Original Article

Bacteriological Study of Acute Appendecitis

Maad M. Abdul Rahman FRCS (Ed)

Key words: Bacteria, Acute appendicitis

Abstract

Background: Acute Appendicitis is the most common acute surgical condition of the abdomen in surgical practice. Regarding the aetiology a number of theories have been proposed; these include abnormalitis in the diet, genetic factors and a variety of infectious agents. The latter is our concern in this study.

Objectives: To determine the rate of different classes and types of bacterial isolates by type of the appendix and the association of age and gender with the risk of having a normal looking appendix at operation.

Methods: A cross sectional study was performed at Baghdad teaching Hospital and AI-Kindy Hospital from January 1999 to December 2001 inclusive and a series of 120 patients is collected randomly with clinical features of acute appendicitis had appendectomy, swabs were taken from the bases of the removed appendices and sent immediately for bacteriological study.

Results: The rate of normal looking appendix was 19.2 %, and the acutely inflamed appendix (65 %). In general Gram negative bacterial isolates were more frequently isolated (84.2%) from cultured specimens than Gram positive types (16.7%). Patients with gangrenous/ perforated appendix had a significantly higher prevalence of both Gram negative and positive bacterial isolates (100% and 31.6% respectively) than normal looking and acutely inflamed appendices. Pure isolates were less frequently seen in patients with gangrenous / perforated appendix (15.8 %) as compared to mixed cultures (84.2 %). The most frequently isolated bacteria from cultured specimens were E.coli (49.2%), followed by Bacteroids (25%) and Klebsiella (15.8%). Gram positive bacteria on the other hand were less frequently isolated.

Conclusions: There is high incidence of normal looking removed appendix in the female. The bacterial flora of the appendix ensures rapid secondary invasion of the damaged tissue with the host bacteria among which Escherichia coli is the most prominent. The result of culture is affected by the pathological state of appendix. Gram-negative bacteria were predominant in both pure and mixed growth.

Introduction:

Acute Appendicitis is the most common acute surgical condition of the abdomen in surgical practice[1], the disease occurs in all ages but it is most frequent in the second and third decades of life[2]. The heavy lymphoid aggregation in the submucosa of the appendix in the early teens of life reflects the high incidence of the disease in this age

Prof. Maad M. Abdul Rahman, FRCS (Ed), Chairman Surgical Scientific Council, Iraqi Commission of Medical Specialization, P.O. Box 5116, Bab Al-Moudam, Baghdad - Iraq.

group[3]. Regarding the aetiology a number of theories have been proposed; these include abnormalitis in the diet[4] genetic factors[5] and a variety of infectious agents[6]. The latter is our concern in this study. The appendix and colon are heavily swarmed by pathogenic microorganism[7]. The effects of the intestinal bacteria on their host are complex, some being definitely beneficial and others possibly harmful[8]. Haematogenous spread of bacteria-particularly streptococci-may also occur[9].

Patients and Methods: A cross sectional study was performed at Baghdad teaching Hospital and Al-Kindy Hospital and a series of 120 patients is collected randomly. There were 61 male and 59 female patients, of different age groups range between 4-50 years and from different social classes. They presented to the surgical emergency department with symptoms suggesting acute appendicitis. The duration of the study is from January 1999 to December 2001. When a diagnosis of acute appendicitis was made, appendectomy was urgently performed. Neither pre operative nor pre operative antibiotics were used. At operation, swabs were taken from the base of the appendix after transaction. The swabs were sent within 30 minutes to the bacteriological laboratory to be cultured. The inoculated plates were kept at 37C° and examined after 24 and 48 hours. Plates that showed no growth were further incubated up to 96 hours before and examined after 24 and 48 hours. Plates that showed no growth were further incubated up to 96 hours before discarded as negative.

Statistical Analysis: The data were translated into codes using a special designed coding sheet and then entered into a computerized data base structure. Statistical analyses were done using SPSS (statistical package for social sciences) computer software after having appropriate statistical consultation. Frequency distribution for selected variables was done first. The statistical significance of association between two categorical variables was assessed by Chi-square test. P value less than 0.05 level of significance was considered statistically significant. The risk of having a certain outcome (like mixed culture) in the presence of a certain criteria (like having a gangrenous / perforated appendix) compared to its absence (having any of the other 2 types of appendix) was assessed by Odd's ratio (which is the best estimate of relative risk in a cross sectional design).

Results: The results presented in this study were based on the analysis of 120 patients with a provisional diagnosis of acute appendicitis. Male to female ratio was 1:1. About half (52.5%) of the study sample were 11-30 years old, a quarter (25%) were less than 11 years of age and 22.5% were older than 30 years, table 1.

