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Prevalence of parasitic infections in surgically removed appendices: parasitological and histopathological studies

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Summary

Intestinal parasites may cause symptoms similar to acute appendicitis. Moreover, the diagnosis of parasitic infections is only done by post-operative histopathological examination of the appendices. Therefore, our aims are to assess the prevalence of intestinal parasitic infections among patients who were be appendectomized at Tanta Hospitals, Egypt and to investigate the possible association between these parasitic infections and appendicitis. To achieve these objectives, we performed a cross-sectional study including 65 patients chosen randomly who had undergone appendectomy over a period of one year from Oct 2015 to Oct 2016. Demographic data were retrieved. Complete blood picture was done. Moreover, appendiceal faecolith were examined macroscopically then by direct smear examination, formol-ether concentration technique, modified Ziehl-Neelsen stain and rapid immunochromatographic test. Histopathological examination of resected appendices was done. We found that parasitic infections were detected in 24.6 % of examined cases. Most of parasitic infections were prevalent in patients belonging to the school age group. Different parasitic infections were detected in the faecolith specimens. Moreover, *Enterobius vermicularis* adult female and *Schistosoma mansoni* granuloma were detected in histopathological sections. Also, a spectrum of pathological changes in the appendices was found ranging from lymphoid hyperplasia to acute inflammation with peritonitis. In conclusion, intestinal parasites may cause clinical picture similar to that of acute appendicitis. Therefore, careful attention to clinical history, stool examination and high eosinophilia may aid diagnosis and avoid unnecessary appendectomy. Moreover, the presence of different parasitic stages in the narrow lumen of the appendix may have a role in the development of appendicitis and this needs further studies.

Keywords: appendicitis; immunochromatographic test; intestinal parasites; *Hymenolepis nana*; *Enterobius vermicularis*; appendiceal faecolith

Introduction

Parasitic infections are common on a worldwide basis and are seen in high numbers of developing countries especially in Africa where a warm, moist climate and standards of hygiene are low. However, parasitic diseases are now occurring in developed

countries in higher frequency than before due to immigration and increased foreign travel (Jan *et al.*, 2010).

Many health problems result from these infections, including malnutrition, iron-deficiency anemia and surgical morbidities such as intestinal obstruction, cholecystitis, liver abscess and appendicitis which need surgical intervention (Hesse *et al.*, 2012).

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Table 1. Distribution of parasites in relation to the age and gender of positive studied cases.

Age group	N.	%	P value	Gender	Positive	X ²	P-value
6 – 18 y.	10	62.5	0.157	Male	N 9	12.802	0.001*
					% 90 %		
				Female	N 1		
					% 10 %		
> 18 y.	6	37.5		Male	N 4	1.332	0.248
					% 66.67 %		
				Female	N 2		
					% 33.33 %		

* extremely significant

Acute appendicitis is a common cause of an emergent surgical intervention. The prevalence of appendicitis is reported to be around 7 %, being slightly higher in male patients. It can be seen at any age but the peak incidence occurs in older children and young adults (Yabanoglu *et al.*, 2014).

In the 21st century the incidence of appendicitis is higher in newly industrialized countries in Asia, South America and the Middle East as compared with Western countries due to different environmental exposures (Ferris *et al.*, 2017).

There are many theories for pathogenesis of appendicitis; one of these theories is the obstruction of the lumen of the appendix. The other theory is really a group of theories, which assumes the existence of a labile factor in the appendix that responds to a variety of external and internal stimuli that causes injury to the appendix which permits bacterial invasion by the flora normally present (Bohrod, 1946). Moreover, changes in symbiont composition in the ecosystem of the human body have led to immune dysfunction and subsequent disease (The biota alteration theory, or biome depletion theory); that may predispose to appendicitis (Parker, 2017).

These theories may account for the association of appendicitis with distant infections and the influence of climatic and other environmental alterations.

