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PARASITOFAUNA IN THE GASTROINTESTINAL TRACT OF THE CERVIDS (CERVIDAE) IN NORTHERN POLAND

BURLINSKI P*, JANISZEWSKI P*, KROLL ANNA** and GONKOWSKI S**

*University of Warmia and Mazury in Olsztyn, Faculty of Animal Bioengineering, Poland **University of Warmia and Mazury in Olsztyn, Faculty of Veterinary Medicine. Poland

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The aim of this study was to determine parasite species and their prevalence in the gastrointestinal tract of the roe deer (Capreolus capreolus L.), red deer (Cervus elaphus) and fallow deer (Dama dama) from selected areas of northern Poland. A total of 1668 fecal samples, obtained from animals living in three various regions, were analyzed. The samples were examined by coproscopic techniques, including flotation (according to Fülleborn), decantation (according to Zarnowski and Josztowa) and flotation in the McMaster chamber. The experimental materials were collected from January 2008 to January 2009, in the hunting districts belonging to two hunting associations, "Bór" in Wipsowo and "Gwardia Slupsk" in Lupawa. In order to compare the health status of cervids kept under farm-like conditions, fecal samples were also collected in the paddocks of the Municipal Zoological Garden "Wybrzeza" in Gdansk. Parasites (eggs and oocysts) were found in 42.09% of the analyzed samples. The most frequently observed roundworm was Chabertia sp. (13.79% in roe deer, 12.07% in red deer, 11.36% in fallow deer). High counts of Ostertagia sp. eggs (11.55% in roe deer, 6.03% in red deer, 13.18% in fallow deer) and Trichostrongylus sp. eggs (10.69% in roe deer, 2.93% in red deer, 10.68% in fallow deer) were also noted. The highest infestation rates (51.17%) were determined in the studied areas of the Zoological Garden in Gdansk.

Key words: coccidiosis, fallow deer, gastrointestinal nematodes, infestation rates, red deer, roe deer

INTRODUCTION

According to most authors, parasites significantly affect the size and the quality of deer (*Cervidae*) populations (Pilarczyk *et al.*, 2005). Cervids are gregarious animals, and owing to the widespread use of antiparasitic drugs in farm-raised ruminants, the animals growing resistance to parasites increases the concentrations of various invasive forms in the environment. The above increases the prevalence (infestation rate) and the intensity (parasite counts per individual)

of parasite invasions (Wedrychowicz, 1996). This, in turn, could lead to deteriorating antler quality, adverse changes in the animals appearance and higher mortality rates (Pielowski, 1988). The absence of effective schemes for monitoring parasitosis in wild cervids occupying border areas, as well as species translocation zones could have adverse consequences, such as the appearance of the blood sucking nematode *Ashworthius sidemi* in Poland. This parasite was first detected in 1997, and by 2000, published sources pointed to its 100% extensiveness in southern border regions (Drozdz *et al.*, 2000).

Since roe deer occupy a small home range area (Mysterud, 1998; Morellet *et al.*, 2009) and they move within a limited radius, this species was regarded as a good indicator of parasite contamination in a given region. Red deer and fallow deer travel long distances, therefore, they may be a vector that plays an important role in parasite transmission in the natural environment (Geist, 1998). The above arguments support an analysis of endoparasitic invasions in the discussed species.

The objective of this study was to determine the prevalence of parasitic infestations in popular game species such as the roe deer (*Capreolus capreolus*), fallow deer (*Dama dama*) and red deer (*Cervus elaphus*). The prevalence of parasite populations in the gastrointestinal tract of the studied animals was determined subject to the season. This study also aimed to determine infestation rates in the investigated areas.

MATERIAL AND METHODS

The study covered three local sub-populations of roe deer and red deer, including two wild populations and one farm population, and two populations of fallow deer, including one wild population and one farm population. The experimental material comprised fecal samples obtained from the analyzed areas. The samples were collected in three regions marked by similar environmental and geoclimatic characteristics (Fig. 1):



Region A - 54°39'N, 17°44'E - hunting district of the "Gwardia Slupsk" hunting association in Lupawa,

Region B - 53°86'N, 20°84'E - hunting district of the "Bor" hunting association in Wipsowo,

Region C – 54°41'N, 18°52'E – "Wybrzeza" Municipal Zoological Garden in Gdansk--Oliwa.