Table 1: Distribution of the study sample by age and gender.

Table 1: Distribution of the study sample by age and gender.

	Male	Female	Total	
	N	N	N	%
<11	19	11	30	25.0
11 - 30	27	36	63	52.5
30+	15	12	27	22.5
Total	**61**	**59**	**120**	**100**

About two thirds (65%) of surgically removed appendices showed signs of acute inflammation, 15.8% were gangrenous and/or perforated, only 19.2% were normal looking appendix. The prevalence of gangrenous/ perforated appendix was significantly perforated appendix was significantly higher among males (23%) than females (8.5%). The prevalence of normal looking appendix was significantly higher among females (30.5%) than males (8.2%), Table 2.

Table 2: Relative frequency of 3 types of

Table 2: Relative frequency of 3 types of appendix by gender.

Type of appendix	Male N	Male %	Female N	Female %	Total N	Total %	P(χ^2)
Acutely inflamed	42	68.9	36	61.0	30	25.0	0.37[NS]
gangrenous/Perforated	14	23.0	2	8.5	63	52.5	0.03
Normal	5	8.2	18	30.5	27	22.5	0.002
Total	**61**	**100**	**59**	**100**	**120**	**100**	

The operative findings in patients with normal looking appendix shown in figure 1.

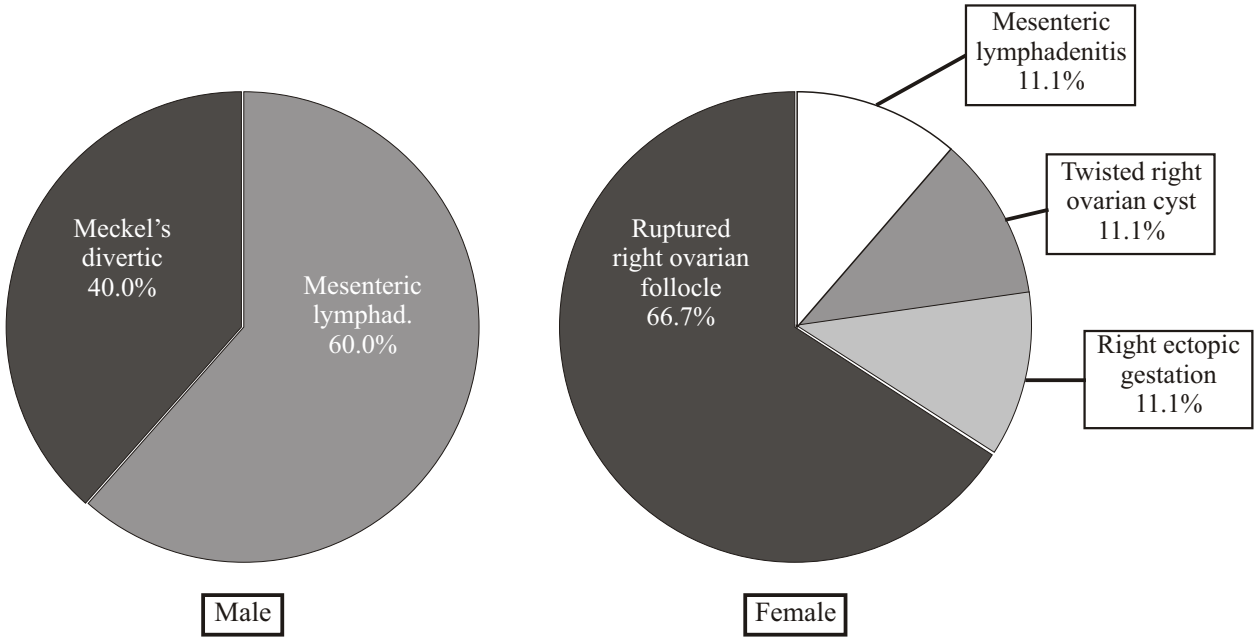

Figure 1: Pie chart showing the relative frequency of operative findings of normal looking appendix in males compared to females.

In general bacterial isolates were more common among patients with gangrenous/ perforated appendix, while no important differences in prevalence of different microorganisms were observed between patients with acutely inflamed appendice an those with normal looking appendix. The most frequently isolated bacteria from cultured specimens was E.coli (49.2%), followed by Bacteroids (25%) and Klebsiella (15.8%), all these were Gram negative bacteria. Gram positive bacteria on the other hand were less frequently isolated, table 3.