It has been reported that the cause of appendicitis is intraluminal obstruction caused by lymphoid hyperplasia, parasite-associated fecal matter and ingested foreign bodies (Pasupati *et al.*, 2008).

Some parasites such as *Enterobius vermicularis*, *Ascaris lumbricoides*, *Schistosoma* spp. and *Taenia* spp. are reported in the appendectomy specimens. Also, some protozoa are reported such as *Entamoeba histolytica*, *Balantidium coli* and *Cryptosporidium parvum* (Hegazi, 2012).

Therefore, the current study aimed to assess the prevalence of intestinal parasitic infections among patients who were appendectomized at Tanta University Hospitals, Egypt and to investigate the possible association between these parasitic infections and appendicitis through parasitological and histopathological examinations of the surgically removed appendices.

Patients and Methods

A cross-sectional study was conducted within one year from October 2015 to October 2016 on 65 cases of appendectomized patients chosen randomly after taking their consent. Arrangement with General Surgery department, Tanta University Hospitals to obtain the appendices after the surgery was done. Interviewing questionnaire including demographic data such as age, sex,

Table 2. Detected parasites in positive studied cases.

Parasites in positive cases	Methods of detection	No.	%
<i>Enterobius vermicularis</i>	Histopathological examination of appendiceal specimens	1	6.25 %
<i>Schistosoma mansoni</i>		1	6.25 %
<i>Hymenolepis diminuta</i>	Microscopic examination of appendiceal faecolith	1	6.25 %
<i>Hymenolepis nana</i>		2	12.5 %
<i>Giardia duodenalis</i>		4	25 %
<i>Entamoeba histolytica</i>		4	25 %
<i>Cryptosporidium</i> spp.		2	12.5 %
<i>Blastocystis</i> spp.		1	6.25 %
Total		16	100 %

name, level of education, residence and clinical data such as abdominal colic, vomiting and diarrhea was done.

Parasitological study

The luminal contents of surgically removed appendices (appendiceal faecolith) were examined macroscopically for consistency, color, odor and the presence of blood, mucus and parasites. Also, they were examined microscopically through:

Direct smear examination: One or two drops of saline was added to the faecolith with a pipette and mixed with pipette tip. The specimen was examined with the low power objective lens (10x) and low light according to Fleck and Moody (1993).

Preserved in 10 % formalin and examined later by formal-ether concentration technique to diagnose intestinal parasitic ova and cysts according to Fleck and Moody (1993).

Stained by modified Ziehl-Nelsen stain to detect intestinal protozoa. Faecal smears were stained with strong carbol fuchsin for 15 – 20 minutes. They were decolorized in acid alcohol (1 % HCl

in methanol) for 15 – 20 seconds. Then, they were counterstained with 0.4 % malachite green (or methylene blue) for 30 – 60 seconds according to Rosenblatt *et al.* (2009).

Copro antigen of *Cryptosporidium* spp., *Giardia lamblia* and *Entamoeba histolytica* were detected in these appendiceal faecolith by rapid immunochromatographic test (Rida Quick *Cryptosporidium/ Giardia/ Entamoeba* Combi Art No, N1723) according to Regnath *et al.* (2006). The test was performed according to manufacturer's instructions. Results were interpreted following the manufacturer's guidelines.

Any appendiceal specimens or faecolith were discarded properly after testing by safe disposal measures in Tanta Faculty of Medicine general incinerator according to the safe disposal rules.

Laboratory blood picture

Blood samples were collected from patients and complete blood picture was done to detect anemia, leukocytosis and eosinophilia. The blood film was fixed with methyl alcohol for 2 minutes. Giemsa

Fig. 1. Luminal content of appendiceal specimens showing: A: *Entamoeba histolytica* cyst with concentrated iodine stain (x1000). B: *Giardia duodenalis* cyst with concentrated iodine stain (x1000). C: *Blastocystis* cyst with concentrated iodine stain (x400). D: *Cryptosporidium* oocysts with modified Ziehl-Nelsen stain (x1000).

