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Coprological study on intestinal helminths in Swiss dogs: temporal aspects of anthelmintic treatment

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Abstract Coproscopic examination of 505 dogs originating from the western or central part of Switzerland revealed the presence (prevalence data) of the following helminths: *Toxocara canis* (7.1%), hookworms (6.9%), *Trichuris vulpis* (5.5%), *Toxascaris leonina* (1.3%), Taeniidae (1.3%), *Capillaria* spp. (0.8%), and *Diphyllobothrium latum* (0.4%). Potential risk factors for infection were identified by a questionnaire: dogs from rural areas significantly more often had hookworms and taeniid eggs in their feces when compared to urban family dogs. Access to small rodents, offal, and carrion was identified as risk factor for hookworm and Taeniidae, while feeding of fresh and uncooked meat did not result in higher prevalences for these helminths. A group of 111 dogs was treated every 3 months with a combined medication of pyrantel embonate, praziquantel, and febantel, and fecal samples were collected for coproscopy in monthly intervals. Despite treatment, the yearly incidence of *T. canis* was 32%, while hookworms, *T. vulpis*, *Capillaria* spp., and Taeniidae reached incidences ranging from 11 to 22%. Fifty-seven percent of the 111 dogs had helminth eggs in their feces at least once during the 1-year study period. This finding

implicates that an infection risk with potential zoonotic pathogens cannot be ruled out for the dog owner despite regular deworming four times a year.

Introduction

The close contact between dogs and humans harbors the risk of transmitting zoonotic agents. Thus, the ascarid worm *Toxocara canis* is considered to be one of the most frequent canine parasites that represents a considerable health risk especially for children. The nematode has a worldwide distribution, including countries with a high hygienic standard (Magnaval et al. 2001). In Switzerland, surveys on *Toxocara* infections in humans resulted in seroprevalences of 2.7–6.5% (Sturchler et al. 1986; Jacquier et al. 1991). Other zoonotic intestinal helminths in dogs prevalent in central Europe include the taeniid worms *Echinococcus multilocularis* and *Echinococcus granulosus*, the causative agents of alveolar and cystic echinococcosis, respectively (Eckert and Deplazes 2004). *E. multilocularis* is widespread in foxes of central Europe and increasingly includes dogs as a definitive host (Gottstein et al. 2001). Conversely, *E. granulosus* has practically disappeared in intermediate hosts in Switzerland and is only rarely found upon immigration or importation of infected dogs originating from endemic areas such as the Mediterranean basin (Eckert 1997). Other helminths that can be detected upon coprological analyses in Switzerland may affect the health status of the infected dogs, e.g., *Trichuris vulpis* or *Ancylostoma caninum*, whereas others exhibit only minor pathological potential, e.g., *Uncinaria stenocephala*, *Capillaria* spp., or *Diphyllobothrium latum*.

A major component for the spread of these parasites is the shedding of eggs via dog. Measures to control the infection risk for humans or to interrupt the cycle within the dog population therefore focus on (1) appropriate deworming strategies of the dogs and (2) minimization of the risk of fecal contamination in public places (e.g., playgrounds). For the latter, dog owners are requested to collect and

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remove the dog feces, whereas veterinarians have to advise dog owners about medication and frequency of anthelmintic treatments. In the present study, we used a questionnaire to document the deworming concept of dog owners and analyzed the efficacy of their treatment concept upon coprological examination of dogs while entering the study (prevalence). Subsequently, the incidence of infection was analyzed upon an anthelmintic treatment strategy followed for a period of 12 months.

Materials and methods

Dogs

A total of 505 dogs originating from the western or central part of Switzerland (cantons of Fribourg, Bern, and Zurich) participated in this study. The majority ($n=470$) was arbitrarily selected by veterinarians, with the owners' consent. A general examination of the health status was carried out in the veterinary practice, and a fecal sample was rectally collected at the same time point. Thirty-five animals within this group were dairy farm dogs, and the dog owners were asked to collect fresh fecal samples by themselves.

Anthelmintic treatment of dogs

The veterinarians who participated in the present study selected 111 healthy dogs from the above mentioned group, and the owners complied to apply a defined anthelmintic treatment schedule for a period of 12 months. The dogs were perorally treated with a combination of pyrantel embonate, praziquantel, and febantel every 3 months (Drontal Plus, Bayer, Germany). The dosage was according to the manufacturer's instructions.

