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Original papers

The Alice – „Follow the White Rabbit" – parasites of farm rabbits based on coproscopy[1]

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ABSTRACT. The aim of the study, conducted in the years 2011–2013, was to determine the level of gastrointestinal parasites infection in New Zealand White rabbits, kept at the Experimental Station of the University of Agriculture in Krakow. The study showed rabbits protozoan infection with the genus *Eimeria*, belonging – based on the sporulation method – to the following species: *E. magna*, *E. media*, *E. perforans*, *E. stiedae* and *E. irresidua*. The highest prevalence of infection, as well as the intensity of oocysts output (OPG – oocysts per gram of faeces), was noted for *E. magna* and *E. media* – respectively 31.4 % (19477.3 OPG), and 40.0 % (14256.07 OPG). The infection of rabbits with *Eimeria* spp. differed significantly between years. With regard to oocysts output, the level of infection was strongly connected with the age of rabbits, being higher in young animals. However, the range of infection was highest among adults. Among nematodes, *Passalurus ambiguus* pinworm was regularly found (prevalence reached 21.9%), other species – *Trichuris leporis*, and *Graphidium strigosum* were rarely noted. The overall infection with nematodes did not differ between years. Similarly, as in the case of *Eimeria* older individuals were more often infected by nematodes. We observed some trends in parasite oocysts/eggs output; the protozoan oocysts were recorded more often in faecal samples collected in the evenings, whereas the nematodes eggs occurred frequently in the mornings. This situation may be related to the phenomenon of coprophagy occurring in the mammals of Lagomorpha order. The results of the study indicate that especially coccidiosis constitute permanently throughout the years an important problem in the rabbitry examined.

Key words: coccidia, nematodes, rabbits, faecal examination

Introduction

The parasites in rabbit farms are pathogenic. Among parasitoses, the most important is coccidiosis caused by protozoa from *Eimeria* genus. The coccidian, due to the direct and short developmental cycle, can quickly spread in the traditional farming units, as well as in large meat farms of those animals. These parasites multiply in the cells of small intestine but in the case of *E. stiedae* – liver hepatocytes. The infection has dynamic course and because of the intense diarrhoea, leads to dehydration and consequently death of animals. Strong infections of coccidia cause vast economic losses in rabbit farms. Gastro-intestinal nematode infections can lead to dangerous diseases, also mortal, with clinical symptoms like anaemia (hematophagic species), and discomfort such as in case of infection by *Passalurus ambiguous*.

The main aim of this study was to define the level of parasite infection in New Zealand white rabbit herd during the period of the greatest risk of parasite occurrence, in the context of improving animal welfare, as suggested by the European Union regulations.

Material and Methods

The research was conducted on New Zealand White breed of rabbits between June of 2011 and

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Table 1. The results of coproscopical examination of rabbits in the years 2011–2013

Parasite	Parameters	2011	2012	2013	Total
	No. examined	70	22	40	132
Eimeria spp.	P [%]	94.3	100	90	93.9
	I [OPG mean (min–max)]	11608.3 (100 – 57000)	4536.4 (50 – 23050)	3508.1 (50 – 21360)	8001.4 (50 – 57000)
Passalurus ambiguus	P [%]	15.7	9.1	40.0	21.9
	I [EPG mean (min–max)]	254.6 (100 – 500)	50.0 (50)	243.1 (15 – 1600)	234.1 (15 – 1600)
Trichuris leporis	P [%]	0	36.4	0	6.1
	I [EPG mean (min-max)]	0	328.8 (15 – 750)	0	32.7 (15 – 750)
Graphidium strigosum	P [%]	2.9	0	0	1.5
	I [EPG mean (min–max)]	100.0 (100)	0	0	100.0 (100)

Explanation: P – the prevalence of infection; I – the OPG/EPG output: the mean, and in the parentheses range

2013, in a rabbitry at the Experimental Station of the University of Agriculture in Krakow. The animals were housed in the cages on slatted floor, with nesting boxes placed in. The animals were fed the forage of farm origin with the feeders and drinkers placed directly on the floor. Sulfatyf, as the coccidiostat, was applied once, after weaning, in drinking water. Adult males as well as the offspring were kept in the same management system.

