Academia Open Vol 9 No 2 (2024): December

Vol 9 No 2 (2024): December DOI: 10.21070/acopen.9.2024.9035 . Article type: (Medicine)

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Academia Open



By Universitas Muhammadiyah Sidoarjo

Vol 9 No 2 (2024): December DOI: 10.21070/acopen.9.2024.9035 . Article type: (Medicine)

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High Appendicitis Incidence in Young Adults Calls for Routine Histological Examination

Tingginya Insiden Usus Buntu pada Orang Dewasa Muda Memerlukan Pemeriksaan Histologi Rutin

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Abstract

This study explores the histopathological changes associated with appendicitis in appendicectomy specimens from Nasiriyah General Hospital. Employing a retrospective analysis on twenty specimens, it aims to underscore the importance of routine histological examinations. Findings reveal that all specimens were afflicted with appendicitis, featuring notable occurrences of mucosal glandular hyperplasia (43%), mucosal and submucosal layer necrosis (42%), and severe inflammatory cell infiltration (71%). These results highlight the necessity of comprehensive histological assessments in appendicectomy specimens to enhance diagnostic accuracy and improve patient management, particularly among adolescents and young adults.

Highlights:

- All specimens confirmed appendicitis with significant alterations.
- Routine histological exams critical for accurate diagnosis.
- Enhanced scrutiny improves adolescent, young adult treatment.

Keywords: Appendicitis, Histopathological Changes, Appendicectomy, Histological Examination, Adolescents

Published date: 2024-05-28 00:00:00

Vol 9 No 2 (2024): December DOI: 10.21070/acopen.9.2024.9035 . Article type: (Medicine)

Introduction

The appendix is an organ that is part of the digestive system and is typically thought of as a useless organ in humans. The possibility that this organ contributes to immunity is now well acknowledged [1]. This organ is located in the lower right quadrant of the abdomen. Even though this organ is not very important, illnesses including hepatitis, pneumonitis, cholecystitis, tonsillitis, and encephalitis can affect it [2]. Because the appendix has an end and is difficult to obtain normally, it is known to be a site of both acute and chronic infections. They are always prone to inflammation, are congested, and their contents do not renew quickly [3].

One of the most prevalent inflammatory gastrointestinal disorders is acute appendicitis [4]. Acute appendicitis affects roughly 7% of people on the planet at some point in their lives. Every year, approximately 250,000 instances of appendicitis are reported in America [5], [6]. An estimated 70,000 cases of infection occur in the UK annually [7]. The symptoms of appendicitis usually appear in the later stages of and rarely occur before the age of two years [8]. Early life and teenage years. The age range of 10 to 30 years old has a higher incidence of appendicitis [9].

Moreover, between 20 and 40 percent of appendices are performed passively [10]. Acute appendicitis does not have a definite age range associated with it, and it may be caused by parasites [11]. The majority of instances of acute appendicitis are caused by swelling of the lymphoid-mucous tissue from bacterial, viral, or parasitic infections—such as those caused by the pinworm parasite—or obstruction of the appendix's lumen as a result of hardened stool [12].

Surgery is often required to treat the common condition of appendicitis in order to avoid complications. In the US, appendicitis affects about 250,000 people a year. Men are more likely than women to experience this inflammation by 1.4–1.3 times, with 80% of instances occurring in those under 45 [13].

The intestines and appendix open up naturally, but obstructions such as hardened excrement, enlarged tissue, parasitic worms, or germs can form and cause appendicitis. Even while it can happen at any age, appendicitis is most common in those between the ages of 30 and 10 [14]. In children younger than two years old, it is uncommon [15]. There have been reports of appendicitis everywhere. It was expected to affect 250,000 cases annually in the USA [16].

Iraq has no reliable records regarding appendicitis. There were 530 general cases registered annually at Nasiriya General Hospital and 450 cases annually at Al Habobi Hospital. Numerous microorganisms, including bacteria, viruses, and parasites, can infect the appendix. This organ needs to be removed surgically in order to safeguard the body from harm and preserve its health.

