TITLE: STUDY OF PARASITIC ETIOLOGY OF CHOLECYSTITIS AND APPENDICITIS IN BASRA GOVERNORATE, IRAQ

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ABSTRACT

The current study dealt with the causes of appendicitis and cholecystitis, and it was found that the most parasitic causes are due to the *Isospora* and *Microsporidia*. The rate of infection of the appendix with *Isospora* was 54% and *Microsporidia* was 4% to have 58% of total percentage was also the *Isospora* which the main cause of cholecystitis with 52% infection rate, *Microsporidia* was 6% and *Cryptosporidium* was 4% to have 62% of total percentage and the parasites was mostly found in the area of the lamina properia and then the muscles.

Keywords: isospora microsporidia, appendicitis, cholecystitis

I. INTRODUCTION

Acute cholecystitis is an acute inflammatory condition and 95% of acute cholecystitis cases are caused by stone obstruction in the neck of the gallbladder or cystic duct [17]. The obstruction is caused by small, medium and large gallstones blocking the neck of the bile sac or Cystic duct obstruction of the cystic duct leads to excessive dilation of the bile sac and its pressure. This subject increases the pressure in addition combining the presence of hypercholesterolemic bile leads to an acute inflammatory response [29] and this may cause a secondary bacterial cholecystitis. Some studies have reported a 20% of bacterial infection.

*Isospora belli*

One cases of cholecystitis with this parasite were reported in immunocompromised persons, which was accompanied by watery diarrhea and severe pain. Histological tests showed the presence of C. belli infection [34,33,24,1].

*Cryptosporidium parvum*

Infection of the gallbladder with *C. parvum* is rare, as [4] the author reported a case of the parasite in an immunocompromised child who had inflammation of the bile sac, with severe watery diarrhea the parasite can also cause infections in immunocompetent persons [35].

Appendix

It is a finger-like tube with a blind tip located at the junction of the small intestine with the large intestine [15]. The average length of the human appendix is 9 cm but can range from 5 to 35 cm. The appendix is 6 mm thick, and more than 6 mm is considered a thickened or inflamed appendix.

Appendicitis is the most common cause of abdominal pain and one of the most common causes of emergency gastrointestinal surgery. Appendicitis may lead to abscess or peritonitis and may lead to death if left untreated [9].

The incidence of appendicitis is about 11 cases per 10,000 individuals per year [22,28]. Chronic appendicitis does not require urgent surgical operation and the incidence rate is 1.5% of all cases with acute appendicitis [16]. Appendicitis occurs for several reasons, including fecal obstruction, lymphatic enlargement or the entry of fruits and vegetables seeds into it and tumors of the large intestine [10]. Ingested foreign bodies may also cause appendicitis [32].
Parasites are among the rare causes of appendicitis. The most common parasites that cause appendicitis are *Enterobius vermicularis*, *Schistosoma spp*, *Taenia spp*, *Ascaris lumbricoides* and *Entamoeba spp*. [37, 2, 5, 8].

**Materials methods**

Tissue samples: tissue samples were taken from the hospital were approved after conducting the fundamental approval.

Appendix and gallbladder

Appendix and gallbladder samples were obtained from hospital, (50) samples were tacked for each, some of them were embedded with paraffin wax which have signs of malignancy while the other were preserved for a long time and ready to be disposed of which not have suspicious signs of malignancy.

Preserved samples were treated with formalin to prepare wax blocks according to Drury and ----1967

The samples were sectioned with a rotary microtome, with a thickness of 7 microns. Then it was stained with different stains like Eosin- Col, s haematoxylin, Periodic Acid Schiff reaction and Ziehl-Neelsen stain. Samples were examined using a Leica compound microscope, observations were recorded and photographed.

**II. RESULTS**

Appendix

The current study proved that the main causes of appendicitis in Iraq are parasitic infection, as the rate of infection is about 58%, and the infection was distributed between 54% with the *Isospora* and 4% with the *Microsporidia* and no infection with yeasts or *cryptosporidium* was recorded (Table 1).

Table (1) shows the rates of appendicitis with parasites

<table>
<thead>
<tr>
<th>Total Percentage</th>
<th>Infected Microsporidia percentage</th>
<th>Infected Isospora percentage</th>
<th>infected</th>
<th>Total Number</th>
<th>Stain Type</th>
</tr>
</thead>
<tbody>
<tr>
<td>%2</td>
<td>0%</td>
<td>2%</td>
<td>1</td>
<td>50</td>
<td>HE</td>
</tr>
<tr>
<td>%58</td>
<td>4%</td>
<td>54%</td>
<td>27</td>
<td>50</td>
<td>PAS</td>
</tr>
</tbody>
</table>

The current study showed that parasites have a great responsibility in appendicitis, diagnosed as *isospora* and *microsporidia* parasites, diagnosed in histological sections and the infection rate of the first parasite was the highest in appendicitis, as the parasite was present in most of the appendix samples, and the parasite was especially present in muscle areas. In varying stage of development, it is important to note that one sample was diagnosed using Eosin-Haematoxylin stain in the serosa layer Fig (1) and in muscle as shown in fig (2).

The study showed that the most species of parasites that infect the appendix are *isospora* of the parasite which present in the sporulation stage in the lamina properia fig (3), and large quantities of the parasite were found in other specimen fig (4).
Fig (1) A, B Infection of the appendix in the serosa layer with *microsporidia* arrow. The dark areas can be observed on both sides of the parasite indicating the location of the tube.

Fig (2) Appendix infected with *Microsporidia* stained with acid fast showing negative staining of parasite arrow.
Fig (3) Infection with *Isospora* in separate cells, showing the parasite's proliferation and the formation of spores arrow. Periodic acid Schiff stain.