Figure 1. Location of research areas

Samples were collected between January 2008 and January 2009. Around 20 fresh feces samples were collected from each analyzed region at 30-day intervals. A total of 580 roe deer feces samples, 440 fallow deer feces samples and 648 red deer feces samples were examined (n=1668). Since the rearing period of cervids falls in the summer months, in particular in July and August, samples were not collected during that time.

The collected samples were analyzed by standard coproscopic techniques applied in studies of parasite infestations, including flotation (according to Fülleborn), decantation (sedimentation)(according to Zarnowski and Josztowa) (Ziomko and Cencek, 1999; Zajac *et al.*, 2006) and flotation in the McMaster chamber (Zajac *et al.*, 2006). In order to assess species of gastrointestinal nematodes, infective (L3) larvae were cultured. The results were used to estimate the prevalence and the intensity of endoparasitic infestation. To compare the data on infestation intensity, the results were processed in terms of standard EPG (eggs per gram of feces) and OPG (oocysts per gram of faeces) units.

Statistical data obtained from each of the investigated regions was subjected to a comparative analysis to determine the quantitative variability of endoparasites in each month of the study. The significance of differences was estimated with the use of Duncan's test. Data were processed statistically in the STATISTICA 8.0 application (StatSoft Inc., Tulsa, Oklahoma, USA).

During the study, the cervids at the Municipal Zoological Garden in Gdansk underwent preventive deworming with a phenebedasol-containing drug (FENBENAT, 4% premix, Vetos-Farma, Poland). Red deer and fallow deer were dewormed in March and September, and roe deer – only in April. Regardless of the cervid species, the drug was administered for 6 consecutive days at a dose of 5 g/40 kg b.w.

RESULTS

The results of the study revealed four parasite groups: nematodes colonizing the gastrointestinal tract, coccidia of the genus *Eimeria*, digenea and tapeworms. The overall infestation rate in the analyzed cervids was 42.09%. The highest parasite prevalence was noted in region C (roe deer – 71.25%, red deer – 32.73%, fallow deer – 55%), while the lowest infestation rates were observed in region A (roe deer – 33.49%, red deer – 22.64%) (Table 1).

The differences in the contamination level of feces samples obtained from roe deer from different regions were highly statistically significant. The differences noted between samples from region A and region B proved to be statistically significant. No statistical differences were noted in the contamination level of red deer and fallow deer samples from various regions. The analysis revealed statistically significant (p=0.05) differences between feces samples of roe deer and red deer collected in region C (Table 2). No statistical differences were observed in the contamination level of feces samples from region B.

An analysis of the parasitic composition of feces samples from different cervid species (Table 3) revealed that gastrointestinal nematodes (34.66%) and coccidia (13.10%) were the most prevalent parasites in roe deer, while other

		Prevalence (%)		48.18	55.00	51.59
Fallow deer	allow deer	positive samples	I	106	121	227
	Numt	tested samples	I	220	220	440
		Prevalence (%)	22.64	27.78	32.73	27.78
Red deer	Red deer Number of	positive samples	48	60	72	180
		tested samples	212	216	220	648
		Prevalence (%)	33.49	48.51	71.25	50.86
Roe deer	Roe deer Number of	positive samples	73	98	114	295
		tested samples	218	202	160	580
	Area of sample collection			В	O	Total

Table 1. Extensiveness of endoparasitic invasions in selected cervids species in the investigated areas

parasite groups were rarely found. 58.12% of the samples collected from roe deer inhabiting a small home range area (region C) were infested with helminth eggs, while coccidia had a high share of the contaminated samples collected from wild roe deer (16.51% in region A and 17.33% in region B). Similar results were noted for the parasitic composition of red deer samples. The highest infestation rate was reported in respect of gastrointestinal nematodes (22.22%) which were determined in 28.64% samples from region C, in 18.98% samples from region B and in 18.87% samples from region A. The highest coccidia counts were observed in the samples from region B (10.19%). Gastrointestinal nematodes were also most prevalent in fallow deer samples, and the highest infestation rates were noted in region B (41.36%). Similarly to other cervid species inhabiting this area, a high rate of coccidia infestation (13.64%) was observed in the fallow deer. As regards protozoa, only oocysts of the genus *Eimeria* were found. Flotation analyses revealed the presence of tapeworm eggs of the genus Moniezia. In the group of digenea, the results of flotation and sedimentation tests confirmed the occurrence of Paramphistomum sp. and the common liver fluke.