Coprological analyses

All fecal samples were tested by a standardized flotation method according to Rommel et al. (2000). This technique uses zinc chloride in a concentration of 44% (w/v) and is

suitable for the coproscopic detection of helminth eggs exhibiting a specific weight of less than 1.2.

Fecal samples were further analyzed for the presence of intestinal *Echinococcus* sp. with an *Echinococcus* coproantigen ELISA (Deplazes et al. 1999). If taeniid eggs were found, these were further isolated and purified by a combination of sequential sieving and flotation in zinc chloride solution. Purified eggs were then subjected to an *E. multilocularis*-specific PCR (Mathis et al. 1996). For the present study, speciation will not be addressed for data evaluation, and we will refer only to the family of Taeniidae. We considered this acceptable by two reasons: (a) private clinics do not diagnostically discriminate taeniid eggs in view of speciation, and (b) praziquantel used in this study does identically affect all taeniid species known to occur in Swiss dogs.

All dogs that participated in the anthelmintic treatment study were followed up monthly in the veterinary practices. A clinical examination and collection of fecal samples was done as outlined above for entering the study.

Questionnaire

The dog owners had to fill out a questionnaire to provide information on the husbandry system [farm dog (including dogs from rural areas with access to farms), urban (family) dog], feeding habits, and anthelmintic treatment. Questionnaire parameters are outlined in "Results".

Statistical analyses

Coprological results (positive/negative) and risk factor information collected from questionnaires were cross-tabulated and analyzed using a two-tailed Fisher's exact test (FET; 2×2 tables) or chi-square test (CST, $2 \times j$ tables). To identify risk factors for overall helminth infestation, a multivariable logistic regression (LR) approach was used (NCSS 2004; <http://www.ncss.com>). All risk factors with P values less than 0.20 in the univariable analysis were jointly entered into the LR model. Subsequently, non-significant variables were removed as risk factors, together with all other factors of interest tested for their confounding

Table 1 Detection of single and multiple parasite infections in 505 Swiss dogs

Number of parasite species detected	Negative Number of dogs	Toxoc	Toxas	Hookw	Trich	Capill	Diphy	Taeni	Total
4	–	1	1	1	1	–	–	–	1
3	–	2	–	2	1	–	–	1	2
2	–	7	2	7	5	1	–	4	13
1	–	26	4	25	21	3	2	2	83
0	406	–	–	–	–	–	–	–	406
Total	406	36	7	35	28	4	2	7	505

Number of parasite species detected in the fecal samples of 505 dogs

Toxoc *Toxocara canis*, *Toxas* *Toxascaris leonina*, *Hookw* hookworm, *Trich* *Trichuris vulpis*, *Capill* *Capillaria* spp., *Diphy* *Diphyllobothrium latum*, *Taeni* Taeniidae

Table 2 Influence of age of Swiss dogs on the prevalence of helminth eggs in Swiss dog feces

Age	Negative Number of dogs	Toxoc	Toxas	Hookw	Trich	Capill	Diphy	Taeni	Total
<1	16	0	0	1	1	0	0	0	18
1–6	224	23	4	23	15	3	1	4	297
>6	138	9	3	9	10	1	1	2	173
Total	378	32	7	33	26	4	2	6	488
<i>P</i> values	–	0.27	0.83	0.52	0.96	0.81	0.90	0.87	–

Age in years; abbreviations of parasites as listed in Table 1. For individual parasites, a chi-square test was performed. *P* values less than 0.05 are considered statistically significant

effect on the remaining risk factors [more than 10% change in the odds ratio (OR) estimates]. Finally, two-way interactions between all remaining variables in the model were assessed for their statistical significance. In all statistics, *P* values less than 0.05 were considered significant, and those less than 0.01 were considered highly significant.

Results

First sample analysis (prevalence) and questionnaire

From 505 examined dogs (prevalence study), 99 (19.6%) had at least one helminth species detectable in their feces. The most commonly found parasites were *T. canis* (6.9%), hookworms (6.9%), *T. vulpis* (5.5%), *Toxascaris leonina* (1.3%), Taeniidae (1.3%), *Capillaria* spp. (0.8%), and *D. latum* (0.4%). Multiple infections with four, three, or two different species were found in 16 dogs (Table 1). From seven dogs with taeniid eggs, one was positive only in the *E. multilocularis* coproantigen ELISA, and one was positive for both coproantigen ELISA and *E. multilocularis*-specific PCR.