In coproscopy, we used a quantitative McMaster method with centrifugation, and saturated NaCl with sucrose as the flotation solution, to determine the prevalence of infection (P, %), and the intensity of coccidian oocyst or nematode eggs output in 1 gram of host faeces (I, OPG/EPG; the mean and the range: min – max). The isolation and sporulation of oocysts was performed in potassium dichromate 2% solution, using the method described by Coudert et al. [1]. Identification of a particular species of coccidia was done according to the dimensions and morphology of oocysts and sporocysts [2]. Nematodes were diagnosed in accordance with Thienpont et al. [3].

The proportion of infected and uninfected with

Table 2. *Eimeria* species diversity in the rabbits based on the sporulation in the year 2011

Coccidian species	Parameters based on coproscopy	Values based on sporulation
	No. examined	70
*Eimeria magna**	P [%]	31.4
	I [OPG mean (min–max)]	19477.3 (2000 – 56400)
*Eimeria media**	P [%]	30.0
	I [OPG mean (min–max)]	14256.1 (100 – 56400)
Eimeria perforans	P [%]	2.9
	I [OPG mean (min–max)]	750.0 (750)
Eimeria stiedae	P [%]	2.9
	I [OPG mean (min–max)]	100.0 (100)
Eimeria irresidua	P [%]	1.4
	I [OPG mean (min–max)]	50.0 (50)

Explanation: see Table 1; * – 2 species only were taken into account

Table 3. The level of parasite infection in rabbits in relation to daytime sampling, based on faecal samples collected in the morning and evening (year 2011)

Parasite	Parameters	Morning sampling	Evening sampling
	No. examined	15*	55*
Eimeria spp.	P [%]	100	92.7
	I [OPG mean (min–max)]	7523.3 (150 – 57000)	12809.8 (100 – 56400)
Passalurus ambiguus	P [%]	66.7	1.8
	I [EPG mean (min–max)]	250.0 (100 – 500)	300.0 (300)
Trichuris leporis	P [%]	0	0
	I [EPG mean (min–max)]	0	0
Graphidium strigosum	P [%]	13.3	0
	I [EPG mean (min–max)]	100.0 (100)	0

Explanation: see Table 1; * – the number of collected faecal samples varies as the result of difficulty in obtaining the faecal material

Table. 4. Gastrointestinal parasites infection according to the age group of rabbits in the years of study

Parasite	Parameters	Young animals	Adult animals
	No. examined	39	93
Eimeria spp.	P [%]	100	91.4
	I [OPG mean (min–max)]	10539.7 (100 – 46200)	6836.7 (50 – 57000)
Trichuris leporis	P [%]	3.8	2.3
	I [EPG mean (min–max)]	356.0 (15 – 750)	283.3 (250 – 300)
Passalurus ambiguus	P [%]	5.3	16.7
	I [EPG mean (min–max)]	140.0 (15 – 500)	264.1 (15 – 1600)
Graphidium strigosum	P [%]	0	1.5
	I [EPG mean (min–max)]	0	100.0 (100)

Explanation: see Table 1

Eimeria spp. and nematodes was compared by χ^2 test in relation to type of infection, age of rabbits, and year of conducted research. Statistical analysis was performed using the Statistica 9.0.

Results

The studies showed that the examined rabbits were infected with coccidia of *Eimeria* genus, as well as with nematodes in total (Table 1). The rabbit were more often infected with *Eimeria* spp. than nematodes (χ^2=45.65 P<0.001) (Table 5).

Protozoan infection was reported most often in 2011. Due to very high intensity of infection, in order to determine the species composition, the sporulation of *Eimeria* oocyst was carried out. *E. magna* and *E. media* were found to be the most dominant, followed by *E. perforans*, *E. stiedae* and *E. irresidua* (Table 2). In addition, we found out that the difference between infection with *Eimeria* spp. in relation to year of research was significant (χ^2=24.45 P<0.001) and more young individuals had coccidian (χ^2=17.06 P<0.001) (Table 5).

