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Helminthofauna of European bison from Borisov-Berezinsky free-living population

Alexander Kashtalian[1], Alexander Subbotin[2], Tatiana Shendrik[3]

[1] OOO «Interservis», Vitebsk Region, Belarus
[2] Vitebsk State Academy of Veterinary Medicine, Belarus
[3] The Scientific and Practical Center for Bioresources the National Academy of Science of Belarus, Minsk, Belarus

Abstract: Data on specific and quantitative structure of helminthofauna of European bison (*Bison bonasus bonasus*) from Borisov-Berezinsky free-living population are presented. Degree of seasonal infection of E. bison by different helminth species is estimated. Ways of infecting an organism by most frequent parasites – *Fasciola hepatica* and *Dictyocaulus filaria* are described

Key words: European bison, Belarus, Berezinsky Biosphere Reserve, helminthofauna, population

Introduction

In 1974 according to the decision of Central Administrative Board for Nature of the Ministry of Agriculture of the USSR regarding the maintenance of Soviet protected areas, five E. bison (*Bison bonasus bonasus*) were brought to Berezinsky reserve. Those animals were born in Prioksko-Terrasny reserve. The purpose of their introduction was the creation of free-living population of European bison on the territory of Berezinsky reserve.

On arrival to the Reserve bison were placed at an enclosure, but soon animals have broken the fence and started to penetrate neighbouring territories. As the home range designated for the herd appeared to be not particularly suitable, with numerous flooded woods, extensive marshes, after a while animals left the Reserve, and moved to the agricultural grounds located at its southern borders.

By 1980, in vicinities of the reserve the free-living population of European bison counting 15 individuals was created. Gradually bison have settled neighbouring territories. The herd had developed system of seasonal migrations triggered by changes in forage supply and a disturbance from people. During the summer animals occupied the reserve territory but they wintered in forests along flood plain of the river Tsna, 25 kilometres from the reserve borders. This district due to its conditions is the most suitable for winter dwelling of the herd. Small woodlands alternate here with agricultural fields, there is a large number of overgrown cutover patches. Moraine hills overgrown with

woody vegetation adjoin directly Tsna's flood plain. Despite the existence of a large number of villages, during winter time after the end of hunting season this territory is scarcely visited.

The strategy of seasonal migrations developed in the early eighties by E. bison from Borisov-Berezinsky population, which number totals 34 individuals, is maintained until now. Such territorial behaviour in many respects defines specific and quantitative composition of endoparasites of European bison described in this publication.

Study area

E. bison from Borisov-Berezinsky population inhabit the territory of two districts (Borisov and Logoisk) which belong to Minsk region. Total area of bison natural habitat make up 7.5 thousand hectares. There are two sites most intensively used by the herd. The first, located in southern side of the reserve in vicinities of lake Palik in woods and on agricultural lands at village Barovljany in a buffer zone near reserve borders, animals visit during a warm season – from April until October. For winter the herd moves to the territory of Zembin and Ikany forestries of the Borisov forest enterprise, situated nearby from the river Tsna. Not far from this territory settlements Gantsevichi, Pogranich'e, Zamosh'e, Zembin are situated, there are agricultural fields, pastures for cattle and hay meadows. The total area of mentioned fields does not exceed 3 thousand hectares. Other part of the home range is visited by E. bison mainly during seasonal migrations between winter and summer ranges of the herd. This territory is mosaic and represents a mosaic of settlements, agricultural grounds and small forest tracts.

Materials and Methods

In 2000 we made helminthological estimation of Borisov-Berezinsky population. Specific and quantitative composition of helminthes was defined during full and particulate helminthological dissections, as well as during carrying out coproscopic analyses. Samples (N = 76) for the latter were collected three times a year – in February-March in a herd wintering area, in June-July in summer localities and in October right after returning of animals to their wintering sites. Besides, in autumn, on summer and winter localities we collected molluscs (first of all – *Lumnaea truncatula*) as intermediate hosts of a number of helminthes for helminthological assessment of pastures. The Fulleborn method (Urquhart *et al.* 1996) allowing to trap out eggs of the majority of nematode and cestode species has been chosen. It is based on floatation properties of eggs of helminthes in a saturated solution of salt at the expense of smaller relative density. For a quantitative assessment a batch of 20 g of excrements was taken and determination of all eggs and larvae found

in it was conducted. Analyses of excrements both before dehelminthezation of E. bison in sites of winter foraging and after the treatment, were made in triple replication.

