Journal of Advanced Veterinary Research Volume 7, Issue 4 (2017) 87-92

Journal of Advanced Veterinary Research

http://advetresearch.com/index.php/avr/index

A Preliminary Study on the Helminth Fauna in Necropsied Stray Cats (*Felis catus*) in Beni-Suef, Egypt

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ARTICLE INFO

Original Research

Received:
03 August 2017

Accepted:
08 September 2017

Keywords:

Beni-Suef
Helminths
Prevalence
Stray Cats

ABSTRACT

Stray cats play a crucial role in the epidemiology of endoparasites, particularly helminths, due to predating a wide range of both vertebrate and invertebrate hosts, often of veterinary and zoonotic importance. Therefore, a total of 62 stray cats were necropsied in Beni-Suef province, Egypt and examined for helminth parasites. The overall prevalence of infection was 87.0%. The recovered helminths consisted of 10 species of trematodes (*Heterophyes heterophyes*, *Pygidiopsis summa*, *H. nocens*, *Echinochasmus liliputanus*, *Alaria* sp., *Procerovum varium*, *Ascocotyle* sp., *Haplorchis* sp., *Prohemistomum vivax*, *Euparadistomum herpestesi*), five cestodes (*Dipylidium caninum*, *Diplopylidium acanthoterta*, *D. nolleri*, *Joyeuxiella* sp. and *Taenia taeniaeformis*), and two nematodes (*Toxascaris leonina* and larvae of Anisakis simplex). The most prevalent helminths were *Dipylidium caninum* (62.9%), *Toxascaris leonina* (33.8%), *Diplopylidium nolleri* (22.5%) and *Echinochasmus liliputanus* (6.45%). Thirty (48.39%) cats were co-infected by one species, 22 (35.48%) by two and three (4.84%) by more than two species. It has been found that cats aged more than 3 years were the most infected. Both male and female cats were parasitized. The infection was the most prevalent in both summer and autumn. In conclusion, veterinarians must highlight more attention towards both stray and domestic cats, as they are considered reservoir hosts for a wide host range of parasites, particularly helminths, and the zoonotic importance of such parasites should be taken on consideration.

Introduction

Stray cats, *Felis catus*, are known to be as a common predator in the wild natural ecosystem worldwide and they can gain access many sources of preys such as mammals, birds, reptiles, and other invertebrates (Millan and Casanova 2009; Chai *et al*., 2013). Stray cat populations are important as potential reservoir hosts of a variety of parasites posing both veterinary and medical potential importance (Arafa *et al*., 1978).

Such cats live freely in urban and rural areas, and tend to discharge helminth eggs, larvae and protozoan cysts into the surrounding environment (Shahram *et al*., 2002; Bahadori *et al*., 2004; Sharif *et al*., 2007; Zibaei *et al*., 2007; Arbabi and Hooshyar, 2009). They act as reservoirs of various zoonotic infections such as ascarids and hookworms (Labarthe *et al*., 2004). Those parasites may induce great public health problems in both Egypt and worldwide. The frequency of the recovered parasitic species may be affected by several factors; seasons of the year, geographical distribution, habits of the local animal populations, the presence of veterinary care, and the cat populations. Previous surveillance works revealed that stray/feral cats are exposed to a large number of parasitic species, particularly helminths (Calvete *et al*., 1998).

Deposition of the parasitic eggs/oocysts with feces of infected animals in the surrounding environment implies a potential risk to the public health (Robertson and Thompson, 2002). Accordingly, a single nematode may deposit up to 200,000 eggs/day inducing a significant environmental contamination (Sinniah, 1982). Moreover, some eggs are highly resistant to the adverse environmental circumstances and may persist in the soil for long times (O' Lorcain and Holland, 2000). To reduce the widespread of those parasites, it is urgently to prevent the initial environmental contamination through a proper sanitation and disinfection.

Among helminths hosted by cats, the most common tapeworm, *Dipylidium caninum*, and the round worms, ascarids, are the most important gastrointestinal parasites in felids (Robertson and Thompson, 2002).