Addressing a potential age-related distribution of the parasites, no respective findings could be shown. The different helminth species were found in all age categories, and a chi-square test did not reveal significant differences, neither for qualitative grouping (helminth positive/negative; *P*=0.5) nor for individual parasites (Table 2; due to the small numbers in several categories, the obtained values have to be interpreted with caution).

Based on the questionnaire, 26.9% of dogs that demonstrated the presence of helminth eggs in their feces were farm dogs, whereas the respective prevalence for dogs from

urban areas was 16.6%. The difference between farm and urban dogs was significant (FET *P*=0.016). At the individual worm level, significant differences were observed for hookworms (*P*<0.01) and Taeniidae (*P*=0.015), while the difference between *T. canis* (*P*=0.82), *T. leonina* (*P*=0.68), *T. vulpis* (*P*=0.67), *Capillaria* spp. (*P*=0.66), and *D. latum* (*P*=1.0) was not significant.

Only a minority of the dogs (12.7%) did not receive any anthelmintic treatment, whereas 73.1% were treated once or twice a year. Additional treatments were given to 13.7% of the dogs every second month, while three dogs were treated monthly (0.5%; Table 3). The frequency of treatment did have little effect on the presence of helminth eggs in dog feces and only resulted in a significant reduction of hookworm eggs (CST *P*=0.018), while no significant difference was observed for other parasites (Fig. 1).

The dog owners had to categorize the feeding behaviors of their dogs. Of the 398 dogs, 78 (19.6%) regularly received fresh uncooked meat. The prevalence of helminths in such dogs (26.9%) was not significantly higher than in dogs receiving uniquely canned or cooked food (16.6%; FET *P*=0.27). The FET was also performed for individual helminthes and gave no *P* values lower than 0.1 for *T. canis*. (*P*=0.119), *T. leonina* (*P*=1.0), hookworms (*P*=0.823), *T. vulpis* (*P*=0.613), *Capillaria* spp. (*P*=0.584), *D. latum* (1.0), and Taeniidae (*P*=0.335).

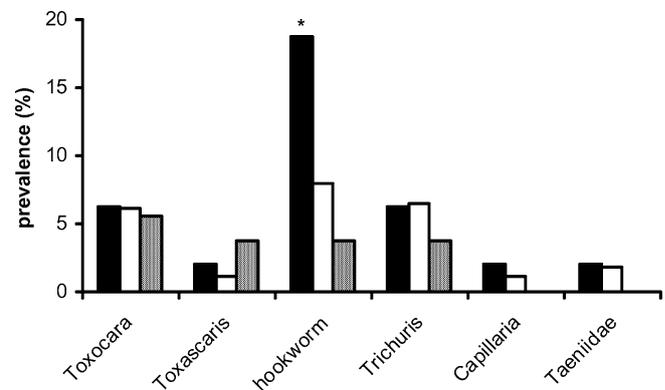
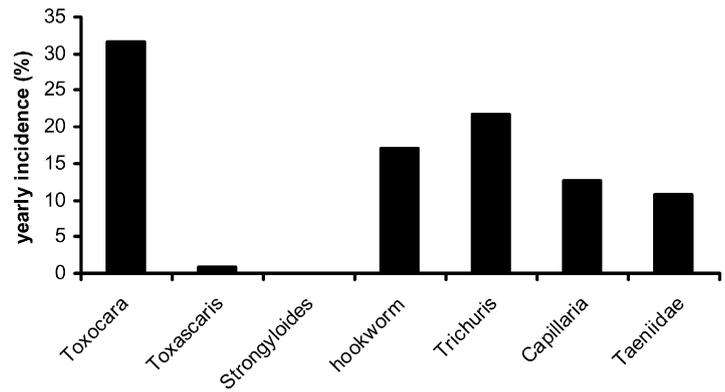


Fig. 1 Prevalence of helminth eggs in Swiss dog feces in dependence of anthelmintic treatment (based on the questionnaire). Findings of the first coprological analysis (*n*=379). Frequency of treatment based on the questionnaire: no treatment (black bar), once to twice a year (open bar), more than twice a year (grey bar). Significant differences are marked with an asterisk