Methods

A. Study Area

The study was carried out in 2022/2023 in Nasiriyah, in the Thi Qar Governorate in Iraq. This governorate is bordered to the south by Basra, to the east by Maysan, to the west by Muthanna, and to the north by Wasit. The study focused on the prevalence of appendicitis in this governorate. Patients having this infection provided samples for the study. The long duration of the study, which ran from October 30, 2022, to April 30, 2023, was undertaken in order to identify the primary causes of appendicitis. The site and the structure of the histology. Being aware of its signs and determining its diagnosis.

B. Sample Collection

Ten appendicitis samples were taken, with a maximum collecting duration of ten days, and fixed in 10% formalin. When patients with appendicitis underwent procedures to remove their appendices, they were retrieved from Nasiriyah General Hospital. They were taken straight to the Al-Hussein Teaching Hospital laboratory for a bacteriological analysis. Where The process of gathering samples began with obtaining a letter from the Thi Qar Health Department authorising a task for the purpose of gathering appendicitis samples. There were various sorts of appendices, some thick and some thin. At Thi Qar Governorate's Al-Hussein Teaching Hospital, a number of preparatory actions were taken. In the manner described below: [17]

- 1. Appendix 8.5×5 cm two pieces taken.
- 2. Appendix 5×1 cm two pieces taken.
- 3. Appendix 8×1 cm around fatty tissue two pieces taken.
- 4. Appendix 5×1 cm around by fatty tissue two pieces taken.

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- 5. Appendix 7.5×1.5 cm fatty tissue two pieces taken.
- 6. Appendix 6.5×0.5 cm fatty tissue two pieces taken.
- 7. Appendix 6.5×1 cm two pieces taken.
- 8. Appendix 7×1 cm around fatty tissue two pieces taken (thick wall).
- 9. Appendix 5×0.5 cm two pieces taken.
- 10. Appendix 10×1 cm around fatty tissue two pieces taken.

C. Histological Cutting

First, the doctor removes the sample from the patient After that we do a macroscopically examined for the sample possibility of ulceration, damage to the outer wall of the sample, inflammation or swelling $\[\]$ The sample is then passed through the steps below.

It maintains the shape of the tissue from deterioration by preventing Autolysis and rotting process by small organisms by using fixatives such as 10% formalin

[10%formalin it's Consists of 10% formalin and 90% tap water]

, gives rigidity to the tissue, and Facilitates the staining process later.

Tap water is used instead of distilled water in formalin because tap water prevents swelling of the sample due to it containing a percentage of salts.

Distilled water is highly osmotic(hypotonic), and this causes its penetration into the tissue and causes swelling in the sample (distilled water free of salts will move to areas of high concentration of salts, and it penetrates the tissue, causing swelling).

1. Fixation

It is the first step and includes three basic functions:

2. Washing

Using tap water for a long (6-24) hours to remove fixative from the tissue ,and It usually depends on the type of fixative.

3. Dehydration

Common dehydration fluids are Ethanol, commercial industrial methylated spirit, methanol, propanol (isopropyl alcohol), acetone. The dehydration process is the removal of water using dehydration liquids and in this samples alcohol was used and alcohol is introduced into the tissue in ascending concentrations are 70%,80%,90%,95%, 100%, 100%, 100% thus, the water was eliminated and the alcohol became inside the tissue.

4. Clearing

Substances are used to remove alcohol and are removed by wax these substances are clearing agent and there are common types of clearing agent are Xylene (xylole), toluene chloroform ,benzene ,cedar wood oil.

In these samples, xylol was used and it is used with three changes in each change of one hour, so that the xylol became inside the tissue.

5. Infiltration (Impregnation)

Paraffin wax is used where it is melted at a temperature of 60C, and thus the paraffin wax becomes liquid and the fabric is placed inside it using three changes, and thus the paraffin wax becomes inside the fabric and the paraffin wax gives a support to the fabric, and then the fabric and its cavities become hardened and protected with wax.

6. Embedding

In this step, paraffin wax is also used, but it does not contain residues of xylene, a mold is used, paraffin wax is poured into it continuously, and then the sample is placed inside the mold containing paraffin wax.

