Intestinal Parasitic Infection in Multi-Cat Shelters in Catalonia

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ABSTRACT

The aim of this study was to characterize the prevalence of intestinal parasite infections in multi-cat shelters and to evaluate facility and the management-related risk factors. The study included adult-cats housed for long periods under two different multi-cat environments: rescue shelters where both dogs and cats were housed and shelters with only cats (cat shelters). A total of 423 fecal samples were collected from 11 rescue shelters and five cat shelters, and examined by coprological examination using a centrifugation-flotation technique with zinc-sulfate solution (ρ=1.18). The overall prevalence of intestinal parasites was 57% (242/423 fecal samples). The following parasites were detected in the study: *Giardia* spp. (116/423; 27%), *Toxocara cati* (71/423; 16.7%), *Cystoisospora* spp. (67/423; 16%), hookworms (36/423;8.4%), Taenidae (33/423; 7.8%), metastrongilids (20/423; 5%), *Toxascaris leonina* (10/423; 2.4%), *Dypilidium caninum* (4/423; 1%), *Capillaria* spp./*Eucoleus aerophilus* (2/423; 0.5%) and *Spirometra* sp. (1/423; 0.2%). *T. cati* and *Cystoisospora* spp. were the most prevalent parasites in rescue shelters, whereas prevalence of *Giardia* spp. was similar in both populations studied. Rescue shelters showed higher prevalence of intestinal parasites than cat shelters ($P < 0.05$). The facilities and management were similar in both shelter types, and therefore it is hypothesized that the vicinity to dogs in rescue shelters constitutes a stressful factor for cats, potentially predisposing them to a higher prevalence of intestinal parasites.

Keywords: *Giardia* spp.; *Toxocara* spp.; Rescue Shelters; Cat Shelters; Risk Factors; Stress; Feline.

INTRODUCTION

Animal shelters harboring multi-cat population include rescue shelters, in which free roaming or stray dogs and cats are kept, and cat shelters which house only cats. High population density in such shelters is likely to increase direct and indirect contact and subsequent animal stress (1). The continuous introduction of new (often juvenile) animals into such shelters, high population turnover and presence of newly introduced cats with long-term residents might favor frequent exposure to parasites and environmental contamination, resulting in higher risk infectation (2). Therefore, protozoal and helminthic intestinal parasites are among the main feline enteropathogens of such cats, and such infections are more prevalent in shelter cats than in owned-cats (3). Financial constraints and untrained staff in such rescue organizations, often result in inadequate hygiene and management and inappropriate deworming protocols (2).

Previous studies of stray cats and shelter cats showed intestinal parasite prevalence of 23.1% to 91% (4-7) and 26.8% to 100% (8-12), respectively, while in privately owned cats the prevalence is much lower (10.1% to 39.6%) (13-16), likely due to better overall health and management (7).

Several factors may impede the parasitic infection control and may contribute to re-infection, especially regarding the facility (e.g., floor material) and the general management (e.g., housing conditions, presence of a quarantine facility, implementation of adequate disinfection and deworming protocols (17).

Euthanasia is forbidden by the Animal Protection Law in Catalonia. Therefore sheltered cats await adoption or rehoming. Zoonotic diseases, such as *Toxocara cati* infection, recently regarded by the public and health authorities as a zoonosis (18), is a major concern. Toxocariasis might induce visceral and ocular larva migrans in humans accidentally ingesting embryonated infective eggs (19,20). Additionally, the risk of zoonotic transmission of *Giardia* spp. from cats to humans must be considered as well, because cats may be carriers of zoonotic assemblages (A and B) (7).

The aim of this study was to determine the prevalence of intestinal parasite infection in adult cats, housed long-term in multi-cat shelters, and to evaluate facility and management-associated risk factors for such infections in two different multi-cat shelters: rescue shelters where both dogs and cats are housed and cat shelters which held only cats.

MATERIAL AND METHODS

This study was conducted in several multi-cat shelters in Catalonia, Spain, from November 2012 to April 2015.

The inclusion criteria of the study cats included adult age (above one year of age), long-term (>6 months) stray and no antiparasitic treatment over the three months prior to sampling.

A questionnaire of information of the cat population (including feline immunodeficiency virus [FIV] and feline leukemia virus [FeLV] status), the facility (including deposition tray material such as sawdust, newsprint or cat litter, and the vicinity to dogs) and management (i.e., population size, available quarantine, disinfection protocol, including products used, frequency and deworming protocol, including products used and frequency, and tick and flea control) was filled-in at the time of sampling.