Fig. 2. Luminal content of appendiceal specimens with concentrated iodine stain showing: A: *Hymenolepis nana* ova (x400). B: *Hymenolepis diminuta* ova (x100) (left) and (x400) (right).

stain 1:9 dilution was poured with buffer over the smear for 8 – 10 minutes. Then, the film was washed off with buffer and dried. The dry and stained films were examined without a coverslip under oil immersion objective (Houwen, 2002).

Histopathological examination

Two transverse sections from the base and the middle portion of the appendix and a longitudinal section from the tip of the appendix were taken. Sections of 4 μ m thickness were prepared and stained with haematoxylin and eosin. The histologic diagnosis was confirmed by reviewing one to four original sections of the specimen. Stained sections of appendiceal specimens were subjected to microscopic examinations to detect any pathological changes such as inflammation, granuloma of the appendix and the presence of parasites (Allen, 1992).

Statistical analysis

Quantitative values of the measured parameters were expressed as mean $\pm$ standard deviation (SD). The data were analyzed by one way-ANOVA to determine significance of differences between groups using Statistical Package for Social Sciences (SPSS), version 14.0. The probability of significant differences was determined by chi-square test for the histopathological studies. Differences were considered significant at $P < 0.05$ and extremely significant at $P < 0.001$.

Results

Parasitic infections, helminth and protozoa, were found in 16 (24.6 %) of cases in this study either by fecolith or tissue sections examined. The present study was conducted on two age groups: school age (6 – 18 years, 60 % of appendectomized cases) and

Table 3. Sensitivity and specificity of different diagnostic techniques for stool examination.

	<i>Giardia duodenalis</i>		<i>Entamoeba histolytica</i>		<i>Cryptosporidium</i> spp.	
	Formol ether technique	Immunodiagnostic technique (Copro antigens)	Formol ether technique	Immunodiagnostic technique (Copro antigens)	Formol ether technique	Immunodiagnostic technique (Copro antigens)
Sensitivity	75	100	100	100	-	-
Specificity	-	-	100	-	100	100
PPV	100	100	100	75	-	-
NPV	-	-	100	-	100	100
Accuracy	75	100	100	100	100	100

PPV: positive predictive value

NPV: Negative predictive value

Fig. 3. Chart of diagnostic performance of different diagnostic techniques for stool examination.

adult age (19 – 60 years, 40 % of cases). In the school age group, there were 29 males and 10 females while in adult age, there were 17 males and 9 females. Among 16 parasitic infected cases, ten cases (62.5 %) were in school age. A significantly higher prevalence of parasitic infections was detected in males of this group (90 %) compared to females. While six cases (37.5 %) were in adult age with also increase in the prevalence of parasitic infections among male (66.6 %) of this group (Table1). Regarding residence, Out of the 16 positive parasitic infected cases, there were 13 (20 %) from rural areas while three (4.6 %) from urban areas. Ten cases had history of diarrhea with or without blood. A single case from all studied patients had a history of *Schistosoma mansoni* infection.

Parasitological study

Parasitological examination of luminal content of removed appendices and histopathological examination of stained specimens

revealed that 16 (24.6 %) positive cases from a total of 65 appendectomized patients had parasitic infections.

Out of the 16 positive parasitic infected cases, there were four cases had *Giardia duodenalis* cysts (25 %), four cases had *Entamoeba histolytica* cysts (25 %), two cases had *Cryptosporidium* spp. oocysts (12.5 %), two cases had *Hymenolepis nana* ova (12.5 %), one case had *H. diminuta* ova (6.25 %) and one case had *Blas-tocystis* spp. cysts (6.25 %) that were detected by examination of appendiceal faecolith (Figs. 1 and 2). Other two cases, *Schistosoma mansoni* ova (6.25 %) and *E. vermicularis* adult (6.25 %), were detected by histopathological examination of stained specimens. Regarding different methods of stool examination used in diagnosis of parasitic infections, the number of positive parasitic cases detected by direct smear examination (17 %) was higher than those detected by formol-ether concentration technique (15.4 %), immunodiagnostic technique (12.3 %) or modified Ziehl-Neelsen (3.1 %). While the number of positive cases of *Cryptosporidium*

Table 4. Blood picture findings in school age group.

