



The First Report on Parasites in Cats from Chapala, Mexico

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Abstract

Background: The increasing number of companion animals in urban areas, primarily cats and dogs, poses serious public health, veterinary, and socioeconomic issues worldwide. The control of parasitic zoonoses is of utmost importance because, despite mass drug administration and preventive measures, they have not been successfully eradicated.

Objectives: This research was conducted to determine the prevalence of parasites in cats from Chapala, Mexico.

Methods: Analyzing a total of 113 fecal samples from both domestic and feral cats. The samples were analyzed by the Faust flotation technique and the Kinyoun specific technique. The fecal samples were carefully examined under an optical microscope at magnifications of 40x and 100x, systematically scanning each field of view across the entire slide. A sample was classified as positive if at least one cyst or trophozoite was observed. Any parasitic stage was identified based on its previously described morphological characteristics. The variables were categorical, and therefore, non-parametric tests were used.

Results: A total of 113 cat fecal samples were analyzed, and an overall prevalence of gastrointestinal parasites of 52.21% was calculated (59 positive and 54 negative). Of the positive cats, 27.11% had two species of parasites in the following order: *Dipylidium caninum*, *Ancylostoma*, *Taenia*, *Cystoisospora*, *Giardia*, *Toxocara*, and *Hammondia*. Additionally, 70.80% of the cats had fleas.

Conclusion: Intestinal parasitic infections continue to be prevalent in companion animals, despite the availability of highly effective drugs and control measures taken by owners and veterinarians. The prevalence of parasites in domestic and feral cats in the municipality of Chapala is high, highlighting the necessity to enhance their control.

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Introduction

Paleoparasitology studies the parasites of ancient human settlements. Most intestinal helminth eggs can be preserved for thousands of years under suitable conditions. Mummification has allowed the identification of previously unidentified endoparasites in skeletal burials, some of which are zoonotic, such as those causing dracunculiasis, filariasis, leishmaniasis, malaria, toxoplasmosis, and trichinosis [1].

Currently, these and other parasites continue to cause morbidity and mortality, and the increasing number of companion animals in urban areas, primarily cats and dogs, poses serious public health, veterinary, and socioeconomic issues worldwide [2]. The control of parasitic zoonoses is of utmost importance because, despite mass drug administration and preventive measures, they have not been successfully eradicated. This may be related to the resistance shown by certain parasites. To control them, there are programs often based on active monitoring, their distribution, and the specific identification of risk factors [3,4].

Animals that roam freely are a significant risk factor in the transmission of diseases. In the case of cats, they have close contact with humans, much like dogs. Cats are definitive hosts for a wide range of zoonotic parasites responsible for toxocariasis and ancilostomatidosis, toxoplasmosis, and giardiasis. Additionally, they serve as reservoirs for other infectious agents such as *Bartonella* spp. and *Rickettsia* spp [5]. Conducting constant studies has a significant impact on public health as they help define potential transmission risks, parasitological control, and infection prevention [6]. This research was conducted to determine the prevalence of parasites in cats from Chapala, México.

Materials and Methods

The objective of this study was to determine the prevalence of parasitic infections and associated risk factors in cats from the municipality of Chapala in the state of Jalisco, Mexico. A cross-sectional study was conducted, analyzing a total of 113 fecal samples from both domestic and feral cats during the months of June to December 2019. Samples were collected without regard to breed, age, gender, or general health status. The overall prevalence of gastroenteric parasites in cats, both owned and unowned, was calculated using the results of the Faust flotation technique and the Kinyoun specific technique.

All cat owners who agreed to participate were provided with informed consent, which explained the study's purpose. Additionally, an epidemiological survey was conducted to collect data on risk factors associated with enteric parasitic infections.

The samples were analyzed using the modified acid-alcohol resistant staining technique known as Kinyoun staining (1860-1919), also referred to as the modified Ziehl-Neelsen stain. This staining method is widely used due to its simplicity, speed, cost-effectiveness, and sensitivity, which can range from 37% to 90% [7].