Fresh stool samples were collected from the deposition tray, transported to the Veterinary Faculty and stored at 4ºC pending analysis. Feces were macroscopically examined to detect adult stages or proglottids. Coprological examination was done using a previously described centrifugation-flotation technique (21). Concentration was carried out using zinc sulphate solution 33% (specific density 1.18 g/ml) flotation. Slides were examined at X100 and X400 magnifications using bright field microscopy.

Prevalence of infection and evaluation of eventual risk factors were analyzed in Epi-Info v. 6.0 (CDC, Atlanta, GA, USA) using Chi-square test. Significance was set at $P \leq 0.05$, unless otherwise indicated.

RESULTS

The study included 16 shelters, 11 rescue shelters harboring both dogs and cats and five cat shelters, where only cats were admitted. The study analyzed 423 fecal samples (rescue shelters, 263; cat shelters, 160). All cats were housed in communal housing in groups of five to 30 cats (n=215) and or 35 to 80 (n=208).

Prior to be admitted in the shelter, cats (n=405) were tested by commercial ELISA screening tests, with 387 negative for both FeLV and FIV, 10 positive for FIV or/and FeLV and the FIV/FeLV status being unknown in 8 cats.

The overall detected intestinal parasite prevalence was 57% (n=242). Infections were noted in all 16 facilities, with prevalence ranging from 19.4% to 94%. The type and frequency of enteric parasitic infection is shown in Table 1.

Rescue shelters showed higher intestinal parasite prevalence than cat shelters ($P < 0.001$). Statistical differences

Table 1: The observed occurrence of enteric parasites in rescue shelters and cat shelters from sixteen multi-cat centers.

Parasite species	Rescue shelter (n=263)	Cat shelter (n=160)	All cats (n=423)
Protozoa	108 (41%)	55 (34%)	163 (38.5%)
Giardia spp.	61 (23%)	55 (34%)	116 (27%)
Cystoisospora spp.	62 (23.6%)	5 (3%)	67 (16%)
Tapeworms	29 (11%)	8 (5%)	37 (8.8%)
Taenidae	29 (11%)	4 (2.5%)	33 (7.8%)
Dypilidium caninum	Ø	4 (2.5%)	4 (1%)
Spirometra spp.	1 (0.4%)	Ø	1 (0.2%)
Nematodes	89 (34%)	17 (10.6%)	196 (25%)
Toxocara cati	64 (24.3%)	7 (4.3%)	71 (16.7%)
Toxascaris leonina	10 (4%)	Ø	10 (2.4%)
Hookworms	29 (11%)	7 (4.3%)	36 (8.4%)
Metastrongilids	15 (6%)	5 (3%)	20 (5%)
Capillaria sp. / *Eucoleus* sp.	2 (1%)	Ø	2 (0.5%)
Total	171 (65%)	71 (44.4%)	242 (57%)

were observed when comparing helminthes prevalence and shelter, being higher in rescue shelters ($P \leq 0.001$;); whereas no statistical differences were observed with respect to the prevalence of protozoa ($P > 0.05$).

Single infection was detected in 66% of the positive samples. Coinfection with two and greater than 2 parasites was observed in 23.5% and in 11% of the cases, respectively. The most common coinfection was with *Giardia* spp. and *Cystoisospora* spp. (n=12). Coinfection was significantly more common in rescue shelters ($P = 0.001$).

The most frequently deposition tray material used was cat litter (n=312) followed by soil (n=51), sawdust (n=34) and pellet (n=26). The highest intestinal parasite prevalence was observed in samples collected in soil (76%), followed by sawdust (70.6%), cat litter (54%) and pellet (38.5%), and there was a significant difference in the prevalence of infection among the different tray materials ($P = 0.017$). Both *Giardia* spp. and *Cystoisospora* spp. were the most prevalent (35.3%) in samples collected in sawdust, while in those housed on cat litter, *Giardia* spp. and *T. cati* were the most prevalent (35.3% for both). *Giardia* spp. was the most prevalent parasite (23%) in cats housed on pellet.

Quarantine was applied in 8 of the 16 shelters (n=206, 48.7%). There was no association between the prevalence of parasites with presence of quarantine.