Therefore, the current study aimed to determine the prevalence of helminths recovered from stray cats necropsied in Beni-Suef province, Egypt, evaluating the distribution pat-

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tern of the recovered trematodes, cestodes and nematodes and the detection of the relationship between infection rates and both age and sex of necropsied cats.

Materials and methods

Collection and examination of cats

Sixty two stray cats obtained from different localities in Beni-Suef province (coordinates: 29°04'N 31°05'E), Egypt during the period from May 2015 to November 2016.

After necropsy, the abdominal cavity was opened and the internal organs of cats including stomach, intestine, liver, lungs and heart were thoroughly examined by the naked eyes. The entire alimentary tracts of all carcasses were removed and the different compartments (oesophagus, stomach, small intestine, caecum and colon) were tightly ligated with fine gauze. The large intestine was opened and the feces were checked for the presence of cestodal segments. The small intestine, in particular, was longitudinally opened with a scissor and washed in 0.85% saline until the supernatant was cleared. The mucosa was scraped with a scalpel. The epithelial scrapings were passed through 60-80 mesh wire sieves. The contents of the sieves were washed with tap water and the intact helminths were collected. The contents of the gastrointestinal tract were then carefully checked with the naked eyes as well as under a stereomicroscope (Georgi and Theodories, 1980; Soulsby, 1982).

Fixing, staining and mounting of adult helminths

The large trematodes were compressed between two slides; small trematodes and cestodes were compressed between two cover slips. The parasites were then fixed in 10% neutral buffered formalin, washed with distilled water, and stained with potassium alum carmine (5 min for small-sized helminths and up to 4 hours for large helminths). They were dehydrated in ascending grades of ethyl alcohol (70, 80, 90, 95, and 100%), cleared in xylene, and mounted with Canada balsam. Intact nematodes were cleared in lactophenol and mounted using a glycerol-jelly mixture onto clean glass slides (Rozsa *et al*., 2000; El-Dakhly *et al*., 2012).

Results

Out of 62 examined stray cats (36 males and 26 females), helminths were detected in 87.0% (54/62) of cats and the recovered helminth fauna consisted of ten species of trematodes [*Heterophyes heterophyes*, *Pygidiopsis summa*, *H. nocens*, *Echinochasmus liliputanus* (Fig. 1), *Alaria* sp., *Procerovum varium*, *Ascocotyle* sp., *Haplorchis* sp., *Prohemistomum vivax*, *Euparadistomum herpestesi*], five cestodes [*Dipylidium caninum*, *Diplopylidium acanthoterta*, *D. nolleri* (Fig. 2), *Joyeuxiella* sp. and *Taenia taeniaeformis* (Fig. 3)], and two nematodes [*Toxascaris leonina* (Fig. 4) and larvae of *Anisakis simplex*] (Table 1).

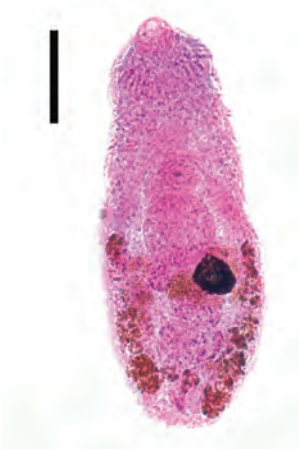

Fig.1. Adult digenean, *Echinochasmus liliputanus*, recovered from the necropsied cats in Beni-Suef province. Scale bar =100 µm

Table1. The overall prevalence of helminths in stray cats in Beni-Suef province (n=62)

Helminths	Number of infected cats	Prevalence
Trematodes	8	12.9
Heterophyes heterophyes	2	3.2
Heterophyes nocens	2	3.2
Haplorchis sp.	1	1.6
Pygidiopsis summa	1	1.6
Procerovum varium	1	1.6
Prohemistomum vivax	1	1.6
Echinochasmus liliputanus	4	6.45
Euparadistomum herpestesi	3	4.8
Alaria sp.	1	1.6
Ascocotyle sp.	1	1.6
Cestodes	53	85.4
Dipylidium caninum	39	62.9
Diplopylidium nolleri	14	22.5
Joyeuxiella sp.	9	14.5
Diplopylidium acanthotetra	5	8.0
Taenia taenaeiformis	6	9.6
Nematodes	22	35.4
Toxascaris leonina	21	33.8
Larva of *Anisakis simplex*	2	3.2