School age		Range	Mean $\pm$ S. D	t. test	p. value
Hb g/dl	Infected positive cases	8 – 12	10.10 $\pm$ 1.29	20.777	0.001*
	Negative cases	10 – 12	11.45 $\pm$ 0.57		
TLC /mm ³ of blood	Infected positive cases	8000 – 11000	10.4 $\pm$ 6.9	0.208	0.651
	Negative cases	1000 – 11000	10.3 $\pm$ 4.7		
Eosinophilic Count %	Infected positive cases	10 – 15	13.80 $\pm$ 2.04	46.152	0.001*
	Negative cases	6 – 8	8.00 $\pm$ 0.00		

* extremely significant

Fig. 4. Gross appearance of inflamed appendices showing: A: inflamed appendices removed for acute appendicitis showing swollen, dull serosa. B: congested appendix filled with faecolith. C: chronic appendicitis showing marked thickening in the wall and fat creeping on serosa.

spp. detected by modified Ziehl-Neelsen was higher than other methods (Fig. 3).

Using direct smear as the gold standard, the sensitivity of formol-ether concentration and immunodiagnostic techniques for *E. histolytica* detection were 100. While the sensitivity of formol-ether concentration and immunodiagnostic techniques for *G. duodenalis* detection were 75 and 100, respectively. The specificity of formol-ether concentration and immunodiagnostic techniques for *Cryptosporidium* was 100 (Table 3).

Laboratory blood picture findings

In this study complete blood picture was done. Regarding haemoglobin (HB) count in school age group, there was anemia in cases infected with parasites as their HB level ranged from 8 to 12 g/dl in appendectomized infected patients with a significant difference between infected and non infected cases. While in adult group there was no difference between infected and non infected cases. Leucocytic count in all age groups ranged from 8000 – 11000 with a significant difference between parasite-infected and non infected cases in adult age group. Concerning the eosinophilic count in all age groups, there was elevation of the count (10 – 15 %) in para-

sitic infected appendectomized cases in comparison to non infected cases with significant difference between them (Table 4 and 5).

Histopathological findings

Gross appearance of inflamed acute appendices showed swollen, dull serosa. Some of them are congested filled with faecolith. Moreover, subacute appendicitis cases showed marked thickening in the wall associated with fat creeping on serosa (Fig. 4). Histopathological features of acute inflammation of the appendix were evident in 44 (67.7 %) cases in the form of transmural infiltration by acute inflammatory cells, mainly polymorphnuclear leucocytes (PNLs), pus cells and macrophages (Table 6). Fourteen cases (21.5 %) of acute appendicitis showed eosinophilic infiltration that was associated with parasitic infections. Moreover, there was one case of acute suppurative inflammation associated with *E. histolytica* infection as appeared by PAS stain that stained trophozoites red (Fig. 5).

Subacute appendicitis was represent in 19 (29.2 %) of the studied cases. These cases showed chronic inflammatory reaction associated with reactive follicular hyperplasia in Peyer's patches and lymphoplasmacytic infiltration of the appendix wall.

Table 5. Blood picture findings in adult age group.

	Adult age	Range	Mean $\pm$ S. D	t. test	p. value
Hb g/dl	Infected positive cases	10 – 12	11.17 $\pm$ 0.75	1.142	0.296
	Negative cases	11 – 12	11.45 $\pm$ 0.51		
TLC /mm ³ of blood	Infected positive cases	9000 – 12000	10.5 $\pm$ 1.22	14.740	0.001*
	Negative cases	9000 – 11000	9.2 $\pm$ 5.23		
Eosinophilic count %	Infected positive cases	10 – 15	12.83 $\pm$ 2.23	75.391	0.001*
	Negative cases	6 – 8	6.10 $\pm$ 0.45		

* extremely significant

Fig. 5. Histopathological changes of resected appendices showing: acute suppurative appendicitis showing ulceration of the mucosa and amoeba trophozoites are found among pus cells in the lumen (arrows) (PAS stain x400).

