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## *Eimeria* Species (Apicomplexa: Eimeriidae) Infecting *Eliomys quercinus* in an Alpine Habitat

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ABSTRACT: Coccidian parasites were detected in an Alpine population of the garden dormouse (*Eliomys quercinus*), and 55–82% of the fecal samples collected during a two-year study (2000 and 2002) contained one or two eimerian species. We report the presence of *Eimeria myoxi* and confirm for the first time the presence of *Eimeria melanuri* in the garden dormouse. These *Eimeria* species can be considered common parasites of the garden dormouse and the Asian garden dormouse. The high prevalence might be due to group hibernation by the dormice.

*Key words: Eimeria melanuri, Eimeria myoxi,* garden dormouse, host parameters, parasite interaction.

The garden dormouse (Eliomys quercinus) is a nocturnal rodent distributed from the Iberian Peninsula, through Italy and some Mediterranean islands, to central and eastern Europe, and the Ural Mountains. The species is found in wooded areas, hedgerows, and rocky places. It shelters in hollow trees, in the branches of trees or shrubs, and in rocky crevices. In some areas dormice are highly arboreal, but in other areas they prefer to move on the ground. Their diet consists of acorns, nuts, fruits, insects, and other small animals. They generally gain weight in late summer and autumn, and hibernate from October to April. Groups of individuals can hibernate in the same cavity.

Coccidian parasites of the garden dormouse are poorly known. The only species reported in the literature is *Eimeria myoxi*, described for the first time by Galli-Valerio (1940) and also found by Pellerdy (1974). Golemanski and Darwish (1993) reported the presence of *E. myoxi* and *E. melanuri* in the Asian garden dormouse (*Eliomys melanurus*). This species, present in northern Africa and the Middle East, was recently separated from *Eliomys quercinus*  on the basis of allozyme data (Filippuci et al., 1988).

Previous studies of the coccidia of the genus *Eliomys* have analyzed only a few animals. Here we describe the coccidian species in an Alpine population of the garden dormouse, including their patterns of prevalence.

The study was carried out in the Champdepraz Valley (northwestern Alps, Italy, 45°36'N, 7°21'E). The study area was a Scots pine (*Pinus sylvestris*) woodland at about 1,300 m above sea level. In the lower part of the woodland, hazels (*Corylus avellana*) were also present.

Dormice were livetrapped for seven days each month from May to September 2000, during a capture and recapture study, and in June 2002 when animals were captured for a radiotracking project. Animals recaptured in different months in 2000 or in different years were considered only once (first capture) for the parasitological analysis. One hundred Sherman livetraps were set 20 m apart. The traps were baited with hazelnut cream and apple, and provided with bedding material. Each trapped dormouse was individually marked with a numbered ear tattoo, and its sex and age were recorded (Bertolino et al., 2001).

After a trapped animal was released, its feces were collected from the trap and placed in a vial containing 2.5% (w/v) aqueous potassium dichromate (K<sub>2</sub>Cr<sub>2</sub>O<sub>7</sub>). The suspension was maintained at room temperature for at least 2 wk to allow sporulation of oocysts. In the laboratory, a fraction of each sample was smeared on a microscope slide; oocysts were examined using an oil immersion  $100 \times$  objective with a Nomarski differential interference con-



FIGURE 1. Sporulated oocysts: a) *Eimeria melanuri*; b) *Eimeria myoxi*.

TABLE 1.. Oocyst and sporocyst measures for the two eimerian species found in the garden dormouse.

	E. myoxi n = 64		E. melanuri $n = 62$	
	$Mean \ (\mu m)$	SD	$Mean \; (\mu m)$	SD
Oocyst length	18.87	1.20	22.40	0.92
Oocyst width	17.29	1.03	19.85	0.94
Sporocyst length	8.01	1.14	9.32	1.35
Sporocyst width	5.99	0.74	7.12	1.20

trast system on a Leitz microscope. Oocysts were measured with a calibrated ocular micrometer and photographed with a SONY ExwaveHAD digital camera.