For the detection of *Cryptosporidium* spp. oocysts in fecal samples [8], the technique involved creating a thin smear of feces on a glass slide, applying Kinyoun's carbol fuchsin for 15 minutes, briefly rinsing it (3 to 5 seconds) with 50% ethanol, immediately rinsing it with tap water [4], immersing it in 1% acid-methanol for 10-15 seconds, rinsing it again, and applying 0.4% malachite green for 30 seconds. The slide was then air-dried, and the specimen was observed

under a microscope using a 40x-100x objective lens [8].

The fecal samples were carefully examined under an optical microscope at magnifications of 40x and 100x, systematically scanning each field of view across the entire slide. A sample was classified as positive if at least one cyst or trophozoite was observed. Any parasitic stage was identified based on its previously described morphological characteristics [3].

Szwabe et al. [3] concluded that the Kinyoun staining technique has high sensitivity and specificity in detecting microsporidial spores in fecal samples, recommending its use in the diagnosis of intestinal microsporidiosis.

The data obtained in this study were recorded in an Excel database, and the verification of the analyzed samples was performed through electronic photographs, which were subsequently evaluated by the researcher.

The variables were categorical, and therefore, non-parametric tests were used. The Chi-square test was applied to determine the association between the variables, and Odds Ratios (OR) were calculated with $OR \geq 1$ to determine the risk factor for the presence of Gastrointestinal parasites (GI) in feline feces, with a significance level (alpha) of $P \leq 0.05$. For the analysis, statistical software JMP 8.0 was utilized.

Results

A total of 113 cat fecal samples were analyzed, and an overall prevalence of gastrointestinal parasites of 52.21% was calculated (59 positive and 54 negative). Of the positive cats, 27.11% had two species of parasites in the following order: *Dipylidium caninum*, *Ancylostoma*, *Taenia*, *Cystoisospora*, *Giardia*, *Toxocara*, and *Hammondia*. Additionally, 70.80% of the cats had fleas (**Table 1**).

Cats under 6 months of age were associated ($\text{Chi}^2 = 6.49$, $P = 0.03$) with the prevalence of gastrointestinal parasites, and it was established as a risk factor ($OR = 3.34$, $P = 0.0001$). On the other hand, gender did not show an association, but females exhibited a tendency (**Table 2**).

Table 1: Prevalence by Parasite in Jalisco's Cats.

GI Parasites	Number of Positive Cats	Prevalence (%)
<i>Ancylostoma</i>	10	8.85
<i>Cystoisospora</i>	5	4.42
<i>Dipylidium</i>	27	23.90
<i>Giardia</i>	4	3.54
<i>Toxocara</i>	4	3.54
<i>Taenia</i>	8	7.08
<i>Hammondia</i>	1	0.88
Positives	59	52.21
Negatives	54	47.79
Total	113	
Fleas		
Positives	80	70.80
Negatives	33	29.20
Total	113	

Table 2: Association of GI Parasite Prevalence with Age and Gender of Cats and Risk Factor.

	Positives n= 59	%	Negatives n= 54	%	Chi ²	P	OR	P	CI
Age									
≤ 6 months	20	17.70	9	7.96	6.49	0.03	3.34	0.0001	2.18-5.10
7 to 12 months	20	17.70	16	14.16					
≥ 13months	19	16.81	29	25.66					
Gender									
Female	31	27.43	38	33.63	3.76	0.052	2.14	0.052	0.98-4.66
Male	28	24.78	16	14.16					

Chi²= Chi- square, OR= odds ratio, • 95% CI = 95% Confidence Interval.

Table 3: Habits and Their Association with the Presence of GI Parasites and Risk Factors in Cats.