Table 3 Effect of anthelmintic treatments (based on the questionnaire) on the detection of helminth eggs in Swiss dog feces

	Helminthes		Total
	+	–	
Frequency of deworming (per year)			
0	16 (33%)	32 (67%)	48
1–2×	51 (18%)	226 (82%)	277
>2×	9 (17%)	45 (83%)	54
Total	76	303	379

Fig. 2 Yearly incidence of helminth eggs in Swiss dog feces after treatment every 3 months with pyrantel embonate, praziquantel, and febantel ($n=111$)



According to the dog owners, 60.9% of the dogs were not permanently under control and had the possibility to hunt small rodents, had access to offal and carrion, or were eating garbage. Among these dogs, the prevalence of helminth eggs was 25.1%. The prevalence in “controlled” dogs was 14.1%, which is significantly lower (FET $P=0.008$). At the individual helminth level, only hookworm eggs (FET $P=0.009$) were significantly more often detected in “uncontrolled” dogs, while no difference was found for *T. canis* ($P=0.140$), *T. leonina* ($P=0.411$), *T. vulpis* ($P=0.413$), *Capillaria* spp. ($P=0.160$), *D. latum* ($P=0.152$), and Taeniidae ($P=0.085$).

In the final multivariable LR model, deworming (coded yes/no) was identified as a protective factor for detection of helminth eggs in dog feces [OR 0.52, 95% confidence interval (CI) 0.26–1.04], while uptake of offal, carrion, or garbage was a risk factor (OR 2.12, 95% CI 1.08–4.14). The variable farm dog remained in the model as a confounder but was not significantly associated with the helminth status ($P=0.22$).

Effects of regular anthelmintic treatment

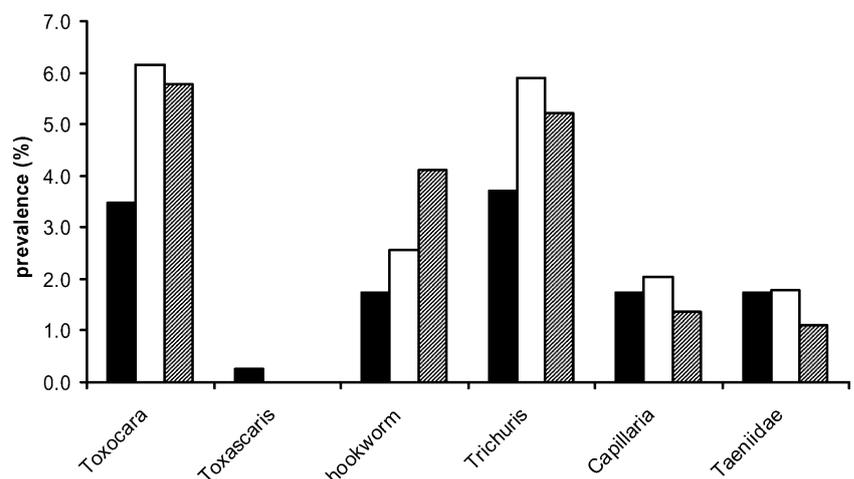
A group of 111 dogs was treated every 3 months with combined medication of pyrantel embonate, praziquantel, and febantel. The yearly incidence of *T. canis*, hookworms,

T. vulpis, *Capillaria* spp., and Taeniidae was between 11 and 32% (Fig. 2). Sixty-three dogs (56.8%) had helminth eggs in their feces at least once a year. The treatment did not result in a significant reduction of egg secretion, neither for nematodes nor for cestodes, as determined by chi-square test. Fig. 3 shows the prevalence of the individual parasites detected 1, 2, and 3 months after treatment (mean prevalence of four samples collected during the 1-year period).