The following organs and tissues were sampled from dead animals: liver, cordis, kidney, spleen, lungs, diaphragm and muscles. Analysis of the samples was carried out according to Skryabin complete helminths animal thanatopsy (Shevzov 1970).

The following sources were used for comparative analysis: literature data for species composition of E. bison helminth fauna in the Belorussian part of Białowieska Forest and in Prioksko-Terrasny reserve, as well as parasite fauna data for animals of Borisov-Berezinsky population, obtained in 1990s.

Results and Discussion

When introducing the animals to a new area, it is important to take the necessary quarantine measures and to perform dehelminthization in order to respond to parasitic diseases. However, as archived materials suggest, no such measures have been taken when transporting bison to Berezinsky Reserve. The correspondence about the reference "On state of free-living bison, transported to Berezinsky Reserve", presented in 1976 to the Department of Environment Protection, Reserves, Forest and Game Sector of the USSR Agriculture Ministry by Reserve director Ju.N. Chichikin, states that "...none of E. bison, transported to Berezinsky Reserve, have been analyzed for parasites. None of them has been subjected to preventive dehelminthization either…". This is also proved by veterinary certificates, issued by Serpukhov Veterinary Station on February 6th and 13th, 1974. These state that only some of animals were under veterinary inspection (i.e. quarantined). However, none of these have been subjected to analyses, vaccination or any other treatment. Actually, even the first several years after bison introduction to the Reserve revealed that "...they are highly infected with bowels strongylatosis, rumen paramphistomatosis, liver fasciolosis..." (abstract from the correspondence between Ju.N. Chichikin and E. bison breeding nursery of Prioksko-Terrasny reserve M.A. Zablotsky).

Fasciola has been detected at Berezinsky reserve immediately after E. bison were released to the wild (Litvinov *et al*. 1978). All analyzed animals older than one year were infected with this trematode. E. bison fasciolasis is a chronic disease. Its clinical presentation is not obvious. However the *Fasciol*a impacst a bison organism significantly. Due to liver dysfunction, gastrointestinal tract performance is negatively impacted, fodder digestion and nutrient availability deteriorate. Animal fatness is also insufficient. For some of the Berezinsky reserve bison, weight loss and a delay of exuviations were registered. Analysis of the animal excrements revealed 100% invasion with trematode eggs. Infected animals were treated with Dertil B drug. The drug was admixed

to combined food, which was used as extra nutrition. Animal excrements were analyzed for *Fasciola* eggs again in two weeks. The analysis revealed that all animals were free from parasites. Later their fatness increased (Litvinov, Karasev 1981).

Paramphistmidae parasite with many ruminants. The rogues, collected from the bison, belong to *Liorchis scotiae* species. The trematode infects up to 50% of Borisov-Berezinsky population. Invasion intensity is often very high. Some of the animals show up to 22,000 of parasites per individual (Litvinov, Karasev, 1981).

The parasitological analysis of digestive tract of a bison carried out in 1994 by Karasev (Berezinsky Biosphere Reserve nature chronicles, 1995) revealed four more parasitic worm species:

- *Bunostomum phlebotomum* – a nematode from Ancylostomatidae family, which belongs to Strongilata and is localized in small intestine. Invasion intensity was 188 rogues;
- *Cooperia oncophora*, *Nematodirus helvetianus* and *Ostertagia ostertagi* – nematodes from Trichostrongylidae family. The first two ones are localized in small intestine. The last one is localized in true stomach and in duodenum. Invasion intensity for *Cooperia oncophora* and *Ostertagia ostertagi* was 460 rogues each, and for *Nematodirus helvetianus* – 22 rogues.

The helminthological analysis of Borisov-Berezinsky population, carried out during the winter of 2004, revealed 7 species of helminthes, trematodes: *Fasciola hepatica* and *Paramphistomum ichikawai*, nematodes: *Trichocephalus globulosa*, *Capillaria bilobata*, *Haemonchus contortus*, *Dictyocaulus filaria* and *Dictyocaulus viviparous*. Some species were very frequently registered with high invasion intensity (Tabl. 1). Eggs of *F. hepatica* and *H. contortus* were registered in excrement samples with frequency of 65.0% and 80.0%, respectively, and larvae of *D. filaria* were registered with frequency of 75.0%. The frequency of