	Positives n= 59	%	Negatives n= 54	%	Chi ²	P	OR	P	CI
Lives with other cats									
Yes	57	50.44	45	39.82	5.65	0.017	5.70	0.017	1.17-27.70
No	2	1.77	9	7.96					
Lives with other animals									
Yes	56	49.56	45	39.82	3.98	0.045	3.73	0.045	0.95-14.60
No	3	2.65	9	7.96					
Out door Access									
Yes	53	46.90	40	35.40	4.80	0.028	3.09	0.028	1.09-8.75
No	6	5.31	14	12.39					
Brushing									
Weekly	1	0.88	7	6.19					
Monthly	5	4.42	10	8.85					
Never	53	46.90	37	32.74	8.80	0.012	4.05	0.007	1.46-11.26

Chi²= Chi- square, OR= odds ratio, • 95% CI = 95% Confidence Interval.

Table 4: Associated variables with the Presence of GI Parasites and Risk Factors in Cats.

	Positives n= 59	%	Negatives n= 54	%	Chi ²	P	OR	P	CI
Provenance									
Adopted	23	20.35	35	30.97					
Feral	36	31.86	19	16.81	7.53	0.006	2.88	0.006	1.34-6.19
Hair type									
Long	7	6.19	22	19.47	12.32	0.0004	0.19	0.0008	0.75-0.51
Short	52	46.02	32	28.32					

Chi²= Chi- square, OR= odds ratio, • 95% CI = 95% Confidence Interval.

Table 5: Characteristics of Feces and Their Association with GI Parasite Prevalence and Risk Factor.

Characteristic	Positives n= 59	%	Negatives n= 54	%	Chi ²	P	OR	P	CI
Color									
Yellow	3	2.56	2	1.77	0.60	0.50	1.39	0.72	0.22-8.67
Light Brown	33	29.20	34	30.09					
Brown	23	20.35	18	15.93					
Consistency									
Watery	1	0.88	1	0.88	6.89	0.075	1.44	0.79	0.086-24.12
Hard and Dry	5	4.42	7	6.19					
Firm	20	17.70	29	25.66					

Findings									
Mucus	5	4.42	0	0					
Parasites	7	6.19	1	0.88					
Blood	2	1.77	2	1.77	9.67	0.02	0.91	0.92	0.12-6.71
No Findings	45	39.82	51	45.13					

Chi²= Chi- square, OR= odds ratio, • 95% CI = 95% Confidence Interval.

Table 6: Association of Flea Prevalence with GI Parasites and Risk Factor.

	Flea Positives n= 80	%	Flea Negatives n= 33	%	Chi ²	P	OR	P	CI
Parasite									
<i>Ancylostoma</i>	9	7.96	1	0.88					
<i>Dipylidium caninum</i>	26	23.01	1	0.88	19.46	0.006	15.40	0.008	1.99-119.04
<i>Giardia</i>	2	1.77	2	1.77					
<i>Hammondia</i>	1	0.88	0	0					
<i>Cystoisospora</i>	4	3.54	1	0.88					
<i>Taenia</i>	6	5.31	2	1.77					
<i>Toxocara</i>	3	2.65	1	0.88					
Negative	29	25.66	25	22.12					

Chi²= Chi- square, OR= odds ratio, • 95% CI = 95% Confidence Interval.

The association with the prevalence of GI parasites and the risk factor was analyzed in **Table 3**. The results regarding prevalence and age showed that cats up to 6 months of age had an association with the prevalence of GI parasites and were a risk factor (Chi²= 5.65, P= 0.017, OR= 5.70, P=0.017). Feral cats were associated with the presence of parasite eggs in feces and were a risk factor for parasitosis (Chi²= 7.53, P= 0.006, OR= 2.88, P= 0.006). Likewise, long-haired cats were associated with the prevalence of GI parasites, and having long hair was a risk factor (Chi²= 12.32, P= 0.0004, OR= 0.19, P= 0.0008) (**Table 4**).

Regarding the characteristics of the fecal samples, watery consistency and the presence of blood were associated with the presence of parasite eggs in feces (Chi²= 6.89, P= 0.075, and Chi²= 9.67, P= 0.02, respectively), as shown in **Table 5**.

