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Short communication

Cryptosporidium, *Giardia* and *Enterocytozoon bieneusi* in cats from Bogota (Colombia) and genotyping of isolates

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Abstract

The prevalence of *Cryptosporidium*, *Giardia*, and *Enterocytozoon bieneusi* in cats from Bogota (Colombia) was determined from fecal specimens and scrapings of duodenal and ileal mucosa screened by PCR. All PCR-positive specimens were sequenced to determine the genotype(s) present. Of 46 cats, 6 (13%) were positive for *Cryptosporidium*, 5 (11%) were infected with *C. felis* and one (2%) with *C. muris*. Three (6.5%) cats were infected with *Giardia duodenalis* Assemblage F. Eight (17%) cats were infected with four genotypes of *E. bieneusi*: genotype D-like (9%), K (4%), Peru 10 (2%), and Peru 5 (2%). This is the first report on the presence of zoonotic species/genotypes of *Cryptosporidium* and *E. bieneusi* in cats in Colombia.

Keywords: Cats; *Cryptosporidium*; *Enterocytozoon bieneusi*; Genotypes; *Giardia*; PCR

1. Introduction

Zoonotic transmission of *Cryptosporidium*, *Giardia* and *Enterocytozoon bieneusi* from domestic animals has been inferred following reports of humans becoming ill after exposure to infected animals (Morgan et al., 2000; Traub et al., 2004; Didier, 2005).

Cryptosporidium has been reported in a wide variety of vertebrate hosts (Fayer et al., 2000a; Xiao et al., 2004). Humans are infected primarily with two species, *C. hominis* that infects humans almost exclusively, and *C. parvum*, a zoonotic species with numerous hosts. To a lesser extent other species and genotypes of *Cryptosporidium* have been identified in infections of immunocompetent and immunocompromised humans. These include *C. canis*, *C. felis*, *C. meleagridis*, *C. muris*, and *C. suis*, as well as cervine and monkey genotypes (Pieniazek et al., 1999; McLauchlin et al., 2000; Morgan et al., 2000; Guyot et al., 2001; Pedraza-Diaz et al., 2001; Xiao et al., 2001; Caccio et al., 2002; Ong et al., 2002; Gatei et al., 2003). In cats two species of *Cryptosporidium* have been reported, *C. parvum* and *C. felis* (Sargent et al., 1998). Although both species are zoonotic (Xiao et al., 2004), *C. felis* has a much narrower host range primarily infecting cats (Fayer et al., 2006).

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Giardia duodenalis isolates from different host species are morphologically indistinguishable from each other and have been grouped into Assemblages (genotypes) based on molecular characteristics (Monis et al., 2003). *Giardia* isolates from humans have been identified as Assemblages A and B (Caccio et al., 2005). These Assemblages also have been found in feces of wild and domestic mammals, such as beavers, cattle, dogs and cats among others (Sulaiman et al., 2003a; Trout et al., 2004). Assemblages C, D, E, F, and G have been identified only in animal feces and are considered host-specific and non-zoonotic. Assemblages C and D have been identified only from canids, Assemblage E only from sheep, goats, cattle, and pigs, and Assemblages F and G only from cats and rats, respectively (Monis et al., 1999, 2003).

Microsporidia are emerging pathogens of animals and humans. Since the onset of the AIDS pandemic, enteric microsporidia, primarily *E. bieneusi*, have been reported in association with chronic diarrhea in severely immunocompromised patients (Weber et al., 1994; van Gool et al., 1995; Didier, 2005). *E. bieneusi* also has been found to be prevalent in wild, farm, and companion animals (Rinder et al., 2000; Fayer et al., 2003; Sulaiman et al., 2003b). The sources and routes of transmission of microsporidia infecting humans have not been identified directly. However, there is evidence suggesting zoonotic transmission (Dengjel et al., 2001; Buckholt et al., 2002). Recent genetic studies have demonstrated the presence of multiple genotypes, some host-specific and others zoonotic because they have been found in infections of both humans and animals (Rinder et al., 2000; Buckholt et al., 2002; Sulaiman et al., 2003b; Santín et al., 2005).

The prevalence and zoonotic potential of different genotypes of *Cryptosporidium*, *Giardia*, *E. bieneusi* in cats is poorly known. Most surveys of feline gastrointestinal parasites have been limited to microscopy to identify the parasite genus but identification of protozoa below that taxonomic level is problematic or impossible because most species or subspecies are morphologically indistinguishable from one another (Hill et al., 2000; Spain et al., 2001). To identify these and conduct epidemiological studies, molecular techniques are required. The current study was designed to determine the presence of species and genotypes of *Cryptosporidium*, *Giardia*, and *E. bieneusi* in cats from Bogota (Colombia) utilizing such techniques.

2. Materials and methods

2.1. Sources and collection of specimens

Entire intestinal tracts from 46 humanely euthanized unwanted domestic cats (30 females and 16 males, August, 2005) in Bogota (area of Engativa), Colombia, South America were carefully removed and placed in plastic bags that were labeled, sealed, cooled and shipped overnight to the USDA laboratory in the United States. These cats were unwanted or stray and were euthanized by intravenous injection of (Euthanex, Invet, S.A. Bogota, Colombia) by Centro Distrital de Zoonosis when efforts to place them in good homes failed. Cats ranged from 15 days to 10 years of age and were grouped as follows: <6 months ($n = 13$), 6–12 months ($n = 6$), and >2 years ($n = 27$) (Table 1). A fecal sample was collected from the rectum of each cat and placed in a 50-mL centrifuge tube. Feces were cleaned of fecal debris, concentrated, and then DNA was extracted (see below). Duodenal and ileal scrapings were obtained from each animal and placed in a 1.5 mL microfuge tube containing 180 μL of DNA extraction buffer (ATL).

2.2. Recovery of parasites from feces

Oocysts were concentrated from feces as previously described (Fayer et al., 2000b). Briefly, 15 g of feces from

Table 1
Number of cats examined, number of cats positive, and prevalence (%) of *Cryptosporidium*, *E. bieneusi*, and *Giardia* in different ages and sexes

	No. of cats	No. of positives (%)		
		Cryptosporidium	*E. bieneusi*	*Giardia*
Age groups				
<6 months	13	1 (7.7%)	2 (15.4%)	0
6–12 months	6	2 (33.3%)	2 (33.3%)	0
>2 years	27	3 (6.5%)	4 (14.8%)	3 (11.1%)
Sex				
Males	16	1 (6.2%)	5 (31.2%)	1 (6.25%)
Females	30	5 (16.6%)	3 (10%)	2 (6.6%)
Total	46	6 (13%)	8 (17.4%)	3 (6.5%)