Negative appendices (no acute inflammatory cellular reaction) were represented in 3.08 % of cases. One of them showed non gravid *E. vermicularis* adult female in the submucosa associated with mild eosinophilic infiltration and intact mucosa (Fig. 6A, B). Another case showed remnant of *S. mansoni* ova surrounded by a granuloma formed of epithelioid cells, eosinophils, lymphocytes, foreign body giant cells, fibroblasts and fibrous tissue (Fig. 6C, D).

Discussion

Appendicitis is considered a common cause of emergent abdominal surgical procedures. Moreover, epidemiologic studies have revealed that approximately 7 % – 12 % of the population would have appendicitis in their life time (Hardin, 1999; Mowlavi *et al.*, 2004).

The aetiology of acute appendicitis has not been established and is still much debated. Several factors have been suggested which include diet, lymphoid hyperplasia, faecolith, and infections due to bacteria, viruses and parasites (Pasupati *et al.*, 2008).

The association between parasitic infection of the appendix and acute appendicitis has been widely investigated. Therefore, this study was conducted to determine the prevalence of intestinal parasites in luminal content (faecolith) and tissue sections of appendectomy specimens in Tanta University Hospital, Egypt.

Parasitic infections, helminth and protozoa, were found in 16 (24.6 %) of cases in this study by faecolith and tissue sections examined. This percentage is close to reports of Pasupati *et al.* (2008) in Malaysia. On the other hand, Ebrahim (2010) reported the presence of parasitic infections in (57 %) appendix specimens in Alexandria, Egypt. Low incidences of parasitic infections were found in 5.5 % of appendices in Oman and Turkey (Zakaria *et al.*, 2013; Yabanoglu *et al.*, 2014). Moreover, 9 % parasitic infections in appendix specimens were reported by Abdellatif *et al.* (2015) in Egypt.

Recently in Egypt, a retrospective study of appendectomies by Hedyia *et al.* (2012) reported that 11 out of 251 specimens (4.38 %) had parasitic infections. In 2014, Jada *et al.* detected a wide spectrum of parasitic infections (48 %) in one hundred surgically removed appendices in India.

Out of the 16 positive specimens detected in this study by either faecolith or tissue sections examined, there were *Giardia lamblia* cysts (25 %), *E. histolytica* cysts and trophozoites (25 %), *Cryptosporidium* spp oocysts. (12.5 %), *H. nana* ova (12.5 %), *Schistosoma mansoni* ova (6.25 %), *E. vermicularis* adult (6.25 %), *H. diminuta* ova (6.25 %) and *Blastocystis* spp.cysts (6.25 %).

There was slight preponderance of males to females in this work. The percentage of parasitic infections was generally higher in males and this coincides with a recent study by Abdellatif *et al.*

Fig. 6.: Histopathological changes of resected appendices (H&E) showing: A: non gravid adult female *E.vermicularis* in the submucosa of the appendix with inflammatory infiltrate and intact mucosa (x100). B: higher magnification of A showing characteristic lateral alae, the gut and the uterus of the worm with lymphoplasmacytic and eosinophilic inflammatory infiltrate surrounding it (x400). C: *Schistosoma mansoni* granuloma formation in the submucosa surrounding remnant of the ova (x100). D: higher magnification of C showing granuloma formed of epithelioid cells, lymphocytes and foreign body giant cells all surrounded by fibroblasts and fibrous tissue (x400).

(2015) who reported that the prevalence of infection, male: female ratio was 2.1: 1.2. On the contrary, Ahmed *et al.* (2015) mentioned that females are more vulnerable gender to be infected. However, no statistical significant difference was found between the rate of infections in males and females in his study.