The presence of ectoparasites in cats was estimated, and an association was found with the presence of *Dipylidium caninum* egg sacs in feces (**Table 6**). It was a risk factor for this parasite (Chi²= 19.46, P=0.006, OR= 15.40, P= 0.008).

Discussion

This epidemiological study of parasitosis in cats is one of the few or perhaps the only one of its kind conducted in México. The overall prevalence of gastrointestinal parasites in this study was 52.21% (59 positives and 54 negatives). Among the positive cats, 53.82% had a single parasite, 35.87% had two parasites, and 10.31% had three or more parasites. The parasitization rate was lower than the 59% reported by Moskvina *et al.* [6]. The parasite with the highest prevalence in this study was *Dipylidium caninum*, followed by *Ancylostoma*, *Taenia*, *Cystoisospora*, *Giardia*, *Toxocara*, and *Hammondia*, with 70.80% of cats also having fleas. In the study conducted by Moskvina *et al.* [6], they mention that age and the prevalence of the species *Dipylidium caninum* were observed more frequently in cats aged 2 and 5 years or older, with a prevalence of 8% in the microscopic examination but 18% in proglottids found in the feces. The low preva-

lence of this and other cestodes in microscopic techniques is common because proglottids are eliminated intermittently and can be retained in the gauze, preventing the detection of eggs.

RÁCZ *et al.* [1] conducted a study in the streets of São Paulo and Guarulhos to determine gastrointestinal parasite infections in 138 cats, of which 80 (57.97%) tested positive. Among protozoa, the most frequent agent was *Cystoisospora felis* in 36 cats (26.09%), followed by *Cystoisospora rivolta* in 34 cats (24.64%), *Cryptosporidium parvum* (1.45%) in two cats, and *Sarcocystis* spp. in one cat (0.72%). Among helminths, *Toxocaracati* had the highest occurrence with 43 positive cats (31.16%), followed by *Ancylostoma* spp. with 12 positive cats (8.70%). A mixed infection was observed in 25 cats (18.12%), with *T. cati* and *Cystoisospora* spp., as well as *T. cati* and *Ancylostoma* spp., being the most common occurrences, both with 7.97% (11 samples) of occurrence.

Cysticercus fasciolaris is the larval and cystic stage of the tapeworm *T. taeniaeformis*, whose definitive hosts are carnivores from the *Canidae* and *Felidae* families, while rodents and humans serve as intermediate hosts. These larval forms are typically found in the livers of these intermediate hosts, which become infected through the ingestion of water or food contaminated with cat feces [5]. The eggs released from mature proglottids exit the host through feces and, when ingested by a susceptible animal, become activated during their passage through the stomach. The larvae (oncospheres) then migrate to the liver, where they continue to develop within a cyst [4].

Choudhary *et al.* [7] conducted a study in which they mentioned that *T. taeniaeformis* in advanced stages in intermediate hosts demonstrated abundant fibroblasts with neoplastic characteristics such as fibrosarcomas. Therefore, it can be concluded that *T. taeniaeformis* can induce this type of hepatic tumors in rats in advanced cases.

Cryptosporidium and *Giardia* are highly significant in this small number of infections as their stages (oocysts and cysts, respectively) can cause disease in both humans and animals. They are resistant to chlorination and other common water treatments [8]. Infections by *Cystoisospora* are also induced by the ingestion of sporulated oocysts in the environment and can occur at any age and in different management conditions. This includes minimal or severe disease, and effective control of coccidia infections in general requires early intervention to limit tissue damage and environmental contamination with oocysts [2]. *Giardia* spp., however, has been the most frequently detected parasite in dog and cat populations, followed by significant prevalences of ascarids, ancylostomas, and taenia infections [4].

The genus *Giardia* contains six species of anaerobic enteric protozoa isolated from mammals, birds, and amphibians. Among all these species, three infect mammals: *Giardia muris* and *G. microti* in rodents, and *G. duodenalis* has a wide range of mammalian hosts, including domestic cats (Gherman *et al.*, 2018). These findings are also reported in other studies where *Giardia* is considered the most common enteric parasite in dogs and cats in developing countries [4,8].