Discussion

Coprological analysis of samples from 505 healthy dogs provided an insight into the distribution and occurrence of intestinal parasites in Swiss dogs. These findings show similar parasite patterns as described by others, with the exception of lacking lungworm (Deplazes et al. 1995; Barutzki and Schaper 2003; Epe et al. 2004). In the present study, the funnel technique according to Baermann-Wetzel, which is specific for detection of viable larvae, was not used. According to other authors, *T. canis* and hookworms are the most commonly found helminths in dogs. However, the parasite prevalence was lower in our study than in samples obtained from dogs at the veterinary hospital for routine diagnosis or samples collected from stray dogs (Deplazes et al. 1995; Barutzki and Schaper 2003). Interestingly, there was no age-related distribution

Fig. 3 Prevalence of helminth eggs in Swiss dog feces after treatment every 3 months with pyrantel embonate, praziquantel, and febantel ($n=111$). Samples were collected in monthly intervals for 1 year. Bars represent the percentage of positive dogs for the specified parasite: 1 month (black bar), 2 months (open bar), and 3 months (grey bar) after drug application. The respective coprological specimen was collected at the next time point when the next treatment was administered



of the parasites. Especially *T. canis*, which is described as typical parasite of pups, was not found in animals of age less than 1 year (Scothorn et al. 1965). This may be partially explained by optimal anthelmintic treatment that dog owners apply especially in the early phase of life, as all dogs in this age segment received at least one treatment. Furthermore, the low number of pups included in the study may lead to an underestimation of parasite detection and makes a statistical analysis rather difficult. It was shown that treatment had a slight positive effect in reducing the detection rate of helminthes, which was even significant for hookworms. However, more frequent treatment did not necessarily result in a further decrease of parasite detection. A quantification of parasitic stages in the feces was not done, nor was information available on the active substance used and the form of application. Interpretation on partial effects and dosage problems was therefore not possible.

Additional information became available from the controlled treatment with a combined application of pyrantel embonate, praziquantel, and febantel every 3 months. Surprisingly, despite regular treatment, the yearly incidence of *T. canis* was 32%, while hookworms, *T. vulpis*., *Capillaria* spp., and taeniid eggs were detected in 11–22% of the dogs at least once a year. As the prepatency period of the mentioned parasites is less than 3 months, it can be assumed that reinfection most likely occurred. However, a comparison of parasitological findings 1, 2, and 3 months after treatment did not result in significant differences for individual parasites, although we observed a tendency toward a lower parasite detection rate in the first month compared to the subsequent months (*T. canis*, hookworms, and *T. vulpis*). It cannot be excluded that treatment does not completely eliminate the parasites, and helminth eggs still may be detected in fecal samples. The used substances are described to have an efficacy of 92–98% against the parasites described in our study (Lloyd and Gemmell 1992; Prelezov and Bauer 2003; Mehlhorn et al. 2003). As every treatment was done under controlled conditions by the veterinarian, an inappropriate application or wrong dosage is very unlikely.

The feeding of fresh uncooked meat was not identified as a risk factor for infection with intestinal helminths, while the uncontrolled feeding of rodents, offal, or carrion was identified to be responsible for the appearance of hookworm and taeniid eggs. It is probable that especially small rodents serve as intermediate (*Taenia* sp. and *Echinococcus* sp.) or paratenic hosts (hookworms and *T. canis*). Twenty dogs shed parasites which must have originated from ingestion of foreign feces as they were not dog specific (mainly *Eimeria* spp. oocysts; data not shown). With the exception of one, all were defined as “uncontrolled feeder.” Coprophagia is an important factor that should not be neglected when performing and interpreting parasitological analyses. Several parasite stages that are only passing the intestinal tract may not be easily distinguished from dog-specific parasites (e.g., *Capillaria* spp. or taeniid eggs from cat feces). Furthermore, it may be possible that the prevalence of canine parasites is overestimated due to coprophagia of fox feces, which is abundant in rural as well as

urban areas (e.g., taeniid eggs, *Toxocara*, hookworms). This hypothesis is favored by the fact that coprophagia was identified as a significant risk factor for the presence of hookworm (FET $P=0.009$) and taeniid eggs in dog feces ($P=0.002$).

Several parasites detected in the dog feces are of zoonotic potential. Especially *T. canis* (Glickman and Shofer 1987) and *E. multilocularis* (Deplazes and Eckert 2001), the latter was identified in one dog in the prevalence study by antigen testing and PCR, can cause significant health problems in humans.

The present findings show that despite regular treatment and efficient drug components four times a year, only 39 of 111 dogs did not have any helminth stages in all fecal samples, and helminths may be present in almost two third of the dog population. Veterinarians should therefore inform dog owners that even after deworming, dogs may be a potential infection source for humans.

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