Concerning age in this study, parasitic infections were higher in the age group ranging from 6 – 18 years. This finding is consistent with studies done by Pasupati *et al.* (2008) and Zakaria *et al.* (2013) who found infections in ages less than 22 years old. Therefore, there is a considerable range of reported prevalence of parasitic appendicitis in both children and adults, which likely reflects differences in parasite endemicity, demographic factors, and differences in diagnostic method (Hegazi and Patel, 2012)

This study was conducted in Gharbiya Governorate which is considered a rural -urban area (El-Khoby *et al.*, 2000). Most of the parasites detected in the current work are transmitted by faeco-oral route and are widely spread in rural areas. This may explain the significant difference detected in infections according to the patients residence in this work.

These results coincide with the findings of other studies focused

on rural areas around the world such as Tang and Luo (2003) in China, Çeliksöz *et al.* (2005) in Turkey, Ikeh *et al.* (2006) in Nigeria, Jacobsen *et al.* (2007) in Ecuador and Ngrenngarmlet *et al.* (2007) in Thailand, where the infection rate was peaking in comparison to the urban areas. The high rate is back to environmental factors, poor personal hygiene and lack of health education. Therefore, all patients are at risk of harboring intestinal parasites. *S. mansoni* granuloma was found in one case with no acute inflammatory changes of appendiceal specimens were observed. This observation is in accordance with Duzgun *et al.* (2004) who found one case of *S. mansoni* infection and mentioned that the role of *S. mansoni* infection in appendicitis is doubtful. On the other hand, Abdellatif *et al.* (2015) mentioned that *Schistosoma* granuloma was important cause of appendicitis. Other reports mentioned that the actual role of *S. mansoni* infection in the development of appendicitis is still open to debate and has been the subject of controversy (Limaïem *et al.*, 2015). Also, it is mentioned that schistosomal appendicitis occurred when obstruction of the lumen by long standing granulomatous reaction around *Schistosoma* ova occurred (Satti *et al.*, 1987).

Table 6. Histopathological finding of appendix specimens.

Histopathologic type	Acute appendicitis		Subacute appendicitis		Negative appendices		Total	
	No.	%	No.	%	No.	%	No.	%
No. examined	44	67.7	19	29.2	2	3.08	65	100
No. Parasitic Infected cases	14	21.5	0	0	2	3.08	16	24.6
<i>E.vermicularis</i>	0	0	0	0	1	1.54	1	1.54
<i>Schistosoma mansoni</i>	0	0	0	0	1	1.54	1	1.54
<i>G.duodenalis</i>	4	6.15	0	0	0	0	4	6.15
<i>E.histolytica</i>	4	6.15	0	0	0	0	4	6.15
<i>H.nana</i>	2	3.08	0	0	0	0	2	3.08
<i>H.diminuta</i>	1	1.54	0	0	0	0	1	1.54
<i>Cryptosporidium</i> spp.	2	3.08	0	0	0	0	2	3.08
<i>Blastocystis</i> spp.	1	1.54	0	0	0	0	1	1.54
P-value	0.021*							

* significant

Also in the current research, one case of *E. vermicularis* in the studied appendectomy specimens (1.5 %) was found. This is nearly similar to previous report by Ahmed *et al.* (2015) who mentioned that occurrence of pinworms with appendicitis ranged between 0.2 – 41 % worldwide. Other studies reported the frequency of *E. vermicularis* in the appendices was 0.6 – 3.8 % depending on the geographic area of the studied cases (Isik *et al.*, 2006; Sah and Bhadani, 2006; da Silva *et al.*, 2007; Chamisa, 2009).

A unique finding in this study is the presence of *E. vermicularis* adult in the submucosa of the appendix with the characteristic morphological features (lateral alae), not in the lumen as it is used to be. Babady *et al.* (2011) mentioned that rarely the *E. vermicularis* adult worms could become lodged in the intestinal mucosa and cause abscess.

