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OCCURRENCE OF FASCIOLOIDOSIS IN RED DEER (*CERVUS ELAPHUS*) IN BARANJA REGION IN EASTERN CROATIA

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SUMMARY

Fascioloidosis is a parasitic disease caused by the giant American liver fluke Fascioloides magna (Bassi, 1875). In Croatia, the first report of this disease was in January 2000, in red deer (Cervus elaphus* L.*) from the Tikveš Forestry in Baranja region (east Croatia). The aim of this survey was to determine the geographical distribution of fascioloidosis and the infection prevalence in deer.
The survey was carried out in six state hunting grounds that manage with deer game in Baranja region during 2001 – 2004. Parasitological examinations were carried out by qualitative and quantitative faecal exams. The highest prevalence's (35 – 60%) were found in epizootic focuses of two hunting grounds at flooding – bog land area in east Baranja, Danube forestry. The mean intensity of infection, determined on the basis of the number of eggs per gram (EPG) was 30 – 33 EPG (range 1 – 300). High 86% of examined samples was in category to 50 EPG. The highest prevalence and the biggest EPG number too, were determined during the first year of survey. In the Baranja area fascioloidosis represents a potential danger for other game species, mainly roe deer and wild boars, as for domestic animals.

Key-words: Fascioloides magna, fascioloidosis, red deer, Cervus elaphus, Croatia

INTRODUCTION

Fascioloidosis is a parasitic disease caused by giant American liver fluke *Fascioloides magna* (Bassi, 1875). *F. magna* is an important trematode which primary occurs in the liver of a variety of wild and domestic ruminants.
F. magna is prevalent in the North America in cervids on all USA territory and south Canada (Mulvey, 1991). In Europe, it was found for the first time in deer in Natural Park La Mandria near Turin, Italy (Bassi, 1875). The parasite was introduced in 1865 with natural infected wapiti deer from North America. The infections has been retained in this area until now with time-varying prevalence.
During the first decades of twenty century, the parasite was found in red deer from the former Czechoslovakia, with prevalence's ranging 70-80% (Erhardova-Kotrla, 1971). Furthermore, sporadic cases were found in Poland and Germany (Salomon, 1932; Slusarski, 1955). At the end of twenty century, enzootic areas of fascioloidosis in cervids were also found in Austria, Hungary and Slovakia (Rajsky et al., 1994; Majoros and Sztojkov, 1994; Špakulova et al., 1995).
Based on migration paths of game, especially red deer, it was expected that spreading of this parasitic disease will, sooner or later, expand into Croatian territory. In January 2000, *F. magna* liver flukes were found during liver examination of shot red deer from Baranja region (Marinculić et al., 2002). Since then, the parasite was never found in Croatia and its introduction has severely damaged the health status of cervids. Furthermore, the disease is potentially dangerous for domestic livestock, also.
Our survey started in 2001 and was carried out by faecal examinations of deer from areas of epizootic focus, followed by examinations of other hunting grounds in Baranja which manages with deer game. The aim was to determine the geographical distribution and the prevalence of the infection.

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MATERIAL AND METHODS

From 2001-2004, faecal samples of red deer *(Cervus elaphus)* were collected in 6 state hunting grounds (altitude range 82-101 m/sl) in Baranja region, in east part of Croatia. In the hunting ground XIV/9 "Podunavlje-Podravlje" (area of 25.533 ha), where the first Croatian case of fascioloidosis was found, samples were collected in 15 locations. In hunting grounds XIV/10 Munjoroš (2.400 ha) and XIV/11 Šarkanj – Vrblje (1.377 ha), samples were collected in 3 locations, and in hunting grounds XIV/3 Haljevo (1.681 ha), XIV/5 Koha – Kozarac (1.293 ha) and XIV/10A Podravlje (8.410 ha) samples were collected on 2 locations per each hunting ground. Most samples were collected from areas most associated with game, such as feeding areas.
A sedimentation technique for detection and quantification of *F. magna* eggs was carried out to determine fluke egg counts from collected faecal samples (Thienpont et al., 1979). Eggs were counted in Petri dishes under microscope (magnification 100 X and 200 X). Arithmetical mean of eggs per gram (EPG) and prevalence for each locality were accounted. The prevalence was calculated as a percentage of positive samples from collected samples.
The statistical analysis was carried out by a computer program STATISTICA (StatSoft 7.1).

RESULTS AND DISCUSION

Mean EPG counts sorted per year, number of faecal samples and number of positive faecal samples are shown in Table 1.