Table 2. Significance of differences in total contamination of samples in the investigated areas

Statistics	С	В	А	Significance of differences					
	Roe deer (1)								
X S X _{min} X _{max}	59.09 30.48 0 100	48.70 8.80 40 66.66	33.40 13.88 15 55	1C1B, 1A** 1B1A* 1C2C*					
Red deer (2)									
X S X _{min} X _{max}	32.73 25.53 0 70	35.91 22 0 75	22.73 9.04 0 35	No differences					
Fallow deer (3)									
X S Xmin Xmax	55.00 29.15 10 100	48.18 9.82 35 65	- - - -	No differences					

* - level of significance at p=0.05; ** - level of significance at p=0.01

The greatest variation was observed in the group of gastrointestinal nematodes (Tables 4, 5 and 6). Samples from region A were mostly infected with *Chabertia sp.* (roe deer -7.34%, red deer -9.43%) and *Ostertagia sp.* eggs (roe deer -5.96%, red deer -5.19%). In samples from region B, the highest infestation

rates were reported for *Haemonchus sp.* (roe deer – 14.85%, red deer – 5.09%, fallow deer – 14.55%), as well as *Ostertagia sp.* and *Chabertia sp.* eggs. The most prevalent parasite found in samples from region C was *Chabertia sp.* (roe deer – 30%, red deer – 16.82%, fallow deer – 12.73%). A high degree of contamination with the eggs of *Ostertagia sp.* (roe deer – 25.63%, red deer – 3.18%, fallow deer – 19.09%) and *Trichostronyglus sp.* (roe deer – 28.13%, red deer – 4.09%, fallow deer – 10.68%) was also noted.

Table 3. Extensiveness of parasitic invasions in the analyzed cervids species in the investigated areas

Research area	A		В		С		Total		
Parasite group	Number of positive samples	%	Number of positive samples	%	Number of positive samples	%	Number of positive samples	%	
	Roe deer								
Coccidia	36	16.51	35	17.33	5	3.13	76	13.10	
Digenea	1	0.46	1	0.49	0	0	2	0.34	
Gastrointestinal nematodes	39	17.89	69	34.16	93	58.12	201	34.66	
Tapeworms	2	0.92	1	0.49	1	0.625	4	0.69	
Total number of samples	218		202		160		580		
		_	Red	deer		_			
Protozoa	11	5.19	22	10.19	10	4.55	43	6.63	
Gastrointestinal nematodes	40	18.87	41	18.98	63	28.64	144	22.22	
Tapeworms	0	0	3	1.39	2	0.91	5	0.77	
Total number of samples	212		216		220		648		
	Fallow deer								
Protozoa	-	-	30	13.64	2	0.91	32	7.27	
Gastrointestinal nematodes	_	_	91	41.36	80	36.36	171	38.86	
Tapeworms	_	_	3	1.36	2	0.91	5	1.14	
Total number of samples	_	_	220		220		440		

Table 4. Extensiveness of fec	es sample contaminatic	on with different parasite	e families
in the roe deer in selected re	search areas		

Research area	А		В		С		Total	
Parasite	Number of positive samples	%	Number of positive samples	%	Number of positive samples	%	Number of positive samples	%
Eimeria sp.	36	16.51	35	17.33	5	3.13	76	13.10
Fasciola hepatica	0	0	1	0.50	0	0	1	0.17
Paramphistomum sp.	1	0.46	0	0	0	0	1	0.17
Moniezia sp.	2	0.92	1	0.50	1	0.63	4	0.69
Bunostomum sp.	2	0.92	1	0.50	0	0	3	0.52
Capillaria sp.	0	0	1	0.50	0	0	1	0.17
Chabertia sp.	16	7.34	16	7.92	48	30.00	80	13.79
Haemonchus sp.	10	4.59	30	14.85	5	3.13	45	7.76
Nematodirus sp.	0	0	3	1.49	0	0	3	0.52
Ostertagia sp.	13	5.96	13	6.44	41	25.63	67	11.55
Strongyloides sp.	0	0	0	0	15	9.38	15	2.59
Trichostrongylus sp.	7	3.21	10	4.95	45	28.13	62	10.69
Total number of samples, N=	218		202		160		580	