Worth mentioning, that there was only mild eosinophilic infiltration around the *Enterobius* worm in submucosa and no acute inflammation was detected in the appendix specimen containing *Enterobius* worm. This is in agreement with majority of studies which reported low incidence of inflammation with enterobiasis of the appendix (Sinniah *et al.*, 1991; Wiebe, 1991; Panidis, 2011).

On the contrary, some studies found acute or chronic inflammation with pinworms in the lumen of the appendix (Dorfman *et al.*, 1995; Saxena *et al.*, 2001). It is justified to say that our results agreed with the reported cases worldwide that does not settle the controversy about the relation between *E. vermicularis* to appendicitis (Arca *et al.*, 2004; Panidis *et al.*, 2011).

On the other hand, some workers believed that pinworms manifest meticulously in the bowel according to some retrospective studies and it is the most common worm residing in the appendix, leading to pathological changes including inflammation, lymphoid hyperplasia, and subsequently complications like peritonitis and gangrene (da Silva *et al.*, 2007). Keeping in mind those intestinal

parasites may cause symptoms and signs similar to that of acute appendicitis; this may explain the normal histology of appendectomy specimen infected with *E. vermicularis* (Yabanoglu *et al.*, 2014).

Likewise, some minor degree of lymphoid hyperplasia may follow parasitic infection and not always cause symptoms. Moreover, pathogenesis of acute appendicitis may be due to either inflammation secondary to presence of parasites or obstruction of the lumen of the appendix by the parasite (Dorfman *et al.*, 1995).

In respect to *E. histolytica* in the appendix, it is quite rare and the exact incidence of this presentation is not well known (Sartorelli *et al.*, 2005). In this work four cases were found (6.15 %). The literature contains only few reports of amoebic appendicitis. When it occurs, it usually develops as an extension of caecal infection (Nadler *et al.*, 1990; Singh *et al.*, 2010). This infection should be considered in the treating physician's mind (Ito *et al.*, 2014). Aggregated trophozoites in the cecum and small intestine after appendectomy are likely and require treatment. Therefore, the use of antibiotic combined with anti-amoebic metronidazole post-operative therapy will reduce the incidence of complications especially in high risk regions such as Egypt.

Of interest, *H. nana* and *H. diminuta* ova were found in four faecalolith samples. This is a novel finding and their presence could be a coincidental finding with doubtful role in pathogenesis of appendicitis.

Moreover, *Blastocystis hominis*, infection was diagnosed in one case in this study. This is in accordance with Pasupati *et al.* (2008) who detected *B. hominis* in appendiceal luminal content with *Cryptosporidia* and *Microsporidia*. *Blastocystis* has been reported to invade the intestinal lamina propria in humans leading to inflammation (Al-Tawil *et al.*, 1994). In literature, only few cases were recorded in the appendix, therefore its role in appendicitis is questionable.

Giardia duodenalis (25 %) and *Cryptosporidium* (12.5 %) were also found in luminal content and in stool samples of parasitic infected appendectomized patients in this study.

Cryptosporidial enteritis may mimic acute appendicitis (Amer *et al.*, 2016). They reported a patient presenting with right iliac fossa pain and diarrhea and was operated for appendectomy. This appendix was grossly normal and stool samples revealed *Cryptosporidia* oocysts which was the cause of false appendicitis.

The question is whether presence of parasites in the specimens is incidental or a factor of inflammation. Aydin (2007) mentioned that an appendix hosting parasites may suffer from primary or secondary inflammation in the appendix as it has narrow lumen.

The surgeon, handling a suspected case of appendicitis, should be aware that parasites may cause colic similar to appendicitis. Therefore, eosinophilic count and rapid examination of stool samples should be done before surgery especially in children (Fleming *et al.*, 2015). This work sheds light on the differential diagnosis and treatment plans for future cases with abdominal colic similar to that of acute appendicitis.