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There is a phenomenon in paraffin wax that is the phenomenon of surface tension and to prevent this phenomenon from happening, the wax is blown into the mold . Another phenomenon that occurs in paraffin wax is the phenomenon of crystallization, and the reason for this phenomenon is that the wax cools slowly, which are bubbles that are inside the wax, and these bubbles weaken the integrity of the block, and it is possible that during cutting they are crushed by Microtome knives.to prevent this phenomenon, The Block is placed in the refrigerator overnight and as a result, rapid cooling occurs, which prevents the crystallization phenomeno. The purpose of this step Is to support the sample from the outside.

7. Trimming Process

After preparing the wax molds, it is advisable to trim them with a sharp blade so that the sample is in a suitable position For cutting so that its edges become parallel and can apply to the edge of the microtome knife.

8. Cutting

After completing the fixation steps, the process of cutting the tissue (sample) using the microtome device: The block that contains the sample is cut with a microtome device with a thickness of (3_5)µm, as soon as a slice containing tissue appears, it is taken directly and placed in the water bath device at a temperature of (40_45)C[in order to ensure the expansion of the sample. After that, a new slide is used, and using Mayer's egg albumen [It's consist of egg albumen,glycerin and crystals of thymol to prevent rotting] it works as a glue to stick the sample slic on the slide.9_StainingAfter that, the Staining process begins, but first the wax in the slice (tissue) must be removed by dewaxing process, using the oven at 60 degrees for about half an hour. Immediately after it is extracted from the oven, it is placed in three variables of xylene (xylole), and the sample is left for a quarter of an hour in each variable so the wax was removed. Since the medium that carries dyes inside the tissue is water and because of the removal of paraffin wax by xylene (xylole), the tissue contains xylene (xylole), so it must be expelled from the tissue and replaced with water, but the question is how is this done? The xylene (xylole) is removed by using descending concentrations of alcohol (100_70). After the last concentration, the tissue is taken and placed in water, after which the dyeing process becomes possible. The Staining process is done by using two dyes:

a. The first is hemotoxylin; It is a basic stain that is attracted to acidic structures, and since the highly acidic structure is the nucleus, it dyes the nucleus in a blue or violet colour. It also stains any acidic structure such as protein, so the dye must be shortened by differentiation, as the dye sticks to high-acid structures only, and an alcoholic acid 1% (consisting of 99 alcohols and 1 HCL compound) is used. The slide was placed four times with alcohol acid 1% and then washed with water, so the dye was confined to the nucleus only. 2_ The second dye is eosin: it is an acidic dye that is attracted to the base compounds, where it stains the cytoplasm, because its compounds are alkaline and dye pink. Thus, the cell became dyed in two colors, the nucleus is blue or violet, and the cytoplasm is pink.

The water inside the tissue that served as a medium for dye transfer is no longer needed for its presence, so it is expelled from the tissue by dehydration through progressive alcohol concentrations (70_80_90_95_100). It should be noted that the presence of alcohol inside the tissue causes cloudiness, and therefore it must be disposed of, and this is done by using xylene (xylole), so it will filter the tissue as well. At the end of the process, a cover slip is attached to the sample on the slide with a material called DPX. Thus, it becomes a tissue that can be preserved for decades.

Results and Discussion

A. Results

Histopathological examination was conducted for 20 appendices. There are histopathological changes was recorded in all excised appendices. These were stained with hematoxylin and eosin and imaged with differentmagnification power.

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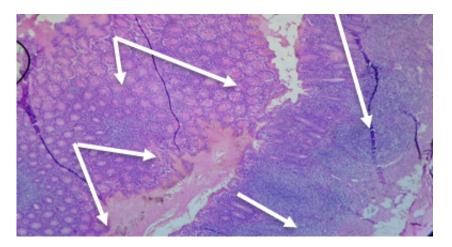


Figure 1. a. mucosal glandular hyperplasia, b. cavitation of lymphatic follicles, c. mucosal ulcerate and necrosis, d. blood sinusoids (40x) (H&E).