Table 1. Results of analyses for the presence of larvae and eggs of helminthofauna in E. bison from Berezinsky Reserve

No	Species	The occurrence (%) of larvae and eggs	The number of larvae per 100 samples	The number of eggs per 100 samples	The number of larvae and eggs per sample
1.	*Fasciola hepatica*	65,0		3600	36,0
2.	*Paramphistomum sp.*	5,0		900	9,0
3.	*Trichocephalus sp.*	1,0		100	1,0
4.	*Capillaria sp.*	1,0		100	1,0
5.	*Strongylata sp.*	80,0		4200	42,0
6.	*Dictyocaulus filaria*	75,0	3420		34,2
7.	*Dictyocaulus viviparous*	4,5	700		7,0

Table 2. Helminthofauna species composition of E. bison populations from Prioksko-Terrasnyi Reserve, National Park «Belovezhskaya Pushcha» and Berezinsky Biosphere Reserve

No	Species	Prioksko-Terrasny reserve*	Belovezhskaya Pushcha**	Borisov-Berezinsky population
1	*Fasciola hepatica*	+ (hg)	+ (av)	+ (hg)
2	*Dicrocoelium lanceatum*	+ (hg)	+ (av)	+ (av)
3	*Dicrocoelium dendriticum*		+ (sp)	
4	*Liorchis scotiae*	+ (sp)		+ (hg)
5	*Moniezia benedeni*	+ (lw)		
6	*Moniezia expansa*		+ (sp)	
7	*Oesophagostomum radiatum*	+ (sp)	+ (sp)	+ (sp)***
8	*Bunostomum trigonocephalum*	+ (sp)	+ (sp)	+ (sp)
9	*Bunostomum phlebotomum*			+ (sp)
10	*Trichostrongylus axei*		+ (sp)	
11	*Ostertagia ostertagi*	+ (hg)	+ (av)	+ (av)
12	*Ostertagia bacuriani*		+ (hg)	
13	*Ostertagia circumcincta*		+ (sp)	
14	*Ostertagia antipini*		+ (sp)	
15	*Ostertagia lyrata*		+ (sp)	
16	*Ostertagia gruhneri*		+ (av)	
17	*Cooperia oncophora*	+ (hg)	+ (lw)	+ (av)
18	*Cooperia memasteri*	+ (sp)	+ (sp)	
19	*Cooperia punctate*		+ (sp)	
20	*Cooperia pectinata*		+ (sp)	
21	*Cooperia zurnabada*		+ (sp)	
22	*Haemonchus contortus*	+ (sp)	+ (av)	+ (hg)
23	*Haemonchus placei*	+ (hg)	+ (av)	
24	*Nematodirus helvetianus*	+ (lw)	+ (av)	+ (sp)
25	*Nematodirus abnormalis*		+ (sp)	
26	*Nematodirus oiratianus*		+ (sp)	
27	*Dictiacaulus viviparous*	+ (lw)	+ (av)	+ (lw)
28	*Nematodirus vitulorum*			+ (sp)
29	*Dictiacaulus filaria*			+ (hg)
30	*Thelazia gulosa*		+ (sp)	
31	*Staria labiata-papillosa*	+ (sp)	+ (sp)	

Table 2. continued

No	Species	Prioksko-Terrasny reserve*	Belovezhskaya Pushcha**	Borisov-Berezinsky population
32	*Setaria cervi*		+ (sp)	
33	*Trichocephalus ovis*	+ (lw)	+ (sp)	
34	*Trichocephalus gazellae*		+ (av)	
35	*Trichocephalus skrjabini*	+ (sp)		
36	*Trichocephalus globulosa*		+ (sp)	+ (sp)
37	*Capillaria bovis*	+ (av)	+ (lw)	
38	*Capillaria bilobata*		+ (av)	+ (sp)
39	*Nematodirella longissimespilata*		+ (av)	
40	*Paramphistomum ichikawai*			+ (lw)
41	*Paramphistomum cervi*		+ (av)	
42	*Oesophagostomum venulosum*		+ (lw)	
43	*Oesophagostomum radiatum*		+ (av)	
44	*Parafasciolopsis fasciolaemorpha*		+ (sp)	
45	*Ashworthius sidemi*		+ (sp)	

* according Beliaeva (1959), Nazarova (1965) and Moskvin *et al.* (2004);
** according Krasochko*et al.* (1996);
*** found only by animal kept in demonstration enclosure of Reserve.
In the parenthesis are given the data on the occurrence: sp – observed sporadically; lw – low; av – average; hg – high

other helminth species was in the range from 1.0% to 5.0%. On the average, one sample contained 5 eggs of *D. filaria*, *H. contortus* and 10 larvae of *D. filaria*. Eggs of *T. globulosa*, *C. bilobata*, *P. ichikawai* and larvae of *D. viviparus* were registered in some cases.

