GASTROINTESTINAL PARASITES OF DOGS AND CATS IN A REFUGE IN NAKHON NAYOK, THAILAND

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Abstract. We collected fecal samples from 500 dogs and 300 cats from an animal refuge in Nakhon Nayok Province, Thailand to test for gastrointestinal protozoa and helminths using a formalin-ether concentration technique. The overall prevalence of parasites in stool from dogs was 36.2% (181/500), 35.7% (177/500) had helminths and 2.8% (14/500) had protozoa. The helminths were: hookworm (30.6%), Trichuris vulpis (16.0%), Toxocara canis (6.6%), Hymenolepis diminuta (1.2%), *Spirometra mansoni* (0.6%), and *Dipylidium caninum* (0.2%). *Giardia duodenalis* (2.8%) was found in the stool of dogs. The overall prevalence of parasites in stool from cats was 44.3% (133/300), 43.3% (130/300) were helminths and 6.0% (18/300) were protozoa. The helminths were hookworm (34.7%), T. cati (9.7%), S. mansoni (4.0%), *Platynosomum fastosum* (2.7%)*, Strongyloides* sp (0.7%)*, and Echinostoma* sp (0.3%)*.* Two species of protozoa, Isospora sp (5.7%) and G. duodenalis (0.3%) were found in the stool of cats. Two percent of dogs and 5.0% of cats had mixed protozoan and helminthic infections. Dogs with double, triple, and quadruple helminthic infections were found at rates of 22.0%, 2.8%, and 0.2%, respectively. Cats with double and triple helminthic infections were found at rates of 9.7% and 1.0%, respectively. Quadruple helminthic infections were not found in cats, and double protozoan infections were not found in either dogs or cats.

Keywords: gastrointestinal parasites, dogs, cats, animal refuge, Thailand

INTRODUCTION

Companion animals, particularly dogs and cats, act as reservoir hosts for several species of gastrointestinal protozoa and helminths that can spread to humans. These parasites include *Toxocara* canis, T. cati, Ancylostoma caninum, A. ceylanicum, A. braziliense, Trichuris vulpis, Strongyloides sp, Dipylidium caninum, Echinococcus granulosus, Taenia taeniaeformis, Spirometra mansoni, Giardia duodenalis, Cryptosporidium sp, Toxoplasma gondii, Sarcocystis sp, Entamoeba histolytica, Balantidium coli, Blastocystis sp (Holyoake, 2008; Palmer et al, 2008), Opisthorchis viverrini (Hinz 1980; Impand et al, 1983; Enes et al, 2010), Gnathostoma spinigerum (Daengsvang, 1980; Maleewong et al, 1992; Rojekittikhun et al, 1996, 2000), Schistosoma

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mekongi (Iijima et al, 1971; Matsumoto et al, 2002) and Ascaris lumbricoides (Traub et al, 2002; Shalaby et al, 2010). There are estimated to be over 9 million dogs and cats in Thailand (Bureau of Disease Control and Veterinary Services, 2008). Thailand is rated 10th in the list of countries with the largest pet dog populations at 6.9 million (Lodish, 2013; Maps of World, 2013). A Thai study of 1.94 million households found 20% raised dogs, of which 51.2% had 1 dog, 2.9% had 5 dogs and 0.6% had more than 10 dogs (National Statistical Office Thailand, 2006). However, in Thailand 5,000 dogs are abandoned every year; in Bangkok alone there are 0.82 million dogs kept as pets (Naewna, 2013).

In Thailand, the number of stray dogs and cats increases each year. The ratios of not-owned and owned dogs and cats are 1:7.6 and 1:4.4, respectively (11.6% of dogs and 18.6% of cats are not owned) (Bureau of Disease Control and Veterinary Services, 2008). Animal euthanasia is not acceptable to most Thai people, resulting in the establishment of several kinds of animal homes, refuges, pounds, guardian associations and foundations by the non-government sector. These include private/personal refuges run by dog and cat lovers, and although kind-hearted, these individuals are generally elderly citizens. These shelters rely completely on donations and sponsorship and are often unorganized operations. These combined issues may cause management and animal-healthcare problems.