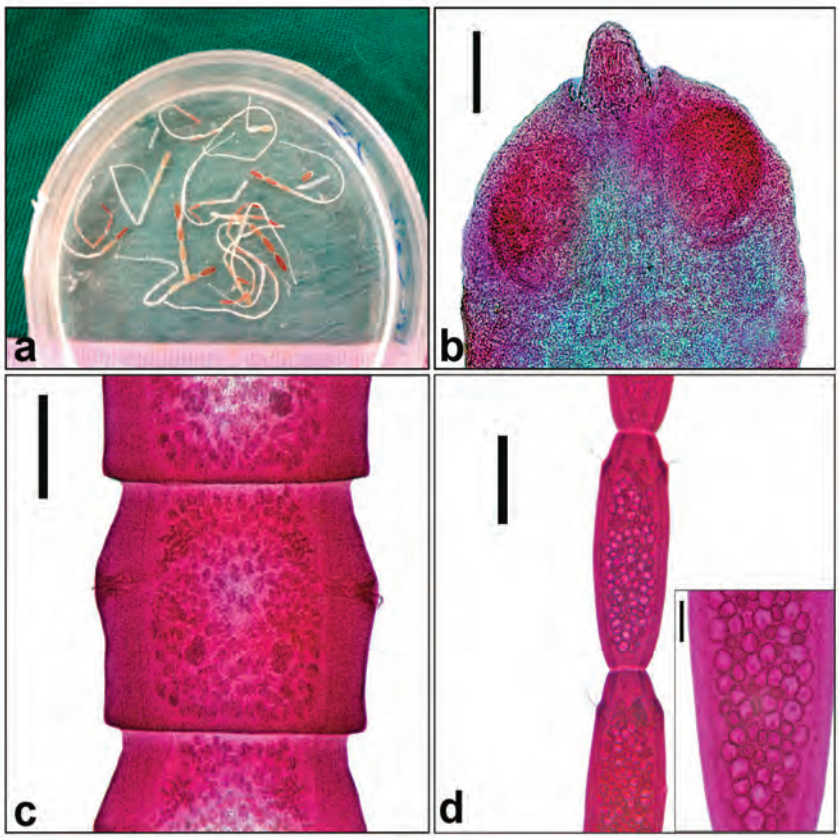

Fig.2. *Diplopylidium nolleri* recovered from the intestinal tract of necropsied cats. a) Intact adult tapeworms. Note the characteristic brownish-red pigmentation in the last portion of strobila. b) Scolex armed with few numbers of claw-hammer-shaped hooks. Scale bar= 100 µm. c) A fully developed mature segment. Note that common genital pores located anterior to the middle of the segment. Scale bar= 500 µm. d) Gravid segment. Note the anteriorly-located common genital pores and egg capsules. Scale bar= 500 µm. Inset: Distinct egg capsules, each with a single ovum. Scale bar= 100 µm.