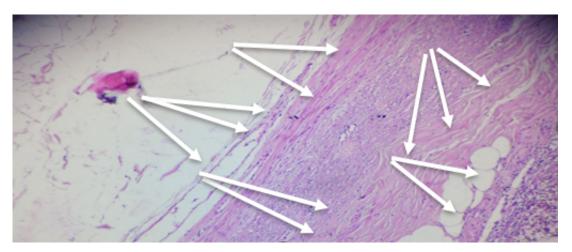


Figure 2. a. erosion of mucosal layer, b. necrosis of mucosal layer, c. necrosis of submucosal layer, d. oedema, e. mild inflammatory cell infiltrations (10x) (H&E).

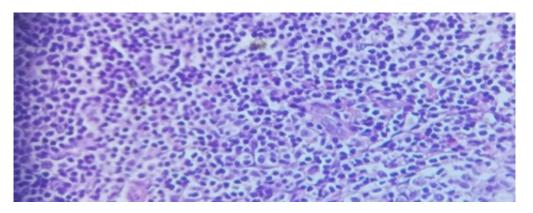


Figure 3. Severe inflammatory cell infiltrations (100x) (H&E)

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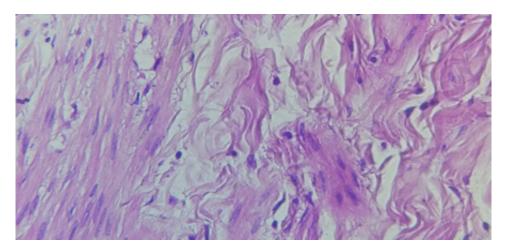


Figure 4. Liquefactive necrosis (400x) (H&E).

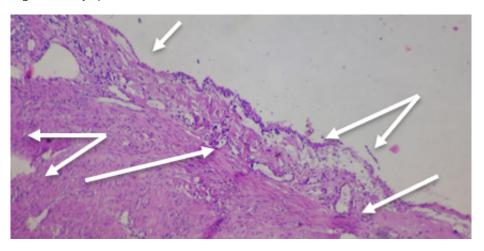


Figure 5. Mucosa atrophy, b. mucosa erosion, c. mild inflammatory cellular infiltrations, d. necrisos of mucosal layer, e. fibroblastic proliferation (100x) (H&E).

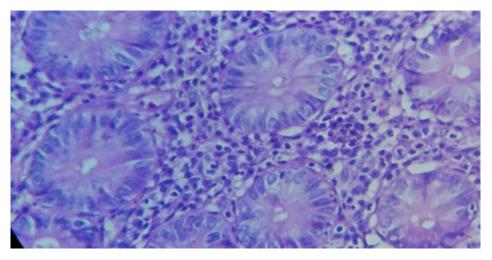


Figure 6. Expansion and hypertrophy of lymphatic ducts and severe inflammatory cell infiltrations (400x) (H&E).

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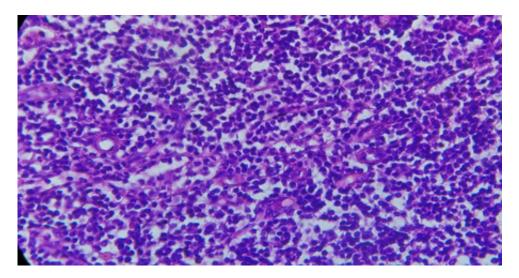


Figure 7. Severe inflammatory cell infiltrations (400x) (H&E).