Helminthoovoscopic analysis of two animals' (10 years old female and 15 years old male) excrements revealed that these were in 100% infected with *D. filaria* larvae. On the average, one sample contained 12 rogues and 13 larva of these helminths.

Following these results, the E. bison were treated with antihelminthic drugs, which were added to dry feed: fenbendazole granulate 10% and albendazole suspension 10%.

After this treatment analysis carried out in May of the same year showed that parasitic worm frequency and invasion intensity decreased. Species composition of registered helminths also decreased. Only 4 species were registered: *F. hepatica*, *H. contortus*, *D. filaria*, *D. viviparus*. Nematode *D. filaria* was the most frequent one (100%). Frequency of *D. viviparus* was 10.0%, of *H. contortus* – 5.0%, and *F. hepatica* – 1.0%. On the average, a single sample contained 9 (*D. filaria*)

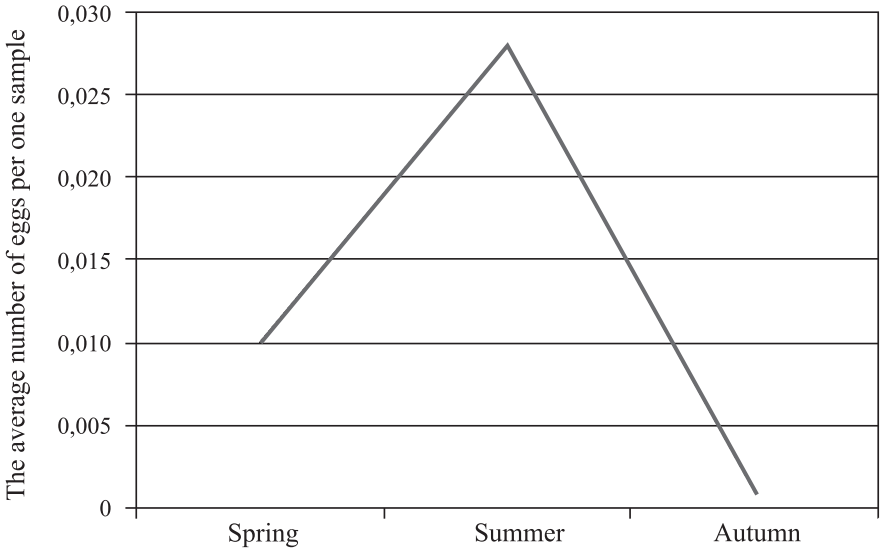

Figure 1. E. bison seasonal infestation with *Fasciola hepatica* in Berezinsky Reserve

and 2 (*D. viviparous*) lungworm larvae and 0.02 eggs of *F. hepatica* and *H. contortus*. Medicated treatment had a positive effect: both helminths frequency in samples and their relative numbers have decreased. Only *D. filaria* nematode frequency increased; however, its relative numbers decreased four times.

Samples taken in September of the same year showed only one helminth species – *D. filaria* (100%). On the average, one sample contained 6.5 larvae of this species. The presence of the nematode both before and after antihelminthic treatment might be conditioned by long usage of the same places for supplemental feeding, which are also visited by wild boars. The latter are one of the transmitting agents of the invasion. This could also lead to quick re-infection of E. bison with lungworms.

Excrement sample analysis was resumed in winter of 2005. Then, 6 types of helminths were found: *F. hepatica* in 30 samples (15%), *Dicrocoelium lanceatum* in 12 samples (6%), *Strongylata sp.* – in 98 samples (49%), *Nematodirus vitulorum* – in 2 samples (1%). A helminthovoscopic analysis of individual E. bison excrements showed that the animals were infected with nematodes *D. filaria* and *D. viviparus*. The invasion intensity was equal to 30% and 20%, respectively. The general infection intensity was up to 80%.