Although dogs and cats are potential reservoirs of zoonotic infections to humans, little data exists on the nature, prevalence, and intensity of animal infections. Most studies were conducted in Bangkok, with few studies from other provinces (Sangvaranond, 2003; Kaewthamasorn *et al*, 2006; Inpankaew *et al*, 2007; Jittapalapong *et al*, 2007). The purpose of this study was to determine the prevalence and species of gastrointestinal protozoan and helminthic infections among dogs and cats at one refuge in Nakhon Nayok Province, Thailand. The study may indirectly reflect on gastrointestinal parasitic infections among abandoned pets.

MATERIALS AND METHODS

Sample collection

Five hundred dog and 300 cat stool samples were collected from Aunt Samruay's refuge in Nakhon Nayok Province, about 100 km northeast of Bangkok (Fig 1). The refuge is one of the largest homes for abandoned and stray dogs and cats in Thailand, accommodating about 1,000 dogs and 500 cats. The sample sizes used in the study were higher than the minimum needed sample sizes for these populations of dogs and cats (Krejcie and Morgan, 1970) (minimum calculated samples: 278 dogs and 217 cats).

Fecal examination

All the samples were examined for gastrointestinal protozoa (trophozoites, cysts and oocysts) and helminths (eggs and larvae) by formalin-ether concentration (Suzuki, 1981). Briefly, 1 g of feces was mixed with 10 ml of water and strained through 2 layers of wet gauze into a centrifuge tube. After centrifugation at 1,000g for two minutes. 7 ml of 10% formalin and 3 ml of ether were added to the sediment. The mixture was shaken vigorously for ten minutes and centrifuged again. The recovered sediment was then examined under a light microscope. Statistical comparisons were performed using chi-square and Fisher's exact tests.

Parasite species	No. positive (% positive)		<i>p</i> -value
	Dogs (N = 500)	Cats (N = 300)	<i>p</i> -value
Protozoa	14 (2.8)	18 (6.0) ^a	0.025 ^c
Giardia duodenalis	14 (2.8)	1 (0.3) ^a	0.013 ^c
Isospora sp	0	17 (5.7) ^b	< 0.0001 ^d
Helminths	177 (35.7)	130 (43.3) ^a	0.025 ^c
Hookworm	153 (30.6)	104 (34.7) ^a	0.233 ^e
Trichuris vulpis	80 (16.0)	-	-
<i>Toxocara canis (T. cati</i> in cats)	33 (6.6)	29 (9.7) ^a	0.116 ^e
Hymenolepis diminuta	6 (1.2)	0^{b}	0.057^{e}
Spirometra mansoni	3 (0.6)	12 (4.0) ^a	0.001 ^d
Dipylidium caninum	1 (0.2)	0^{b}	0.438 ^e
Platynosomum fastosum	-	8 (2.7)	-
Strongyloides sp	0	2 (0.7) ^b	0.068 ^e
Echinostoma sp	0	1 (0.3) ^b	0.196 ^e
Overall parasitic infections	181 (36.2)	133 (44.3) ^a	0.023 ^c

Table 1 Numbers and percentages of fecal dog and cat samples positive for protozoa and helminths.

^aChi-square test, ^bFisher's exact test; ^csignificant difference, ^dhighly significant difference, ^eno significant difference.

RESULTS

The overall prevalence of gastrointestinal parasitic infections in dogs was 36.2% (181/500), of which 35.7% (177/500) were helminthic infections, and 2.8% (14/500) were protozoan infections (Table 1). The helminthic infections were: hookworm (30.6%), Trichuris vulpis (16.0%), Toxocara canis (6.6%), Hymenolepis diminuta (1.2%), Spirometra mansoni (0.6%) and Dipylidium caninum (0.2%) (Fig 2). Only one species of protozoa, Giardia duodenalis, was found in dog feces (2.8%). The overall prevalence of gastrointestinal parasitic infections in cats was 44.3% (133/300); 43.3% (130/300) had helminthes and 6.0% (18/300) had protozoa. The helminths were: hookworm (34.7%), T. cati (9.7%), S. mansoni (4.0%), Platynosomum fastosum (2.7%), Strongyloides sp (0.7%) and Echinostoma sp

(0.3%). Two species of protozoa, *Isospora* sp (5.7%) (Fig 2) and *G. duodenalis* (0.3%), were found in cat feces (Table 1). Mixed protozoan and helminthic infections were found in 2.0% of dogs and 5.0% of cats (Table 2). Double, triple and quadruple infections were found in 22.0%, 2.8%, and 0.2% of dogs, respectively. Double and triple helminthic infections were found in 9.7% and 1.0% of cats, respectively. No quadruple helminthic infections were found in cats. Double protozoan infections were not found in either dogs or cats.