Cats were dewormed when introduced to the shelter and later periodically treated, four times a year (43%; n=154) or once or twice a year (57%; n=206). Deworming included milbemycin-praziquantel combination (158 cats; 37%), fenbendazole in (104 cats; 25%) and praziquantel-pyrantel-febantel combination in (19 cats; 4.5%). The deworming protocol was unavailable in 63 cats. There was no association between the prevalence of parasitic infection and the frequency of deworming ($P > 0.05$).

The cleaning and disinfection schedules were similar in all facilities. Feces were collected and removed daily, whereas tray material was completely replaced every two days or more, except just one center where it was done daily. Bleach (sodium hypochlorite) was the most commonly disinfectant compound used (127 cats; 44.7%) followed by steam water (94 cats; 33.1%), soap (39 cats; 13.7%) and a bleach-ammonia combination (24 cats; 8.5%). For seven facilities (139 cats) the disinfection data was unavailable. There was no association between the prevalence of infection with the disinfectant type used ($P > 0.05$).

Fecal consistency was scored in 288/423 samples. 85.3% of positive samples to protozoa showed a score of 5-6, presenting texture without definite shape, which left residue when picked up, statistical differences ($P = 0.02$) were observed between the prevalence of protozoa infection and fecal consistency.

DISCUSSION

The intestinal parasite overall prevalence in multi-cat environments herein was 57%, while previous studies showed prevalence of 7.19%, 26.6%, 33.6%, 50.9% and 100% (7-11). In the present study, rescue shelters showed higher intestinal parasite infection prevalence than cat shelters, although the management and facilities were very similar in both environments, with the exception of rescue shelters which harbor both dogs and cats, whereas cat shelters housing only cats. We therefore suggest that, in rescue shelters the vicinity to dogs induced visual and auditory stress for the cats, as previously reported (22).

Regarding parasites detected: *Cystoisospora* spp. prevalence was significantly higher in rescue shelters than in feline shelters (23.6% vs. 3%, respectively). These parasites are especially prevalent in kittens, and prevalence decreases with age (23). Such infection in older cats, might occur in immunocompromised animals, including stressed hosts (24, 25). Our results suggest the eventual role of stress in the prevalence of this coccidian parasite.

Similarly, *T. cati* was more prevalent in rescue shelters than in cat shelters. In most previous studies of multi-cat environments, *T. cati* was the most frequent parasite detected, with prevalence of 10.5% to 27.1% (7, 10, 11). However, it should be noted that differentiating between *Toxocara* spp. based on morphometric identification by light microscope is imperfect (26). Thus, in the rescue shelters it is impossible to determine the proportion of detected *Toxocara* spp. eggs that had originated from nearby dogs through environmental contamination and procedures and that proportion of true infection in cats. It has been reported that some cats repeatedly shed *T. canis* eggs in their feces after having been transferred to cages that had served for dogs (27). Nevertheless, both *T. canis* and *T. cati* are important zoonoses (19).

Giardia spp., was the most common intestinal parasite detected in both multi-cat shelters in this study. Presence of *Giardia* sp. is common in dogs, and to a lesser extent, in cats (28). It has been suggested that its prevalence is often

underestimated due to the low sensitivity of conventional detection methods, and because it may be present subclinically, and cysts are excreted intermittently. *Giardia* sp. infection is infrequently associated with clinical disease in dogs and cats (29), however, cases of clinical giardiasis in cats are often reported in multi-cat environments, where overcrowding may cause stress and exacerbate the clinical presentation (30).

Reports of the risk of infection that originates in dogs and cats for humans are conflicting (31-34). Dogs and cats may carry *Giardia* assemblages A and B, and therefore may act as infection sources to humans. A previous study of sheltered cats showed that 8/18 cats that were positive for *Giardia* spp. were infected with assemblages A (2 cats) and B (6 cats), although assemblages C and D (3 cats), mostly of canid origin, were also detected. In that study, genotype F was the most prevalent (7 cats). Nevertheless, inter-species *Giardia* spp. transmission between dogs and cats living in the same area or habitat is considered minimal (35).

Hookworm eggs were detected in 8.4%, and this was the second most prevalent nematode infection, while the prevalence in cats in previous studies was lower (1.1% to 4.9%) (9, 11, 12). Conversely, in another study from a Brazilian cat shelter, hookworms were markedly prevalent (96.1%) (10).