A female bison, which was drowned in the territory of the herd winter habitation, was subjected to partial helminthological autopsy. It revealed 9 specimens of *F. hepatica*, 12 – *Liorchis scotiae*, 14 – *Bunostomum trigonocephalum*, 46 – *Cooperia oncophora.*

When analyzing the excrements of male bison, which is kept in the reserve enclosure, 40 specimens of *Oesophagostomum radiatum* nematode were found

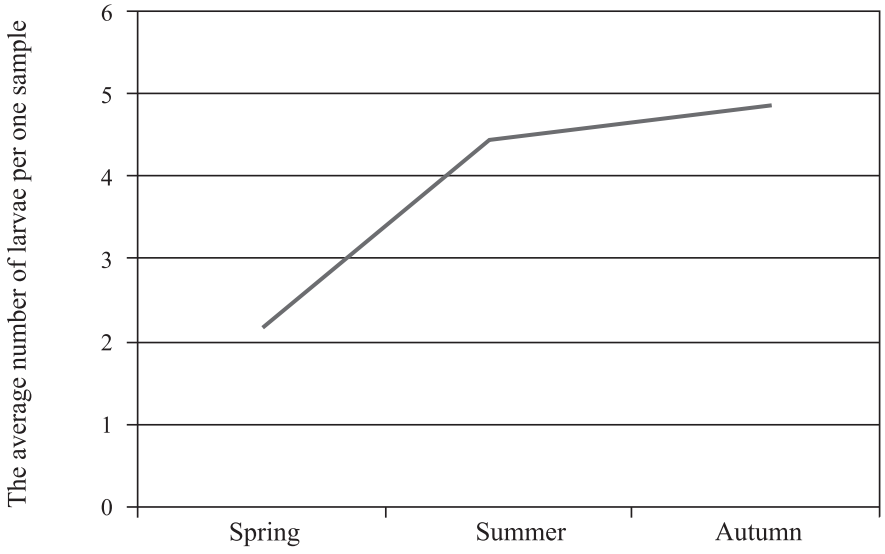

Figure 2. E. bison seasonal infestation with *Dictiocaulus filaria* in Berezinsky Reserve

after antihelminthic treatment. However, later on no grown-up helminthes or their eggs were found with this animal.

A comparison of gathered data with information, obtained by Karasev in mid-1990s, makes it clear that our samples did not contain any nematode species, found by Karasev. Taking these preliminary results into consideration, one may confirm that there are 15 helminth species in Borisov population. Infection with four of these is wide-scale. It is also clear that helminth fauna composition in Borisov population is not that rich as one of Prioksko-Terrasny reserve as well as one in the territory of "Białowieska Forest" nature reserve (Tabl. 2). However it must be noted that in Borisov population helminth fauna was studied only occasionally, and its species composition is not completely defined.

From data presented in Tabl. 2 it can be clearly stated that typical for E. bison helminth species are not characteristic for Borisov population. Parasites that were found there can also be recorded in other hoofed mammals – red deer, roe deer, moose and domestic cattle. Four of parasite species found with Borisov-Berezinsky population (*Bunostomum phlebotomum*, *Dictiacaulus filaria*, *Paramphistomum ichikawai* and *Nematodirus vitulorum*), were recorded in Białowieska Forest and Prioksko-Terrasny reserve. *B. phlebotomum* was occasionally recorded in Mordovsky (Matevosian 1964), Khopersky and Kavkaszky reserves (Kozlov, Nazarova 1979). *D. filaria* were also found in E. bison in the Polish territory of Białowieska Forest (Wroblewski 1927). Literature data about incidence of *P. ichikawai* in E. bison was not found. However, infection with *D. filaria*, *F. hepatica* and *H. contortus* in Borisov--Berezinsky population is wide-spread.

Invasion intensity with the most pathogenic helminth species in E. bison of Borisov-Berezinsky population varies during the spring, summer and autumn periods. Fig. 1 shows the degree of seasonal invasion of *F. hepatica*. It is the lowest during spring, since in that period E. bison are infected only through overwintered shellfish. The highest degree is observed in summer, after that the cercaria larvae develop in shellfish and coming out, become adolescent and lay eggs in pastures and watering places. In autumn the invasion intensity goes down, since in this period shellfish features only redia and embryonic cercaria, which do not cause an infection at pastures and watering places.

The dynamics of E. bison invasion with *D. filaria* nematode follows a similar pattern (Fig. 2). In spring its regularity is low, since a number of overwintered larvae in environment is usually minimal. Therefore, the infection intensity in pastures is low. In this period we registered a bit more than two larvae per sample. The parasite peak number falls to summer, which is followed by an insignificant decrease in number, since in autumn the infection level in pastures and watering places remains high.