The results of the study point to a varied degree of eggs and oocysts excretion in feces. Owing to a large number of parasite families in each of the examined areas, the intensity of contamination was determined solely in respect of parasites whose presence was noted on three or more occasions during the experimental period. As proposed by Zajac *et al.* (2006), parasite counts of 500-1000 EPG/OPG were regarded as indicative of moderate to acute parasite invasion. As regards roe deer in region C, the intensity of *Chabertia sp.* infestation exceeded 100 EPG in the course of three months (February, March and May). A high number of *Trichostrongylus sp.* and *Ostertagia sp.* eggs was also noted, and high parasite counts were reported in both cases throughout the period of the study, reaching the maximum registered level in March. The intensity of infestation by the above parasites reached 2480 EPG and 1420 EPG, respectively. The number of the remaining nematode eggs did not exceed 100 EPG in the studied samples, and they were observed sporadically in comparison with the remaining

three parasites. *Chabertia sp.* egg counts in red deer feces samples exceeded 200 EPG in January, February and October. Maximum counts of this helminth were noted in February at 22300 EPG, which was the highest value observed in the experiment. A very high level of contamination with *Trichostrongylus sp.* (4820 EPG) and *Ostertagia sp.* (1860 EPG) eggs as well as *Eimeria sp.* (1040 OPG) oocysts was reported in the same month. In September, November and December, the intensity of infestation with parasite eggs exceeded 100 EPG (*Haemonchus sp.* – 140 EPG, *Moniezia sp.* – 100 EPG and *Bunostomum sp.* – 160 EPG). In the remaining months, the level of sample contamination remained low under 80 EPG. Parasite counts in the feces samples of fallow deer exceeded 100 EPG only on three occasions during the investigated period. In January, contamination levels reached 100 EPG (*Trichuris sp.*).

Table 5. Extensiveness of feces sample contamination with different parasite families in the red deer in selected research areas

Research area	A		В		С		Total	
Parasite	Number of positive samples	%	Number of positive samples	%	Number of positive samples	%	Number of positive samples	%
Eimeria sp.	8	3.77	22	10.19	10	4.55	40	6.90
Moniezia sp.	0	0.00	3	1.39	2	0.91	5	0.86
Trichuris sp.	1	0.47	7	3.24	3	1.36	11	1.90
Bunostomum sp.	1	0.47	2	0.93	8	3.64	11	1.90
Chabertia sp.	20	9.43	13	6.02	37	16.82	70	12.07
Haemonchus sp.	5	2.36	11	5.09	13	5.91	29	5.00
Oesophagostomum sp.	1	0.47	3	1.39	3	1.36	7	1.21
Ostertagia sp.	11	5.19	17	7.87	7	3.18	35	6.03
Strongyloides sp.	0	0.00	0	0.00	7	3.18	7	1.21
Trichostrongylus sp.	5	2.36	3	1.39	9	4.09	17	2.93
Total number of samples, N=	212		216		220		648	

In region B, *Eimeria sp.* oocysts counts in the feces samples of roe deer exceeded 200 OPG in six different months, and oocysts counts were higher than 2000 OPG on three occasions (February, March, November). The maximum level of contamination was 3820 OPG. In addition to the above coccidia, the presence

of Haemonchus sp. was noted regularly, and the highest egg counts were reported in March (260 EPG). Chabertia sp. eggs were shed between January and June, but their presence was not noted in the fall and winter. The reverse was observed in respect of Trichostrongylus sp. whose presence was clearly demonstrated in the fall and winter, while no eggs of this parasite were found in the spring. The contamination intensity of red deer feces samples from region B was moderate throughout the experimental period. In January, April, September and October, Eimeria sp. oocyte counts exceeded 200 OPG, and they reached 1440 OPG and 3420 OPG in two samples in January. In February, March and January 2009, the intensity of contamination with Ostertagia sp. eggs reached 120 EPG. A high level of contamination with Moniezia sp. eggs was noted in November and December at 640 EPG and 260 EPG, respectively. The number of parasite eggs and oocysts in feces samples collected from fallow deer inhabiting region B was high throughout the experimental period. *Eimeria sp.* oocysts counts exceeded 200 OPG in six different months, reaching 1060 OPG in May, 1020 OPG in September and 1100 OPG in October. The level of contamination with gastrointestinal nematodes was moderate between March and October, and it did not exceed 260 EPG. In the winter and spring, parasite counts higher than 100 EPG were not noted.