Comparisons of the prevalences of some parasite infections in dogs and cats are shown in Tables 1 and 2. *G. duodenalis* infection was seen in dogs significantly more often than in cats (p<0.05). *Isospora* sp was not found in dogs but were found in 5.7% of cats (p<0.0001). For helminths, only *S. mansoni* was significantly more

Parasite species	No. positive (% positive)		<i>p</i> -value
	Dogs ($N = 500$)	Cats (N = 300)	<i>p</i> -value
Multiple protozoan infections	0	0	-
Multiple helminthic infections	125 (25.0)	32 (10.7) ^a	< 0.0001 ^d
Double helminthic infections	110 (22.0)	29 (9.7) ^a	< 0.0001 ^d
HW + Tv	64 (12.8)	-	-
HW + Tcn (HW + Tct in cats)	25 (5.0)	20 (6.7) ^a	0.332 ^e
Tv + Tcn	21 (4.2)	-	-
HW + Sm	0	4 (1.3) ^b	0.010 ^c
HW + Pf	-	1 (0.3)	-
Tct + Sm	-	3 (1.0)	-
Sm + Ss	0	1 (0.3) ^b	0.196 ^e
Triple helminthic infections	14 (2.8)	3 (1.0) ^a	$0.087^{\rm e}$
HW + Tv + Tcn	13 (2.6)	-	-
HW + Tv + Hd	1 (0.2)	-	-
HW + Tct + Sm	-	3 (1.0)	-
Quadruple helminthic infections	1 (0.2)	0^{b}	0.196 ^e
HW + Tv + Tcn + Hd	1 (0.2)	-	-
Mixed protozoan and helminthic infections	10 (2.0)	15 (5.0) ^a	0.018 ^c
Gd + HW	4 (0.8)	1 (0.3) ^b	0.656^{e}
Gd + HW + Tcn	2 (0.4)	-	-
Gd + HW + Tv	1 (0.2)	-	-
Gd + HW + Tv + Tcn	3 (0.6)	-	-
Is + HW	0	8 (2.7) ^b	<0.0001 ^d
Is + Sm	0	3 (1.0) ^b	0.025 ^c
Is + Tct	-	1 (0.3)	-
Is + HW + Tct	-	1 (0.3)	-
Is + HW + Tct + Sm	-	1 (0.3)	-

Table 2 Numbers and percentages of fecal dog and cat samples positive for protozoa and helminths by type of infection.

Gd, Giardia duodenalis; Is, Isospora sp; Hd, Hymenolepis diminuta; HW, Hookworm; Pf, Platynosomum fastosum; Sm, Spirometra mansoni; Ss, Strongyloides rhabditiform larva; Tcn, Toxocara canis; Tct, Toxocara cati; Tv, Trichuris vulpis; ^achi-square test; ^bFisher's exact test; ^csignificant difference, ^dhighly significant difference.

common in cats than dogs (p<0.001). Multiple helminthic infections were more common in dogs than cats (p<0.0001).

DISCUSSION

Previous reports from Thailand focused mainly on Bangkok metropolitan areas and found the most common protozoa and helminthic infections in dogs were: *Ancylostoma* sp, *T. canis, T. vulpis, S. mansoni, G. duodenalis* and *Isospora* sp (Hinz, 1980; Rojekittikhun *et al*, 1998, 2000; Sangvaranond, 2003; Kaewthamasorn *et al*, 2006; Inpankaew *et al*, 2007; Traub *et al*, 2008, 2009); and in cats were *Ancylostoma* GI Parasites of Dogs and Cats in Nakhon Nayok



Fig 1–Aunt Samruay's refuge.