Species identification was based on oocyst size, shape, and internal structure. Golemansky and Darwish (1993) was used as a reference for oocyst identification. Single measures (oocyst and sporocyst length and width) were not sufficient to separate all the oocysts into the two species because of the overlapping of central values in the dimensional ranges (Golemanski and Darwish, 1993). Thus, a classification system considering the four measures together was necessary. We used Kmeans clustering to separate the oocysts into two groups (species). This method allows one to separate a set of data into a user-specified number of clusters that are as distinct as possible. Computationally, the program starts with K random clusters and then moves objects (oocysts) between those clusters with the goal of minimizing variability within clusters and maximizing variability between clusters. To evaluate the appropriateness of the classification, we used ANOVA (analysis of variance) to compare the between-cluster variability with the within-cluster variability, computing the significance test for the hypothesis that the means of each cluster for each dimension (oocyst and sporocyst measures) in the groups were different from each other and evaluating how distinct the clusters were. A discriminant function analysis (DFA) was then performed on the four character variables (oocyst and sporocyst length and width) to evaluate the degree

of correct classification (Gardner and Duszynski, 1990). Mahalanobis distances were used for all comparisons of discrimination (Manly, 1986). Representative parasites were accessioned in the Museo Regionale di Scienze Naturali, Turin, Italy (accession numbers: MRSN Pr1, MRSN Pr2).

We define prevalence as the number of infected hosts divided by the number of hosts examined (Bush et al., 1997).

*Eimeria* oocysts were found in the fecal samples of 14 of the 17 dormice trapped in 2000 and six of the 11 in 2002. The prevalence in males (7/11) and females (13/17) did not differ (Fisher Exact test P=0.67, data from the two years pooled).

In total, we measured 126 oocysts that were classified into two species-groups  $(n_1=64, n_2=62)$  by means of K-means clustering. All the ANOVA tests comparing between-cluster variability with withincluster variability for oocyst and sporocyst length and width were significant (P<0.0001). The two species-groups differed significantly (DFA Wilks' lambda *F*value=88.80, d.f.=4, 121, P<0.0001). Oocyst length and width contributed most to the discriminating function. Using the classification functions, we correctly classified 96.8% of the oocysts previously assigned to one of the two species.

The measures of the two species (Table 1) fall within the ranges reported for *E. myoxi* and *E. melanuri* (Fig. 1) (Golemanski and Darwish, 1993). Using the results of these analyses, we attributed each oocyst to one of the two species and then recorded the presence or absence of each coccidian species for each fecal sample.

The prevalence of *E. melanuri* was 82.4% in 2000 and 36.4% in 2002. *Eimeria myoxi* was present in 64.7% of dormice in 2000 and 54.6% in 2002. All dormice infected by *E. myoxi* in 2000 and by *E. melanuri* in 2002 were also infected by the other species (Fisher Exact test for paired infection P < 0.05). However, the differences in prevalence must be viewed with caution because of the different months of sampling in the 2 yr.

Therefore, we found in the garden dormouse the two *Eimeria* species that Golemanski and Darwish (1993) reported in the Asian garden dormouse. Those authors suggested that the oocysts found in the garden dormouse by Pellerdy (1974) and attributed to *E. myoxi* should instead be considered *E. melanuri* on account of their dimensions. We confirm for the first time the presence of *E. melanuri* in the garden dormouse.

Host specificity of coccidian parasites can be rather broad. There are many reports of eimerian parasites shared among species of the same genus and even among genera (Shults et al., 1990; Patrick and Wilson, 1995). The exchange of parasite species among closely related host species is considered an important factor in maintaining the stability of eimerian assemblages (Seville and Stanton, 1993). On the basis of previous reports of coccidia in the genus *Eliomys* and the results of our study, we conclude that E. myoxi and E. melanuri are common parasites of the garden dormouse and the Asian garden dormouse. It might be possible to find other rarer and more localized eimerian species by investigating new areas throughout the range of the genus *Eliomys* and by increasing the sample size of analyzed animals.

The occurrence of coccidian parasites in the garden dormouse was very high in 2000, when 82% of the fecal samples contained at least one eimerian species. The high prevalence of parasites could have been due to group hibernation by the dormice (Bussy, 1968). Moreover, there might have been a density-dependent effect, but this could not be tested with our data. Other factors, such as the past history of parasite infection, might have played a role in determining the high prevalence. However, larger sample sizes are required to thoroughly investigate the factors responsible for different levels of prevalence.

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