Research area	В		c c		Total		
Parasite	Number of positive samples	%	Number of positive samples	%	Number of positive samples	%	
Eimeria sp.	30	13.64	2	0.91	32	7.27	
Fasciola hepatica	3	1.36	0	0.00	3	0.68	
Paramphistomum sp.	0	0.00	2	0.91	2	0.45	
Trichuris sp.	10	4.55	6	2.73	16	3.64	
Bunostomum sp.	4	1.82	0	0.00	4	0.91	
Chabertia sp.	22	10.00	28	12.73	50	11.36	
Haemonchus sp.	32	14.55	14	6.36	46	10.45	
Nematodirus sp.	8	3.64	0	0.00	8	1.82	
Oesophagostomum sp.	2	0.91	0	0.00	2	0.45	
Ostertagia sp.	16	7.27	42	19.09	58	13.18	
Strongyloides sp.	4	1.82	4	1.82	8	1.82	
Trichostrongylus sp.	29	13.18	18	8.18	47	10.68	
Total number of samples, N=	220		220		440		

Table 6. Extensiveness of feces sample contamination with different parasite families in the fallow deer in selected research areas

Roe deer				
Males	11.05 – 30.09			
Females and young	01.10 – 15.01			
Red deer				
Males	21.08 – the end of February			
Females	01.10 – 15.01			
Young	01.10 – the end of February			
Fallow deer				
Males	01.10 – 31.01			
Females and young	01.10 – 15.01			

Table 7. Dates of hunting on selected cervid species in Poland (In based on Dz.U. nr 48, poz. 459, 2005r)

In region A, *Eimeria sp.* oocyst counts exceeded 200 OPG in the feces samples of roe deer in the course of three months. The highest level of contamination by this parasite was observed in December at 8120 OPG. *Chabertia sp.* and *Trichostrongylus sp.* eggs were relatively frequently identified in feces samples, yet the noted quantities were below the counts reported in the samples from region B. A similar number of observations showing egg counts in exceeded. In November, the intensity of contamination with *Moniezia sp.* eggs was very high at 1100 EPG. The fecal samples collected from red deer in region A were marked by low parasite counts throughout the period of the experiment. Contamination levels exceeded 100 EPG only in April (*Eimeria sp.* – 100 OPG and *Chabertia sp.* – 100 EPG) and in May (*Haemonchus sp.* – 280 EPG).

Regardless of the point of sample collection and the prevalence of local parasites, the highest contamination intensity was observed at the turn of February and March and at the turn of November and December. Parasite counts exceeded 200 EPG in 65 (3.89%) samples and 1000 EPG in 23 (1.38%) samples.

The highest level of contamination in Zoological Garden in Gdañsk may have the connexion with the transmission of the oocysts, eggs, and larves with the food. Small paddocks may be also one of causes unnatural big concentration of invasive forms of parasites.

DISCUSSION

The results of this study are similar to the findings of other authors (Zalewska-Schönthaler and Szpakiewicz, 1987; Romaniuk, 1999; Cisek *et al.*, 2003; Cisek *et al.*, 2004). Most studies investigate feces samples collected directly from the rectum of hunted cervids. Investigations of this type are marked by a relatively high possibility of error because the collection of samples is limited

to the hunting season (Table 7). The results of this study indicate that the hunting season coincides with the infestation peak in the fall and late winter. The hunting season for the studied cervid species falls in the period marked by the highest rate of parasite egg and oocyst shedding.

Gastrointestinal nematodes were most frequently observed in the studied feces samples. In selected Polish regions, including Wielkopolska (Drózdz et al., 1987; Drózdz and Dudzinski 1993), eastern Poland (Drózdz et al., 1989) and north-eastern Poland (Drózdz et al., 1992), the rate of infestation with gastrointestinal nematodes was determined at 100%. Zalewska- Schönthaler and Szpakiewicz (1987) noted the presence of the above parasites in 50% of individuals studied in north-eastern Poland. According to Romaniuk (1999), the extensiveness of contamination with gastrointestinal nematodes was only 30% in free-ranging red deer and 40% in farm-raised populations. As noted by American authors, parasite invasions are one of the main causes of mortality in deer farms. According to Hattel et al. (2007), 32.31% of deaths in Pennsylvania farms resulted from infestations with gastrointestinal nematodes. Cisek et al. (2004) reported infestation rates of 84% in roe deer, 71.43% in red deer and 47.50% in fallow deer in Western Pomerania. In a previous study, the above authors observed parasite prevalence rates of 96.51% in roe deer, 73.53% in red deer and 92% in fallow deer (Cisek et al., 2003). Czech investigators noted growing infestation rates in roe deer, reaching 78.4% (Tománek, 1967), 88% (Dyk and Chroust, 1974) and 100% (Vetyška, 1980). Belarusian researchers also reported very high rates of infestation in cervids at 92% (Shimalov and Shimalov, 2002).