Tapeworms compatible with *Dypilidium caninum* were found in 8.8% of the stool samples in the present study. Egg-capsules were detected in samples from two cat shelters (1.5%). The present prevalence is similar to previous findings in cats (0.2% - 5%) (6, 7, 11 ,13, 16, 36). It is mainly transmitted by ingestion of *Ctenocephalides* fleas containing the larval stage. Data on ticks and fleas control was not reported for any of these cat shelters. *Spirometra* spp. eggs were found in fecal samples from one rescue shelter (0.2%), where raw fish was included in the cats' diet. Previous findings of this infection in cats in New York and Italy showed prevalence of 0.4% and 1.12%, respectively (11, 16).

In this study, the prevalence of lungworms was 8.8%, and mostly (8%) metastrongilidae (*Aelurostrongylus* sp. or *Troglostrongylus* sp.) while 1% of the samples were positive for the trichuroid *Eucoleus* spp. (syn *Capillaria* spp.). Studies conducted in shelters in Germany and New York reported that *A. abstrusus* was the main detected lungworm as well (1% and 6.2%, respectively) (11, 12). As in these two previous studies, in the present one the centrifugation-flotation technique was used, which allows detection of larvae; however, the Baerman sedimentation technique is more sensitive for detecting this parasite (12), and therefore, our results may be an underestimation of its true prevalence. The prevalence of *Eucoleus* spp. (syn *Capillaria* spp.) infection in cats is similar to previous findings in colony stray cats from Italy (1.4%) (6).

The rate of coinfection detected in the present study (34%) was similar to previous findings (27%) (12). Mixed infections are more common in cases of protozoal infections (*Giardia* spp. and *Cystoisospora* spp.) (37). The most frequent nematode coinfection presently was of hookworms and *Toxocara* spp., which is consistent with a previous study (10). Rescue shelter cats are thought to be more prone to harbor several parasites concurrently compared to cat shelter ones, as the vicinity to dogs may constitute a continuous stressful factor (22). Little is known about the consequences of harboring coinfections, but it has been suggested that the presence of several parasites concurrently induces some adverse synergistic effects concerning clinical signs (38, 39).

As in the present study, a previous study did not find a significant correlation between fecal consistency and intestinal parasite infection in multi-cat environments (10). Most positive samples were solid, consistent and of normal-shape. Asymptomatic cats, infected with intestinal parasites may play an important role as the main infection source for other cats. Nevertheless, diarrheic samples were reported to be more likely to be positive for both *Giardia* spp. and *Cystoisospora* spp. (7).

Deposition tray material might play an important role in maintaining some parasites (17). In that sense, a higher prevalence were detected in shelters using soil in the deposition tray, maybe because cats sometimes bury their feces in soil, which promotes a suitable environment where parasites develop to their infective stage, while protecting the parasite from desiccation, which constitutes one of the harmful mechanisms for parasites survival (40).

Regarding the facility management, this study showed no significant association between presence of quarantine in the facility and parasite prevalence. Quarantine is most important for controlling infectious diseases in such facilities (22). Quarantine was not applied in all shelters, and in those in which a quarantine was used, rigorous quarantine protocols were not adopted. There was no association of parasite prevalence with deworming frequency, in agreement with a previous study showing that dogs that were dewormed 1 to 4 times annually showed similar parasite prevalence to untreated animals (41).

Bleach (sodium hypochlorite) and steam water were the most common disinfectants used in the shelters studied. Sodium hypochlorite is cheap, readily-available, and has been shown to be ovicidal *in vitro* against *Toxocara* spp. and *Trichuris* spp. (42, 43). Conversely, other studies showed that *Toxocara* spp. eggs exposed to bleach did develop to the infective larvae despite being damaged (44). Steam treatment also provides an effective method for killing pathogens, and is probably less toxic for animals than chemical disinfectants (45). Quaternary ammonium compounds (as a single disinfectant or combined with other ones) has been shown to be effective at inactivating *Giardia* cysts (46). Although several disinfecting strategies exist, there is no uniformly efficacious and recommended protocol against parasitic infections in shelters (47).

In conclusion, the most prevalent intestinal parasites observed in multi-cat shelters have zoonotic potential. Mixed dog-cat rescue shelters showed higher prevalence of intestinal parasites than cat shelters. Because the facilities and their management factors were very similar for both the mixed rescue shelters and the cat shelters, the vicinity of dogs may constitute a stressful factor for cats, which play a possible role in the observed higher prevalence of intestinal parasites in the former shelters. Further studies are warranted to clarify the role of stress in the prevalence of intestinal parasites in cats in multi-cat environments.

CONFLICT OF INTEREST STATEMENT

The authors declare no conflicts of interest regarding the content of this paper.

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