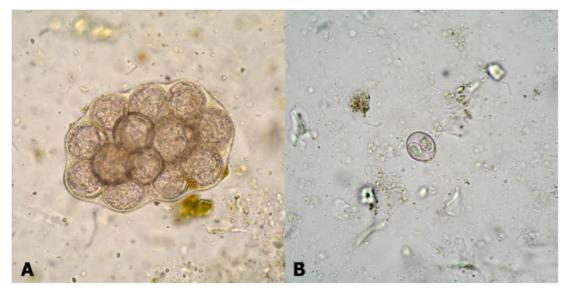


Fig 2–(A) Egg packet of *Dipylidium caninum* (each egg is approximately 35-60 m in size); (B) *Isospora* sp oocyst (38-51 x 27-39 m) found in feces of dogs and cats.

sp, T. cati and Isospora sp (Impand et al, 1983; Jittapalapong et al, 2007; Enes et al, 2010). Our study found similar results except where one cat was found to be infected with Echinostoma sp. Flukes in the genus Echinostoma are found worldwide, since many animals serve as definitive hosts, including aquatic birds, carnivores, and rodents. Ten species have been recorded to infect humans, including: E. malayanum, E. hortense, E. macrorchis, E. revolutum, E. ilocanum and E. perfoliatum (CDC, 2009). E. revolutum, E. ilocanum, E. malayanum and E. hortense have been reported to occur in cats and dogs (Sohn and Chai, 2005; Chai et al, 2009; Shin et al, 2009; Vetbook, 2010; Okanishi et al, 2013).

At Aunt Samruay's refuge, dogs and cats are confined to a large area on the grounds of the temple, Wat Prongphai Chantharangsi. The animals are caged individually, housed together in groups by size, or kept in large, open rooms. In total, there are about 1,500 animals, and although not overcrowded, they are housed rather closely.

The prevalence of *G. duodenalis* was significantly higher among dogs than cats. This may be explained by the fact that *G. duodenalis* (synonym: *G. intestinalis* or *G. lamblia*) is considered to be a complex species that can infect people, dogs and cats. *Giardia* species are divided into assemblages. Assemblage A infects humans and other mammals, including dogs and cats. Assemblage B infects humans and dogs but not cats. Assemblages C and D *Giardia* commonly infect dogs and Assemblage F infects cats (Monis *et al*, 2009; Tangtrongsup and Scorza, 2010).

Isospora sp infections occurred in 5.7% of cats but not in dogs. A reason for this may be a host specificity of the coccidian. Canine *Isospora* do not infect felines

and the reverse is true for feline *Isospora* (Dubey *et al*, 2009; CAPC, 2013). Further studies are needed to idenfity the specific serotypes.

Hookworm parasites were the most common helminth in both dogs (30.6%) and cats (34.7%). This is consistent with previous studies conducted in 2001-2002, of stray dogs and cats in Bangkok temples, where the prevalences of hookworm infections were 70.3-88.3% in dogs and 55.4-76.6% in cats (Rojekittikhun, unpublished data). Similarly, a high prevalence was also observed in studies from the Bangkok area (19.9-58.1% in dogs, 9.9-68.9% in cats) (Sangvaranond, 2003; Kaewthamasorn et al, 2006; Inpankaew et al, 2007; Jittapalapong et al, 2007); from other provinces in Thailand (58.8-73.0% in dogs, 32.1-77.3% in cats) (Impand et al, 1983; Rojekittikhun et al, 2000; Enes et al, 2010); and from other countries (53.8-87.8% in dogs, 37.9-94.2% in cats) (Mukaratirwa and Singh, 2010; Coelho *et al*, 2011; Ngui *et al*, 2012).

Nakhon Nayok Province in Thailand is endemic for gnathostomiasis. The prevalences of *G. spinigerum* in swamp eels was 10.7-44.1% (Rojekittikhun *et al*, 2004a,b), among dogs was 1.2-2.9% and among cats was 1.9-2.9% (Rojekittikhun *et al*, 1996, 2000, 2002). No dogs or cats in this study were positive for *G. spinigerum*. This may be due to: 1) seasonal fluctuation in infection and worm burden of the parasite (Maleewong *et al*, 1992); 2) a low prevalence in canine and feline definitive hosts (Rojekittikhun *et al*, 1996, 2000); and 3) many refuge dogs and cats are not native to the province.

In conclusion, dogs and cats were found to harbor several species of parasites, posing a significant zoonotic risk to humans. Healthy animals usually mean animal keepers are providing good healthcare. This can only be possible when pet owners take good care of their animals and do not abandon them. It is necessary to educate the public, veterinarians, and funding agencies about the work of private refuges. People may contribute by adopting a pet, donating to a shelter, sponsoring animals or supporting compassionate people who care for animals. These are good ways to keep these animals healthy.

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