Varied levels of parasite invasions in cervids were reported in a German study (Rehbein et al., 2000). The highest gastrointestinal nematode egg counts (excluding Nematodirus sp.) in roe deer reached 9520 EPG. The maximum level of feces sample contamination with Nematodirus sp. eggs was 120 EPG, with Trichuris sp. eggs – 30 EPG, and with coccidia oocysts – only 20 OPG. As regards gastrointestinal nematodes (excluding Nematodirus sp.), the intensity of invasions was several times higher than reported in this study. The coccidia counts noted in our experiment significantly exceed the values reported by German researchers. In a study of red deer, Rehbein et al. (2002) noted that the level of contamination with endoparasitic eggs and oocysts did not exceed 140 EPG for gastrointestinal nematodes (excluding Nematodirus sp. eggs), 175 EPG for Nematodirus sp. and 15 EPG for Capillaria sp. The intensity of infestation with Nematodirus sp. eggs was higher than that reported in this study, while the rate of egg shedding by the remaining parasites was several times lower. As regards fallow deer, the above authors noted the following levels of contamination with parasite eggs: gastrointestinal nematodes (excluding Nematodirus sp.) - 50 EPG, Nematodirus sp. - 60 EPG and Capillaria sp. - a 10 EPG. The above values are much lower than those reported in this study.

The results of this study indicate that endoparasites colonizing the digestive tract pose a significant threat to both free-ranging and farm-raised cervids in northern Poland and other parts of the country. The identified parasites are characterized by taxonomic variation, and their distribution is determined by geoclimatic conditions and human activity. Effective parasitofauna monitoring schemes, in particular in border areas and species translocation zones, will limit parasite occurrence and slow down their migration to other regions. The results of this study could contribute to the development of a deworming scheme addressing wild cervids to minimize parasite incidence, reduce egg shedding rates, and lower the intensity and prevalence of parasite invasions in cervids to the levels reported in other countries, such as Germany.

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Address for correspondence: Burlinski Piotr Jozef Katedra Fizjologii Klinicznej WMW Oczapowskiego 13/024 10-759 Olsztyn - Kortowo, Poland E-mail: p.j.burlinski@uwm.edu.pl

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PARAZITOFAUNA GASTROINTESTINALNOG TRAKTA CERVIDA U SEVERNOJ POLJSKOJ

BURLINSKI PJ, JANISZEWSKI P, KROLL ANNA i GONKOWSKI S

SADRŽAJ

Cilj ovog istraživanja je bio da se utvrde parazitske vrste i njihova prevalenca u gastrointestinalnom traktu srne (*Capreolus capreolus L.*), evropskog jelena (*Cervus elaphus*) i jelena lopatara (*Dama dama*) iz odabranih oblasti severne Poljske. Analizirano je ukupno 1668 uzoraka fecesa od životinja iz tri različite regije. Uzorci su analizirani koproskopskim tehnikama, uključujući flotaciju (prema Filebornu), dekantaciju (prema Žarnovskom i Joštovoj) i flotaciju u Mekmasterovoj komori.

Eksperimantalni materijal je sakupljan od januara 2008. do januara 2009. godine u lovištima koji pripadaju lovačkim udruženjima, "Bur" u Vispovu i "Gvardia Slupsk" u Lupavi. Radi poređenja zdravlja cervida koji su držani u farmskim uslovima, fekalni uzorci su sakupljani na pašnjacima gradskog zoološkog vrta "Vibžeža" u Gdanjsku. Paraziti (jaja i oociste) su pronađeni u 42,09% analiziranih uzoraka. Najčešće je otkrivan valjkasti crv, *Chabertia sp.* (kod srne 13,79%, jelena 12,07% i jelena lopatara 11,36%). Takođe je otkriven veliki broj jaja *Ostertagia sp.* (kod srne 11,55%, jelena 6,03% i jelena lopatara 13,18%) i jaja *Trichostrongylus sp.* (kod srne 10,69%, jelena 2,93% i jelena lopatara 10,68%) je. Najveći stepen infestacije (51,17%) je utvrđen u zoološkom vrtu u Gdanjsku.