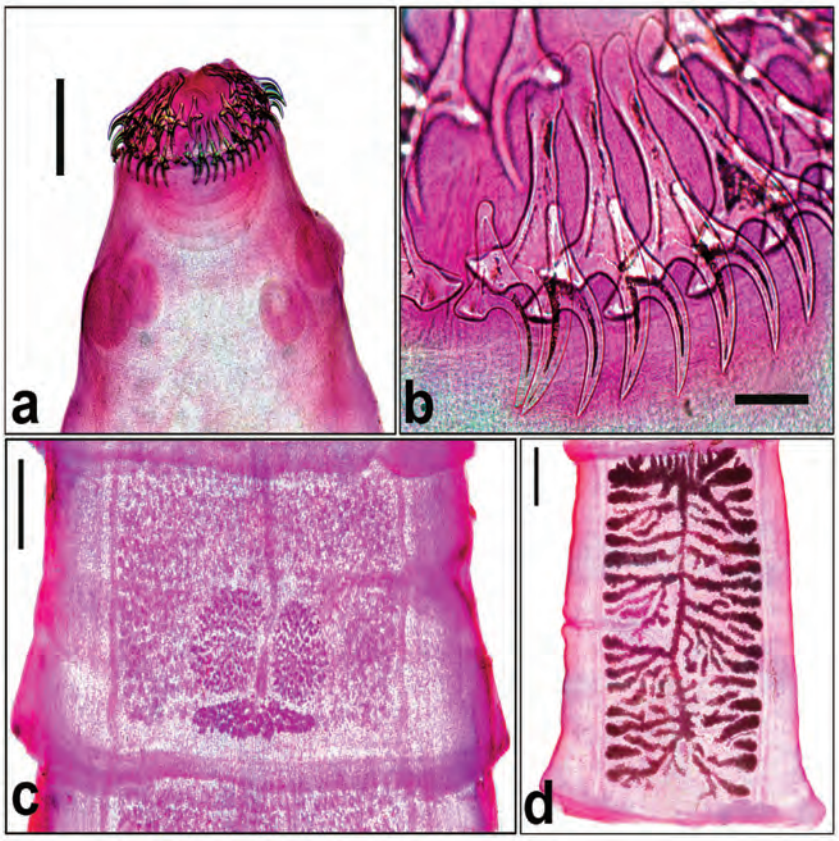

Fig.3. Adult *Taenia taeniaeformis* revealed from necropsied cats. a) Scolex is provided with armed rostellum. Scale bar= 500 µm. b) Typical taenoid hooks. Scale bar= 100 µm. c) Well developed mature segment. Scale bar= 500 µm. d) Gravid segment showing several diverticula filled with fertilized eggs. Scale bar= 1 mm.

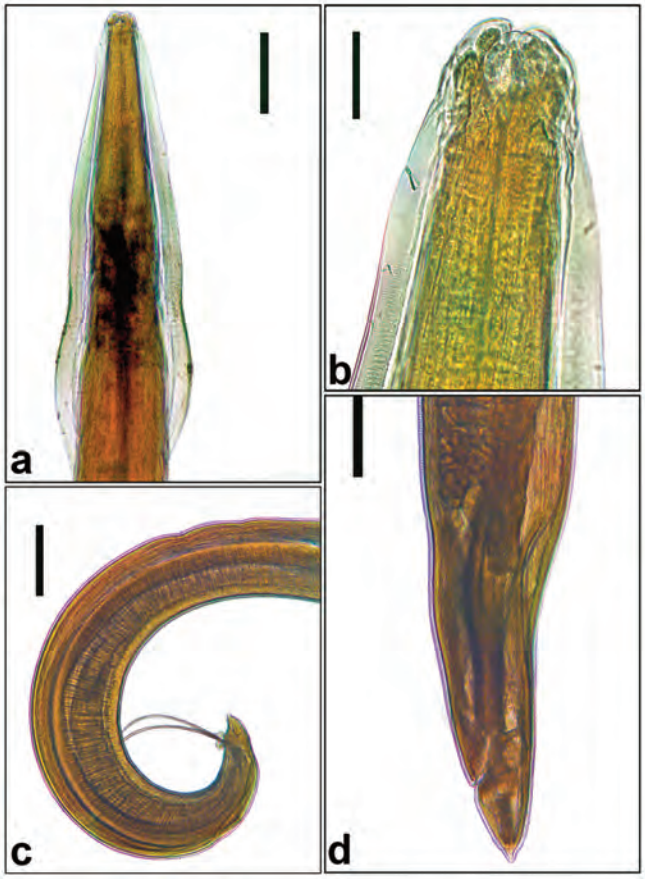

Fig.4. Adult *Toxascaris leonina* recovered from necropsied cats. a) The anterior end showing the typical arrow-headed shape. Scale bar = 500 µm. b) Well distinct fine striations of the cervical alae. Scale bar = 100 µm. Inset: Well defined 3 lips at the most anterior end. Scale bar = 50 µm. c) The posterior end of adult male showing a pair of subequal spicules. Scale bar = 500 µm. d) The posterior end of adult female. Scale bar = 500 µm

Thirty (48.39%) were parasitized by one species, 22 (35.48%) by two and three (4.84%) by more than two species. It was found that cats with multiple infections were lower in number than those with single and double infections, with the former was relatively higher than the later (Table 2).