Among the nematodes, the pinworm *Passalurus ambiguus* was found regularly (Table 1). In contrast to protozoan infection we did not find out significant difference between number of infected rabbits within years 2011 and 2013 (χ^2=1.95

Table 5. Results of χ^2 test based on presence/absence of infection with nematodes and *Eimeria* spp. in relation to age of rabbits, type of infection, and year of research (P – level of significance)

		Eimeria spp.	Nematodes	χ^2	P
		No. infected individuals	No. infected individuals		
	Adults + young	124	38	45.65	0.001
Age of rabitts	Adult	85		17.06	0.001
	Young	39			
	Adult		27	12.71	0.001
	Young		11		
Year of research	2011	66		24.45	0.001
	2012	22			
	2013	36			
	2011		13		
	2012		9	1.95	0.50
	2013		16		

P=0.50) (Table 5), whereas more adult individuals were infected with nematodes than younger (χ^2=12.71 P< 0.001) (Table 5).

We compared protozoa oocysts and nematode eggs output in relation to the daytime of sampling (morning or evening hours) (Table 3). In general, the samples collected in the evening were more often positive for coccidia than those collected in the morning, which was opposite to the nematodes (Table 3). The level of coccidia infection (OPG values) were associated with the age of rabbits, and the young were more infected, which is the common phenomenon of coprophagy for this group of parasites (Table 4).

Discussion

The studies on the rabbits infection were carried out in our research before [4–6] but in the presented farm only once [4]. In the previous studies, we showed a higher level of coccidia infection in young animals (OPG), particularly observed in early–summer months, which could be the results of favourable conditions for sporulation of oocysts under farm conditions (temperature and humidity) [4]. The increase in the infection of coccidia may be also connected with stress, associated with weaning time. Balicka-Ramisz [7] confirmed this relation in young rabbits, aging 35 days, which had the highest level of the protozoan infection. The present study did not show the most pathogenic species of intestinal coccidia, i.e., *E. flavescens* and *E. intestinalis*, found previously in the bigger and intensively managed warrens [2] similarly to Balicka-Laurans [7,8]. However, although the previous research showed the lack of *E. stiedae* in the herd presently examined [3] now the pathogenic hepatitic coccidian species occurred.

Keeping rabbits in the wire cage system can reduce the transmission of parasites, but in the case of protozoa – due to their short life cycle the transmission still may occur. The welfare of the animals, even at ensuring high zoo-hygienic standards, cannot stop the transmission of species from *Eimeria* genus.

In the present work, the most frequently reported nematode was *Passalurus ambiguus*. Pinworms are characterized by a specific development cycle, with the possibility of retroinvasion (developing larvae can return to the host, following hatching from eggs laid by gravid females around the animal anus). This situation may explain the most frequent occurrence of these parasites in the rabbit management. The occurrence of other nematodes (*Trichuris leporis* and *Graphidium strigosum*) was rare. Their occasional presence is associated with the type of feeding, i.e., green forage use (grass, vegetables). The management system of examined rabbits can

also explain the lack of other nematode i.e., *Trichostrongylus retortaeformis*, which larvae develop to the invasive stage in green forage from pastures. However, Nosal et al. [5] observed this nematode with a low infection in one small farm (southern Poland) where green forage was used. The confirmation of this thesis is the frequent occurrence of this nematode in hares *(Lepus europeaus)* in Poland [9–12].

In the breeding farm of rabbits the most important problem is low zoo-hygienic management system that contributes to the transmission of infectious and invasive diseases (large herds of animals). In rabbits kept in wire cages, the infection with parasites from genus *Eimeria* and pinworm *Passalurus ambiguous* may be the most common.

The results of the study indicate that especially coccidiosis constitute permanently throughout the years an important problem in the rabbitry examined. What next with the parasitoses in rabbits in context of EU Regulation (No 1831/2003) limiting the use of anti-parasitic drugs (especially as fodder supplements). Can we get back to the traditional methods of parasite control in the context of emerging drug resistance in different livestock species? May drugs made from herbal formulations be useful in reducing the level of parasite infections [5]? Or should we stop using them in prophylaxy at all (synthetic and natural).

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