The rate of parasitic infections detected in the study group arouse the possibility of some of them may have contracted infection in hospital. According to Lettau (1991), parasitic diseases are generally due to lack of cleanliness in the rooms, bathrooms and kitchens in the hospitals. Similarly, El-Sibaei (2003), reported that 8 % of patients developed diarrhea after admission to Ain-Shams pediatric hospital in Egypt and they were found to be positive for cryptosporidiosis.

Regarding eosinophilia detected in the studied patients it was distributed as follows: 6 % in negative patients for parasites, 15 % in cases with parasitic infection. Although most of protozoa infections seldom cause eosinophilia, the eosinophil count was significantly higher with protozoa infections in patients originally suffering from other disease. As regards enterobiasis, Seybolt *et al.* (2006) reported that it wouldn't cause eosinophilia unless the parasite invades tissues, as reported in our study.

The pathological examination of tissue sections in this study showed a range of findings. Acute inflammation was found in (67.69 %) of cases, with neutrophil infiltration or neutrophil and eosinophil infiltration. Two cases of them were diagnosed as periappendiceal abscesses, chronic inflammatory reaction with lymphoid hyperplasia was found in (29.23 %) which may initiate the start of inflammatory process. Two cases (3.08 %) showed no acute reaction and were considered as negative appendices.

In this study, immunochromographic test for *E. histolytica*, *G. duodenalis* and *C. parvum* copro-antigen detection was selected as a rapid test for stool examination and its performance was compared with microscopic examination as gold standard. This test is recommended for diagnosis of protozoa infection because of simplicity of use. There was no evidence of cross reactivity using the kit with other parasites identified in the stool specimens and can be used for screening purposes in large scale studies or outbreak investigations or as a possible alternative to stool examination

(Swierczewski *et al.*, 2012).

In this study, results of immunodiagnostic test for detection of copro-antigen show sensitivity of 100 % for *E. histolytica* and 100 % for *G. duodenalis*. *E. histolytica* sensitivity was found to be 54.5 % in the work of Gatti *et al.* (2002). Regarding cryptosporidial infection, the specificity of immunodiagnostic technique was 100 and it needs further evaluation since it failed to detect any positive cases. This may be due to low oocyst density. Also, may be due to genetic diversity of *Cryptosporidium* (Goni *et al.*, 2012). Salman (2014) found that direct fluorescent assay and modified Ziehl-Neelsen methods show high sensitivity and specificity in *Cryptosporidium* diagnosis than ELISA-copro antigen. On the other hand, Hawash (2014) recommended this test as rapid and highly sensitive and specific test.

This study has shed light on the problem of patients in surgical ward as being source for spread of parasitic diseases. Many types of food borne intestinal parasitic infections were found in these patients, and intestinal protozoa were of special importance.

Therefore, it is recommended to consider the possibility of parasitic infections in these patients in order to reduce the associated morbidity and suffering. Also detection of copro-antigens should be included in the laboratory diagnostic work-up for parasitic infections in these patients.

Finally, health education and implementation of infection control measures will always be advisable. It is recommended that routine histopathological examination of appendiceal specimens should be done for diagnosing unsuspected conditions such as parasitic infections that require further treatment.

In conclusion, parasitic infections were detected in 24.6 % of appendectomized patients at Tanta University Hospitals, Egypt. Most of parasitic infections were prevalent in patients belonging to the school age group. So, intestinal parasites may cause symptoms and signs similar to that of acute appendicitis. Therefore, careful attention to clinical history, stool examination and high eosinophilia may aid diagnosis and avoid unnecessary appendectomy. The presence of some ova and parasites in the narrow lumen of the appendix may be a co-factor in the development of appendicitis and this needs further studies. Rapid examination of stool samples and eosinophilic count should be done in case of appendiceal colic in endemic areas to diagnose intestinal parasites. The use of antibiotic combined with antiamoebic post-operative therapy will reduce the incidence of complications especially in high risk regions as Egypt. Immunodiagnostic test for the detection of copro-antigen was a successful rapid diagnostic technique, but still needs further evaluation in more cases of cryptosporidial infection, especially when stool samples were microscopically positive.

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