Table 1. Prevalence of *F. magna* infection in red deer (*Cervus elaphus*) and EPG in the examined localities

Hunting Ground	Year	Examined sample	Positive Sample	Prevalence (%)	Mean EPG ± SD
Podunavlje – Podravlje	2001	247	129	52.23	51.01 ± 51.84
	2002	300	151	50.33	32.97 ± 23.64
	2003	300	139	46.33	23.91 ± 14.11
	2004	300	133	44.33	25.69 ± 13.35
Šarkanj – Vrblje	2001	15	7	46.67	47.43 ± 27.39
	2002	60	36	60.00	46.08 ± 33.93
	2003	60	26	43.33	20.88 ± 11.68
	2004	60	21	35.00	10.87 ± 6.03
Koha – Kozarac	2001	4	0	0	0
	2002	30	0	0	0
	2003	30	0	0	0
	2004	30	2	6.67	2 ± 0
Munjoroš	2001	5	0	0	0
	2002	45	0	0	0
	2003	45	1	2.22	1 ± 0
	2004	45	2	4.44	2 ± 0
Podravlje	2001	0	0	0	0
	2002	30	0	0	0
	2003	30	4	13.33	5.50 ± 2.88
	2004	30	4	13.33	3.25 ± 1.50

The highest prevalence of fascioloidosis was found in hunting grounds XIV/9 "Podunavlje-Podravlje", where the first case of fascioloidosis in Croatia was found, and in hunting ground XIV/11 Šarkanj – Vrblje, with slightly decreasing trend during four years period. Both hunting grounds were located in east Baranja region, in flooded forestry area near the Danube River.
In accordance to their biological and ecological characteristics, both areas form a continuous, top quality, biotope for deer game with the highest abundance of animals. Because of daily and seasonal deer moving, the number of animals is quite constant, which is of great importance for the assessment of epizootiologic situation (Cvetnić, 1993). Therefore, the highest prevalence in those hunting grounds

can be explained based on their continental and hydrological connections per Danube. However, the management of the hunting grounds is often different. For instance, in some case deer game is treated with triclabendazole while in other grounds deer are untreated. The similar prevalence in both hunting grounds probably implies that deer game get in to contact with mentioned medicine during the year on certain spots where it was mixed with pelleted feed and left. But still, the more presumptive reason was customize of deer game to this liver fluke, since invasion with other parasites which reacts to triclabendazole weren't significantly reduced (Slavica at al, 2006). For instance, Erhardova-Kotrla (1971) stated that in Danube hardwoods eco-system, overgrown with cane and other marsh vegetation, there was adequate climate and hydrological conditions for development of intermediate hosts to maintain *F. magna* life cycle. In Europe, the most common intermediate host is *Lymnaea truncatula* snail, which is present in Baranja region (Kalinović, 1999).

During our survey, *F. magna* EPG ranged from 1-300 in faecal samples collected in hunting ground XIV/9 "Podunavlje-Podravlje". The highest number was found in the first year (2001) to decrease until 2004. Figure 1 shows the frequency distribution of EPG from 2001 to 2004. Most of faecal samples (86.42%), were in the category up to 50 EPG while only 0.18%, in the category between 250 and 300 EPG. Overall, the arithmetical EPG mean of examined samples was 33 EPG. According to Balbo et al. (1989), values of 100 EPG indicated severe infections, while values around 50 EPG indicated less severe-mild infection. Based on our results and on the lack of parasite-induced mortality, it can be concluded that most of deer population in this area had mild infection. Similar results were found by Majoros and Sztojkov (1994) in their survey on fascioloidosis in Hungary, in Dunaremete and Lipót area, where most of faecal samples accounted less than 50 *F. magna* EPG.