Table 2. Co-infection of helminths in stray cats in Beni-Suef province

Infection	Infected cats	
	Number	Percentage
Single infection		
Trematodes	0	0
Cestodes	28	45.16
Nematodes	2	3.23
Total	30	48.39
Double infection		
Trematodes + cestodes	5	8.06
Trematodes + nematodes	0	0
Cestodes + nematodes	17	27.42
Total	22	35.48
Multiple infection	3	4.84

The prevalence of digenean trematodes was 12.9%; *Heterophyes heterophyes* 3.2%, Heterophyes nocens 3.2%, *Pygidiopsis summa* 1.6%, *Echinochasmus liliputanus* 4.8%, *Alaria* sp. 1.6%, *Procerovum varium* 1.6%, *Ascocotyle* sp. 1.6%, *Haplorchis* sp. 1.6%, *Prohemistomum vivax* 1.6%, *Euparadistomum herpestesi* 4.8%.

Tapeworms of the family Dipylidiidae were the most frequently recorded helminths. The differentiation between *Joyeuxiella* sp. and *D. nolleri* was clear and did not require staining since the freshly isolated mature segments of the later species are dark red to reddish brown with smaller-sized scolex armed only with four rows of hooks. The prevalence of *D. caninum* and *D. nolleri* was 62.9% and 22.5% respectively. *Diplopylidium acanthoterta*, *Joyeuxiella* sp. and Taenia sp. were detected in prevalences of 8.0, 14.5 and 9.6%, respectively. *Toxascaris leonina* nematode was found in a prevalence of 33.8%. Moreover, larvae of Anisakis simplex ware recorded in 2 (3.20%) cats (Table 1).

It has been found that the helminthosis was age related with the highest infection rates in cats aged more than 3 years (Table 3). Moreover, female cats had relatively higher infection rates than males (Table 4). Meanwhile, the current study revealed that all infected cats harbored cestodes in both summer and autumn, with the highest infection rate with digeneans in winter, while nematodes were more abundant in both autumn and winter (Table 5).

Table 3. The prevalence of helminths in stray cats relative to the age

Helminths	Up to one year		1 – 3 years		More than 3 years	
	No.	%	No.	%	No.	%
Helminths	0	0	2	3.23	6	9.68
Cestodes	13	20.97	12	19.35	28	45.16
Nematodes	6	9.68	4	6.45	12	19.35

The highest prevalence rate of helminth infection was found in cats aged more than 3 years.

Table 4. The prevalence of helminths in stray cats relative to sex

Helminths	Infected males		Infected females		Total	
	No.	%	No.	%	No.	%
Trematodes	2	3.2	6	9.6	8	12.9
Cestodes	27	43.5	26	41.9	53	85.4
Nematodes	16	25.8	6	9.6	22	35.4

Table 5. The prevalence of helminths in stray cats relative to seasonal variation

Class/ season	Summer (*n*=14)		Autumn (*n*= 14)		Winter (*n*=19)		Spring (*n*=15)	
	No.	%	No.	%	No.	%	No.	%
Trematodes	3	21.4	0	0	4	28.6	1	6.6
Cestodes	14	100	14	100	13	92.8	12	80
Nematodes	5	35.71	9	45	6	42.8	2	13.3

Discussion

The present study revealed that the overall prevalence of helminths in necropsied stray cats in Beni-Suef, province, Egypt was 87.0%. Such finding was similar to those obtained by Calvete *et al.* (1998) in Spain (89.7%); Shahram *et al.* (2002) in Iran (98.5%); Labarthe *et al.* (2004) in Brazil (89.6%); Schuster *et al.* (2009) in Dubai (87.0%); Borthakur and Mukhariee (2011) in India (85.2%); Khalafalla (2011) in Egypt (91%); Thabit (2011) in Assiut, Egypt (91.7%). The higher prevalence of helminths in stray cats indicated that the meterological conditions are suitable for the spread and survival of eggs/segments of cestodes. The findings confirmed a trend in Egypt of an increased

prevalence of helminth infections in cats, particularly cestodes and nematodes. This might be attributed to the poor hygiene and lack of anthelmintic drugs used. The infection of cats with helminths can occur either through the ingestion of infective stages or as a result of feeding on paratenic hosts (rodents) containing larvae and cysts in their tissues.

