity of 68% in the detection of *Fasciola* eggs, while interpretation can only be done in the patent period. For this reason, more precise data on the prevalence of *F. magna* in deer could only be obtained by more extensive feces and liver sampling over a prolonged period of time.

Accordingly, the spread of *F. magna* among wild animals should be monitored and their contact with domestic animals in terms of common grazing should be prevented in invaded areas.

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