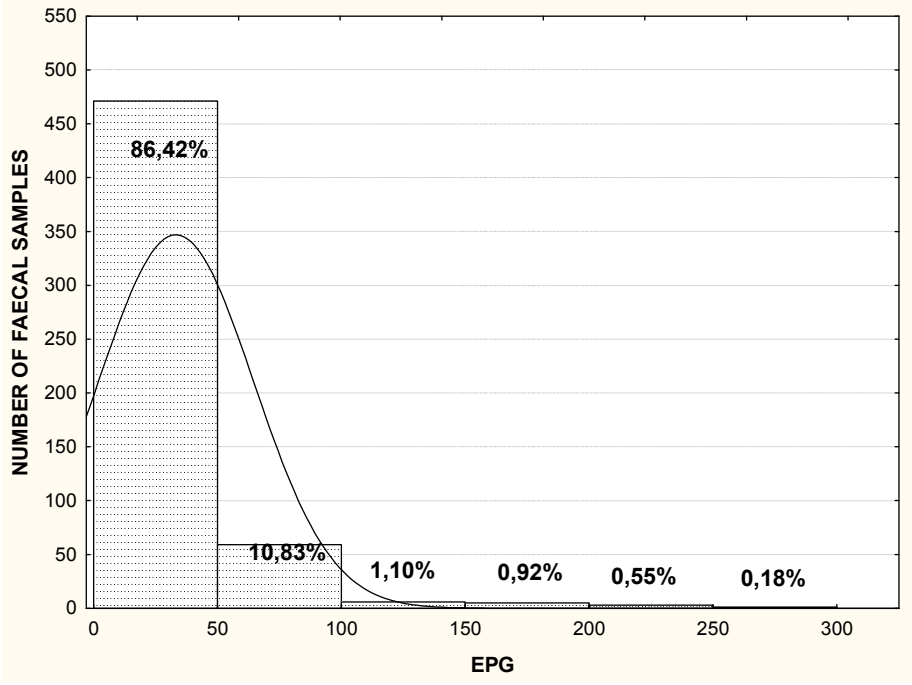

Figure 1. Frequency distribution of *F. magna* EPG in red deer faecal samples in hunting ground XIV/9 Podunavlje-Podravlje (2001-2004)

In 2 hunting grounds in middle Baranja, XIV/3 "Haljevo" and XIV/5 "Koha –Kozarac", no *F. magna*-positive faecal samples were found throughout the survey, but in 2004, when in 2 samples (6.67%) from "Koha –Kozarac" hunting ground was found positive (mean intensity 2 EPG). It should be as a consequence of the infections spreading during the survey period. However, deer probably migrate from Danube valley hunting grounds. This area has no direct hydrological connection with Danube valley, it was not flooded during the year and is located at 10 meter higher altitude. Therefore, the area of Drava River Basin Region and middle parts of Baranja could be characterized as fascioloidosis-endangered areas. Spreading of the infection downwards the river Drava cannot be excluded because of centuries migration paths of deer game which lead from flooded hunting grounds across the bogland of Drava in Baranja to their right riverside, in the area of Slavonia. Our results approve this, because the findings of liver fluke *F. magna* eggs in deer game faeces samples were collected in

hunting grounds XIV/10 ”Munjoroš” and XIV/10A ”Podravlje”, which have been elapsed along the Drava river on the south side of Baranja. During the years 2001 and 2002, using faecal examinations, *F. magna* eggs were not found in those hunting grounds. In year 2003, eggs were found in 6.67% samples and in year 2004 in 8% samples, with quite low intensity (3-4 EPG; Table 1). Our results approve the hypothesis of Majoros and Sztojkov (1994), based on their results in Hungary. They concluded that slowly spreading of fascioloidosis in Danube valley could be expected, which had been approved with the first findings of liver fluke *F. magna* in Danube valley hunting grounds in Croatia. Based on our study, the spreading of the infection throughout the left riverside of Danube River, i.e. in Republic of Serbia, as well as in hunting grounds on the right riverside of Drava River in the Republic of Croatia can be predicted. In such areas, where the environmental conditions are not particularly favourable for *F. magna*, subclinical infections should spread in roe deer (*Capreolus capreolus*) and wild boar (*Sus scrofa*) as well as fallow deer *(Dama dama)* and moufflon *(Ovis musimon)* reared in enclosed hunting grounds. Beside game, in forests and pastures of eastern Slavonia cattle are traditionally reared in opened fields. Furthermore, it has to be stressed that the infection, even mild, to be lethal for pigs, goats and sheep (Stromberg at al., 1985). Although infections are not usually patent in such hosts, necropsy findings would be very important as epizootiological indicator. In eastern Slavonia, and especially in Danube valley and in Drava River Basin Region hunting grounds, health status of game is very important because of trophy value. It is an important economical resource, especially for red deer. Although *F. magna* infection apparently does not increase the deer mortality rate in Croatia, it could lead to a reduced natural resistance and shape of animals, which would result in diminished trophy value.

CONCLUSION

Fascioloidosis in red deer, with relatively high prevalence and infection intensity are limited to the flooded area of hunting grounds in eastern Baranja, along Danube River. Mild, less severe infections were found in hunting grounds of Baranja, along Drava River, and in its flooded middle part. In this area fascioloidosis represents a potential danger for other game, as well as for cattle. Prevention and systematic therapy are necessary measures in all the endangered areas, when the primary aim is control of the disease, especially in the hunting grounds with deer game. In order to hold the disease under constant control, a monitoring program should be carried out systematically. Based on previous epizootiological indicators, spreading of disease on left side of the River Danube, i.e. in the Republic of Serbia as well as in hunting grounds on the right side of the River Drava in the Republic of Croatia could be expected.

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