B. Discussion

Appendicitis is a condition that affects people of all ages and genders, but it is more common in those between the ages of 10 and 30 [18]. It is also very common and can have serious repercussions. Numerous investigations and scientific studies have verified the involvement of bacterial, viral, and parasite pathogens in the development of chronic inflammation. The appendix is blocked at the end, which prevents its contents from regenerating fast and causes it to continuously become a source of both acute and chronic inflammations [19]. which advance in the direction of the bodily cavity [20]. Primary animals and worms reproduce quickly, which poses a serious health risk in developing nations, particularly in South and Southeast Asia. These regions have high population densities, heat waves, monsoons, poor public health and education systems, and biological and mechanical resistance to parasitic growth phases in the environment, all of which contribute to the survival and spread of primary animals and worms in these environments. Parasitic infections have a detrimental effect on growth, health, and nutrition because they can cause major pathological consequences, obstruct important organs, or infiltrate tissues, necessitating surgery in cases like appendicitis [21], as well as conduct a culinary competition [22]. The contribution of parasite causes to inflammation is a topic of discussion and disagreement. Due to the parasite's low prevalence in the inflammatory appendages—as demonstrated by observation of tapeworm body fragments in the appendix cavity—some researchers have demonstrated the existence of certain intestinal parasites in the inflamed appendages without establishing their pathological role in appendicitis [23]. However, other researchers that verified the parasite's pathogenic role-particularly in histological analyses of infected appendages-documented similar results with some relevance., which counted parasites as pathological organisms closely related to the occurrence of inflammation [24]. Confirm [25]. The relationship of pinworms and their association with appendicitis, indicating that the percentage of this relationship varies between 0.2-41.8% worldwide. As for the relationship between the total infection of appendages with parasites and the sex of the infected, the current study showed that there is no significant difference in infection between females and males (47% for females versus 61.7% for males), this may be due to the presence of the same opportunity to cause infection in both sexes, such as the same food, presence in the same environment and drinking the same contaminated water...Etc. This is consistent with the findings of [26] (3.3% of females and 4% of males were injured) and the severity [27], where the injury was 41.9% of females and 58% of males. From this, it was found that there is no influence of the sex factor in causing parasitic infection of appendages, which confirms that gender itself is a non-essential factor in determining the susceptibility and predisposition of an individual to infection with such intestinal parasites. In the current study, no infection of the appendix with the bovine tapeworm Taenia saginata and the fish tapeworm Diphyllobothrium latum was recorded, due to the lack of common consumption of raw or poorly cooked meat in Iraq. The chance of infection of the appendix with schistosomiasis worms or their eggs remains low due to the fact that the rate of infection with the blood splinter is low in the city of Baquba, however, the infection of the appendix with these worms was recorded by 44% [28] at Al-Sabah Hospital in Kuwait .The results of the current study showed the presence of common parasitic infections in one appendix by 3.1% in which the dysentery amoeba parasite shared with other intestinal parasites. Co - infection with parasites in the appendix has been recorded by [29]. Co-infection was noted by [30] between pinworm and whipworm, pinworm and nematode, pinworm and dysentery amoeba, and pinworm and Giardia lamblia. The quality of the parasites reported in each study and their respective infection rates are the primary causes of this little disparity. The recent investigation shown that intestinal worms and primary animals, two types of parasites, infected appendages. Amoeba dysentery was more common than other parasites; at 53.1%, it had the highest infection rate ever seen, compared to [31] 0.97% at Mosul Teaching Hospital in Nineveh [32].

Additionally, it is 0.21% in Mosul, [33] in the Baghdad Teaching Hospital, 0.7%, and (28%), in the province of Najaf. Both positively and negatively, having parasites in the colon lowers immune system effectiveness [34], which weakens the host's defences against bacterial and viral illnesses and increases susceptibility to other pathological

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infections [35].

Conclusions

The present study revealed that adolescents and young adults in the Thi Qar Governorate of Iraq exhibit a high incidence of appendicitis, with histopathological analysis of 20 appendectomy specimens indicating significant mucosal and submucosal layer necrosis, mucosal glandular hyperplasia, and severe inflammatory cell infiltration. The routine histological examination of appendectomy specimens is strongly supported by these findings to prevent missing clinically significant tissue alterations. Furthermore, the study's implications emphasize the critical need for timely diagnosis and intervention in appendicitis cases to mitigate potential complications. Future research should focus on identifying the underlying causes of appendicitis, including the potential role of parasitic infections, and evaluating preventive measures to reduce the incidence and severity of this condition in vulnerable populations.

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