Although Thabit (2011) detected that mixed infection is actually expected to be more prevailing as cats, the definitive host, may harbour the infective stages of a variety of helminths e.g. fish, rodents, insects, the present study revealed that cats with multiple infections were lower in number than those with the single or double infections. This finding might be attributed the uneven distribution of intermediate hosts.

In the present study, 10 species of trematodes were detected with a total prevalence 12.9%. This is in the range of previous studies recorded by Schuster *et al*. (2009); Chai *et al*. (2013); Fang *et al*. (2015). On the other hand, the current findings were lower than those recorded by Fouly (1997) who detected that the prevalence of trematodes was 42%; Thabit (2011) revealed that the prevalence of trematodes was 61.6%; Chai *et al*. (2013) reported 23 *digenean* sp.; El-Azazy *et al*. (2016) identified 14 species of trematodes with a prevalence of 24.6%.

In different localities of Egypt, Kuntz and Chandler (1956) detected 14 trematode species (*Heterophyes heterophyes*, *Heterophyes aequalis, H. pumilio, H. taichui, H. yokogawai, S. falcatus, Pygidiopsis genata, Phagicola longicollis, P. ascolonga, Stictodora sawakinensis, Echinochasmus liliputanus, Stephan oprora denticulatoides, Mesostephanus appendiculatus and Cynodiplostomum namrui*) from domestic cats. Most of these trematode species detected from cats are zoonotic and transmitted via eating raw flesh of freshwater/brackish water fish. Accordingly, it is suggested that stray cats may play an im¬portant role as reservoir hosts in endemic areas of zoonotic trematodes.

Currently, the most common trematodes were those of Family Heterophyidae (*Heterophyes heterophyes*, *H. nocens*, and *Pygidiopsis summa*). Similarly, El-Azazy *et al*. (2016) revealed that the most common trematodes belonged to the genus Heterophyes, particularly *H. heterophyes* and *H. dispar* (15.8% and 10.8%), revealing that stray cats are good indicators of fish-borne trematodes in the environment. Eom *et al*. (1985) described heterophyid flukes, including *H. continua, H. nocens,* and *P. summa*, from cats purchased at Jungang Market in Seoul. Meanwhile, Khalafalla (2011) mentioned that the infestation rate of *Heterophyes heterophyes* was 3%; Schuster *et al*. (2009) recorded that the infestation rate of *Heterophyes heterophyes* was 2.5% and Heterophyopsis continua was 0.4%

Echinochasmus liliputanus and *H. pumilio* are intestinal flukes infecting canids, felids and humans as definitive hosts. In the present work, those trematodes were found in a prevalence of 6.45% and 1.6%, respectively. Oppositely, Thabit (2011) and Fang *et al*. (2015) detected these trematodes in a higher prevalence (28.3% and 53.3%, respectively).

Euparadistomum herpestesi isolated from the gall bladder of three infected cats with a prevalence of 4.8%. Feeding habits of cats i.e. predating rodents or reptiles containing encysted meraeracariae, infective stages, allow the development of such flukes (Ravikumar *et al*. 1979). Large populations of stray cats are present in Egypt, so, it is advisable to do further studies on such fluke to determine all proposed Euparadistomum species.

The overall prevalence of cestodes was 85.4%. Such finding agreed with that obtained by Arbabi and Hooshyar (2009) who recorded cestodes in a prevalence of 90.3%. On the other hand, Khalafalla (2011) recorded a higher prevalence (23%). *Dipylidium caninum* had the highest prevalence (62.9%) of cestodes that were recovered from the gastrointestinal tract of stray cats. Similar results were recorded by Labarthe *et al*. (2004) (52.6%); Arbabi and Hooshyar (2009) (68.1%); Schuster *et al*. (2009) (69.2%); Knaus *et al*. (2011) (83.3%); Waap *et al*. (2014) (53.1%).