Fig (4) An appendix infected with *Isospora* showing the release of large numbers of the parasite's spores arrow. Periodic acid Schiff stain.

**cholecystitis**

The current study proved that the main cause of cholecystitis is parasites, 6% of gallbladder infections are caused by *Microsporidium*, 4% are caused by *Cryptosporidium*, and 52% are caused by *Isospora*, with a 62% total percentage of infection - Table. (2).

Table (2) shows the parasites that cause cholecystitis and the percentages of infection

<table>
<thead>
<tr>
<th>Total Percentage</th>
<th>Microsporidia</th>
<th>Infected</th>
<th>Cryptosporidium</th>
<th>Infected</th>
<th>Isospora</th>
<th>Infected</th>
<th>Total Number</th>
<th>Stain Type</th>
</tr>
</thead>
<tbody>
<tr>
<td>%0</td>
<td>%0</td>
<td>0</td>
<td>%0</td>
<td>0</td>
<td>%0</td>
<td>0</td>
<td>50</td>
<td>HE</td>
</tr>
<tr>
<td>%62</td>
<td>%6</td>
<td>3</td>
<td>%4</td>
<td>2</td>
<td>%52</td>
<td>26</td>
<td>50</td>
<td>PAS</td>
</tr>
</tbody>
</table>

The study showed that most of the gallbladder infections were caused by the *Isospora*, the parasite presented in several region of the lamina properia, the parasite was appeared surrounded by a membranous vacuole (Parasitiforus vacuole). The sporont and sporocyste residium were also observed as shown in fig-(5) also the developed parasite spreads in the tissues of the gallbladder as shown in fig (6). While the *microsporidia* spreads in the lamina properia forming many vesicles which filled with parasites as shown in fig (7), a necrotic tissue wall of the gallbladder in the lamina propria was filled with *Microsporidia* parasite and the parasite’s sporulations observed as shown in Fig (8).
Fig (5) shows the infection with the parasite *Isospora* showing sporont (red arrow) and sporon (black arrow), parasitoforus vacuole (yellow arrow), and sporocyst residium (green arrow).

Fig (6) shows the parasite *Isospora*, showing the large spread of the parasite in the tissues of the gallbladder.
Fig (7) shows the severe spread of the parasite *Microsporidia* arrow causing extensive damage of the lamina propria PAS.

Fig (8) shows a necrotic wall of the gallbladder in the lamina propria filled with *Microsporidia* parasite, black arrow, and the parasite’s sporulations appeared arrow (PAS).

### III. DISCUSSION

*Cryptosporidium* and *Isospora belli* are opportunistic parasites that infect people with immunodeficiency syndrome [26] and *Cryptosporidium* is common in children under five years old, causing watery diarrhea [23].

*Isospora belli* spores multiply when they invade host cells by endodyogeny [18] and this process repeats continuously. Endodyogeny of the parasite may occurs in the villi cells of the small intestine and rarely in the lining cells of the intestine. It can also be found in the crypts and the lamina propria [30,3] are causing serious pathological changes, [12,13].

*Isospora belli* infection may be fatal or causes headache, diarrhea, fever, abdominal pain and vomiting, [36] in immunocompromised patient and rarely in immunocompetent, but the same researcher noticed an infection in women for four years and the infection was accompanied by severe watery diarrhea and confirmed that a transmission of infection outside the intestine is common. Studies by [22] indicated that the oocytes were released from the infected person's feces, and needed 24 hours to complete sporulation.
**Microsporidia** parasites are global in prevalence and infect vertebrates and invertebrates, while microsporidiosis in humans occurs in immunocompromised and immunocompetent persons as well, but the infection is more widespread and virulent in the immunocompromised [11]. **Microsporidia** parasites can also cause inflammation of the eyes, brain, liver and Chronic diarrhea and wasting, which is responsible for cases of severe exhaustion, which leads to a high proportion of deaths.

**Microsporidia** species were classified according to the area of infection. It was noted that the parasite *Enterocytozoon bieneusi* causes diarrhea, Weight loss, cholecystitis, bronchitis and asthma [20, 6, 7].

Several studies indicated that the rarity of appendicitis caused by parasitic causes [2] and food residues and seeds held the major responsibility for this [10] and many studies confirmed the presence of parasites or infection of the digestive tract with eggs of nematodes may cause severe infections of the appendix [27]. Here it cannot be denied that food entering into the appendix duct has a role, but in a small percentage, as only two cases were observed of the presence of food in the appendix in the current study out of fifty samples that were studied and were accompanied by severe necrosis of the appendix tissues.

A recent study presented at the US-Canadian Academy of Pathology meeting (2018) concluded that *Isospora belli* infection was 6.1% of gallbladder samples excised in children [31] and 9.7% of all pediatric patients Immunocompetent cholangiectomy [25].

The parasite may migrate from duodenum and passes through pancreatic duct that joins to gallbladder, forming the hepatopancreatic ampulla (known as the ampulla of Vater) – the gallbladder empties into the duodenum via the main duodenal papilla and this papilla is regulated by a muscular valve [14]. Thus, it is possible for the infection to be transmitted from the intestine to the bile vesicle through this channel, and this may explain what was observed by [19] of the infection of the pancreas with the *G. lamblia*, and since the *Giardia* parasite is unable to invade the tissues, it was not Through this channel, or the parasite is transmitted internally through succession of cell infection, reproduction, and infection of other cells, thus gradually moving to it. The access of parasites to the gallbladder may be through the common pancreatic duct, where there is a duct that allows the bile to exit the intestine from the bile vesicle to the intestine through the pancreas, and the arrival of parasites to the vesicle may be evidence of a liver injury.

The presence of the parasite in appendix may be due to a previous infection in the intestine that has spread to the inside of the appendix.

**REFERENCE**