Not overlooking the fact that Toxocariasis is one of the most significant zoonotic helminth infections in pets, it is primarily caused by the nematodes *T. canis* and *T. cati* in dogs and cats, respectively, as definitive hosts. Additionally, paratenic hosts for *T. canis* have been documented in the literature, including earthworms, mice, rats, chickens, pigeons, sheep, pigs, and even cats. The mode of infection for these hosts is the ingestion of larvated eggs, which are highly resistant to adverse environmental conditions [1,3,8].

Bahk *et al.* [6] reiterated that it is not known for *H. hammondi* to cause clinical disease in any naturally infected intermediate or definitive host. They found microscopic oocysts of *H. hammondi* (referred to as HhCatEt1 and HhCatEt2) in the feces of 2 out of 36 free-roaming domestic cats (*Felis catus*) from Addis Ababa, Ethiopia. These oocysts were orally infectious for Swiss Webster and interferon-gamma gene knockout mice, with the inoculated mice developing tissue cysts in their muscles. Laboratory-reared cats fed with tissues from infected mice shed *H. hammondi* oocysts with a prepatent period of 5 days. DNA extracted from sporulated oocysts reacted with specific *H. hammondi* primers, and the sequences were deposited in GenBank (accession numbers JX477424 and KC223619). This is the first report of *H. hammondi* isolation from cats in the African continent.

Despite the enormous efforts of humans to eliminate parasites from their pets, they continue to be a serious health problem in developed countries, and this impact is even more pronounced in developing countries [6]. The significant impact of zoonoses on human health makes it pertinent and timely to conduct studies that help understand and define the possible risks of transmission of these diseases, especially when they involve pets like dogs and cats that live so intimately with people [2]. Approximately 58% of the 1,400 known human pathogens are of zoonotic origin, and 73% of the 177 pathogens considered by the WHO as reemerging are directly related to people's contact with an animal source [1]. Science fiction movies have even been compared to aspects related to the biology and ecology of parasitic relationships, highlighting mechanisms of transmission and pathophysiology, and even exposing the manipulation by the parasite of the host's behavior by altering its

phenotype. These parasites, in addition to compromising the health of animals, can, under certain conditions, be transmitted to humans, causing various zoonotic diseases [3,5,8]. Risk factors in humans are associated with age, environmental conditions, socioeconomic status, hygiene practices, cohabitation with reservoirs and definitive hosts (González *et al.*, 2017). One of the most challenging global issues is the provision of clean drinking water and soil free from fecal contamination.

The European Scientific Counsel Companion Animal Parasites (ESCCAP) and the Companion Animal Parasite Council (CAPCVet) recommend anti-parasitic treatments every week until 14 days after weaning for puppies and at least four times a year for adult animals. However, it should be noted that frequent treatments can lead to the development of Anthelmintic Resistance (AR). In developing countries, the situation is further complicated due to the uncontrolled growth of the stray canine and feline population, and the fact that many household dogs have outdoor access, increasing the possibility of transmission among animals and to humans [8].

The best way to achieve successful preventive programs is through raising awareness among pet owners, facilitated by veterinarians, about the numerous parasitic diseases that can affect their pets and their potential for transmission to humans (zoonoses) [3,4,5,6]. This includes educating owners about the hygienic measures that should be taken regarding pet waste, especially feces, as potential sources of transmission (handling and disposal of waste at home and during outdoor activities) [6].

Conclusions

The study's results justify the need to improve the control of parasitic infections in cats. This involves administering antiparasitic treatments, maintaining hygiene, and reducing outdoor excursions. Intestinal parasitic infections continue to be prevalent in companion animals, despite the availability of highly effective drugs and control measures taken by owners and veterinarians. The prevalence of parasites in domestic and feral cats in the municipality of Chapala is high, highlighting the necessity to enhance their control.

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