The helminths, found within Borisov-Berezinsky population, fall into two groups: nematodes and trematodes. For trematodes, a bridging host is typically a shellfish. The ways of helminths infecting a final host vary, but all of them need high humidity and optimal temperature to finish their lifecycle successfully. These conditions are fulfilled by summer pastures within the territory of Berezinsky reserve and adjacent territories of Borisov forestry enterprise. Forests there are highly waterlogged, packed with small pools, dampened grassy lots, forest springs and small forest rivers. In spring, after snowmelt and sometimes in rainy summers the surroundings are strongly flooded, what contributes to nematode larvae development and spreading of trematode bridging hosts. For *F. hepatica* trematode, that causes fasciolasis and is widely spread in samples, such host is small pond snail *Lymnea truncatula*. This animal may live not only in the water, but also stay out of it for prolonged periods of time, providing that humidity level is sufficient.

The study on pastures and damp forest lots in the region of bison summer area revealed that a small pond snail is registered in small areas, covering from 0.02 to 1% of studied area in open spaces and also covers some spots in temporary forest pools. Here its quantity reaches 5 – 7 specimens per square meter. The extensity of small pond snail infection with cercaria of *F. hepatica* in October of 2004 reached 20% and intensity was up to 300 specimen per a shellfish. The gathered data proves that the studied areas are the main sources of E. bison infection with dictyocaulosis and fasciolasis carriers, that get to bisons' organisms upon visiting temporary watering places and grazing at highly moist forests.

According to literature sources (Beliaeva 1959; Nazarova 1965; Krasochko *et al.* 1996; Kochko 2003; Moskvin *et al.* 2004), the list of helminths, registered in three E. bison populations (Białowieska within the territory of Belarus,

Prioksko-Terrasny and Borisov-Berezinsky) contains 45 species (Tabl. 2). Most of them are trematodes and nematodes, which are parasitizing mainly in digestive tract, lungs and liver of animals.

In Prioksko-Terrasny reserve, where Borisov-Berezinsky population founders were taken from, 18 species of helminths were registered. The most dangerous (in nosologic aspect) species in Prioksko-Terrasny reserve are *Fasciola* sp., *Dicrocelium, moniezia*, whipworms, gastrointestinal tract Strongilata, Capillarids, and possibly, Setaria, which are widely spread and highly invasive (Moskvin *et al.* 2004).

In the territory of Belarus, E. bison have quite diverse helminth fauna. The papers published in the 1990s (Krasochko *et al.* 1996; Kochko *et al.* 2000), point out 15 species of helminths – 11 species of nematodes, 3 species of trematodes and 1 species of cestodes – found in animals from Białowieska forest. E. bison invasion extensity reaches there 91.2%, 79.18% of animals are infected with nematodes, 66.7% – with trematodes and 7.69% – cestodes (Krasochko *et al.* 1996). Summary publication on bison helminthofauna (Kochko 2003) contains a list of 41 species, recorded within the last century in E. bison in both Belarusian and Polish parts of the Białowieska forest.

In new ecological situation, as a rule, animals lose a part of parasite species diversity due to absence of intermediary hosts, change of humidity and ambient temperature. However, the helminthofauna can be supplemented with parasites, characteristic for domestic and wild animals of the region. For example, about 50% of helminths of E. bison, living in Białowieska forest, are typical for the livestock. This is conditioned by the fact that domestic animals in the national park use the pastures, which are occasionally visited by bison.

B. phlebotomum was detected with bison in the Belarus territory for the first time. As for *C. oncophora, N. helvetianus* and *O. ostertagi*, these are common for animals from Białowieża population.

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Helmintofauna żubra z wolnej populacji Borisov-Berezinsky

Streszczenie: Przedstawiono dane o ilościowej strukturze fauny pasożytniczej stwierdzanej u żubrów (*Bison bonasus bonasus*) z wolnożyjącej populacji Borisov-Berezinsky. Stopień poziomu infekcji zależnie od sezonu zbadano dla różnych gatunków pasożytów. Opisano drogi wniknięcia do organizmu żywiciela gatunków pasożytów najczęściej spotykanych, tj. *Fasciola hepatica* i *Dictyocaulus filaria*.