The prevalence of *D. caninum* was significantly high due to the appropriate environmental conditions for the flea instars. The high individual worm burdens with *D. caninum* might be due to the ability of the larvae to ingest the whole egg capsules from the feces of cats (Nichol *et al*., 1981). Humans, particularly children, acquire the infection by accidentally ingesting infected fleas. On the other hand, Barutzki and Schaper (2003) and Mircean *et al*. (2010) revealed that the prevalence of *D. caninum* was 0.1% and 0.2%, respectively.

In the present investigation, *D. nolleri* was found in a prevalence of 22.5%. This might be lower than previously recorded by El Shabrawy and Imam (1978) in Egypt (36.4%) and Changizi *et al*. (2007) in Iran (54%). The later elucidated that in Iran, humid climates, favourable conditions for transmission of the parasite are adequate, but dry areas are unsuitable for the survival of the insect intermediate hosts.

Joyeuxiella pasqualei and *Diplopylidium acanthotetra* showed a lower prevalence compared to previous studies (Calvete *et al*., 1998, 55%; Millan and Casanova, 2009, 21%). Interestingly, the prevalence and abundance of these two tapeworms were strongly synchronized, and may indicate the use of a similar intermediate host. Cysticercoids of Joyeuxiella and Diplopylidium are often found in snakes, lizards, and amphibians (Schuster *et al*., 2009). There is scarce literature about lizards in Egypt, and cats rarely predate upon snakes.

The prevalence of *Taenia taeniaeformis* was 9.6%, which was similar to that detected by Sohn and Chai (2005) (14.6%); Zibaei *et al*. (2007) (12.3%); Arbabi and Hooshyar (2009) (15%). Oppositely, such result was lower than that recorded by Borthakur and Mukhariee (2011) (70.4%) and Borkataki *et al*. (2013) (60%). Coman (1972) mentioned that the prevalence of the cestode was related to the existence of mice in the diet of cats. The low worm burden might be referred to the rodent intermediate host carrying the strobilocercus.

Herein, *Toxascaris leonina* was found with the highest prevalence (30.4%). This was higher than the estimated prevalence previously revealed by Labarthe *et al*. (2004); Zibaei *et al*. (2007); Michaleczyk and Sokol (2008) and Khalafalla (2011) (11.9, 12.9, 15.4, and 5.0%). Cats usually defecate at night in sandy soils and bury the feces. Such a habit is favourable for ascarid eggs protecting them against desiccation. Furthermore, some helminths such as Toxascaris eggs are resistant to low temperatures and high humidity, but the infectivity decreased as a result of desiccation and direct sunlight.

The age of cats was a potential risk factor associated with parasitic infection. It is probable that the infection may occur at any age, either by eggs/tissues containing larvae, although the highest prevalence of infection exists in elderly cats. However, previous literature revealed the highest infection rate in young ages particularly less than 6 months (Lorenzini *et al*., 2007; Sharif *et al*., 2007; Itoh *et al*., 2012). This can significantly attribute to the dissemination of viable helminth eggs into the environment, and a temperate climate appears to enhance the embryonation of helminthes eggs in the soil and their potential transmission to humans.

The current investigation indicated that the infection with flukes was higher in females than in males, but concerning cestodes and nematodes, the reverse was the case. Oppositely, Thabit (2011) revealed that digeneans were higher in males than females, while tapeworms and roundworms were higher in females than males. Arafa *et al*. (1978) reported that female cats were possibly more vulnerable to the infection than males. Fouly (1997) showed that the infection rate in adult cats was affected by the sex as all parasites were more abundant in males than in females. Meanwhile, Thabit (2011) elucidated that the infection rates among male or female cats are not re-

lated to the sex.

Currently, the prevalence of infection was the highest in both summer and autumn (100%). Similarly, Arafa *et al*. (1978) and Thabit (2011) observed that the infection rate was higher in hot seasons. The effect of seasonal variations may be not only due to climatic conditions but also due to the activity of cats and their available food in different seasons. It is worthy to mention that adult helminths found in cats during the summer actually resulted from the infection occurring few months ago.

Conclusion

In conclusion, *Dipylidium caninum*, *Dipyloplydium nolleri*, *Taenia taeniaeformis*, and *Toxascaris leonina* are the most common helminths in necropsied cats in Beni-Suef provience, Egypt. Older cats had the highest prevalence of infection than young ones.

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