Aerobic bacterial isolates were more frequently isolated (76.7%) from cultured specimens than anaerobic types (31.7%), table 4.

Table 3: Difference in prevalence rate of different bacterial isolated between three types of appendix.

Bacterial isolates	Type of appendix: Acutely Inflamed N (n=78)	%	gengrenous/ perforated N (n=19)	%	Normal N (n=23)	%	Total N (n=120)	%	P value
Gram negative									
E. Coli	35	44.9	17	89.5	7	30.4	59	49.2	<0.001
Bacteroids	12	15.4	12	63.2	6	26.1	30	25	<0.001
Klebsiella	11	14.1	6	31.6	2	8.7	19	15.8	0.1[NS]
Proteus spp	2	2.6	7	36.8	3	13	12	10	<0.001
Ps. aerogenosa	5	6.4	1	5.3	2	8.7	8	6.7	0.89[NS]
Gram positive									
Anaerobic strep.	6	7.7	4	21.1	0	0	10	8.3	0.04
Clostridia spp.	1	1.3	2	10.5	0	0	3	2.5	0.04
Staph. Pyogens	1	1.3	3	15.8	2	8.7	6	5	0.02
Staph. Epidermidis	1	1.3	0	0	0	0	0	0.8	0.76[NS]
Aerobic strep.	1	1.3	2	10.5	0	0	3	2.5	0.04
Bacillus spp.	2	2.6	2	10.5	0	0	4	3.3	0.14[NS]

Table 4: Difference in prevalence rate of anaerobic bacterial isolated between three types of appendix.

Bacterial isolates	Type of appendix: Acutely Inflamed N (n=78)	%	gengrenous/ perforated N (n=19)	%	Normal N (n=23)	%	Total N (n=120)	%	P (χ^2)
Oxygen requirement									
Aerobic	57	73.1	19	100	16	69.6	92	76.7	0.03
Anaerobic	19	24.4	13	68.4	6	26.1	38	31.7	0.001
Negative culture	3	3.8	0	0	3	13	6	5	

Gram negative bacterial isolates were more frequently isolated (84.2%) from cultured specimens than Gram positive types (16.7%), table 5.

Table 5: Difference in prevalence rate of Gram positive and Gram negative bacterial isolated between three types of appendix.

	Type of appendix								
	Acutely Inflamed		gengrenous/ perforated		Normal		Total		
Bacterial isolates	N (n=78)	%	N (n=19)	%	N (n=23)	%	N (n=120)	%	P (χ^2)
Gram stain									
Gram negative	64	82.1	19	100	18	78.3	101	84.2	0.11[NS]
Gram positive	12	15.4	6	31.6	2	8.7	20	16.7	0.002
Negative culture	3	3.8	0	0	3	13	6	5	

The median number of different classes of bacterial isolates was significantly higher among patients with gangrenous/perforated appendix than those with acutely inflamed or normal looking appendix, i.e patients with gangrenous/perforated appendix not only had a higher prevalence rate of different bacterial types and classes but the number of isolated bacterial types in each class was also significantly higher than in patients with acutely inflamed or normal looking bacteria, table 6.

Table 6: Difference in median number of bacterial isolates between three types of appendix.

	Type of appendix				
	Acutely Inflamed	gengrenous/ perforated	Normal	Total	P value
1. Number of bacterial isolates					<0.001
Range	0 -2	1-6	0 -2	0-6	
Median	1	3	1	1	
N	78	19	23	120	
2. Number of aerobic bacterial isolates					<0.001
Range	0 -2	1 -3	0 -1	0 -3	
Median	1	2	1	1	
N	78	19	23	120	
3. Number of anaerobic bacterial isolates					<0.001
Range	0 -1	0 -3	0 -1	0 -3	
Median	0	1	0	0	
N	78	19	23	120	
4. Number of Gram negative bacterial isolates					<0.001
Range	0 -2	1 -4	0 -2	0 -4	
Median	1	2	1	1	
N	78	19	23	120	
5. Number of Gram positive bacterial isolates					<0.001
Range	0 -1	0 -3	0 -1	0 -3	
Median	0	0	0	0	
N	78	19	23	120	

In general the most common type of culture results was pure bacterial isolates (78.3%), followed by mixed type (16.7%). Negative cultures was reported in 5% of the study sample. Pure isolates showed a significantly lower rate among patients with gangrenous/perforated appendix (15.8%) compared to those with normal looking appendix and those with those with acutely inflamed or normal looking appendix. Negative cultures acutely inflamed appendix (78.3% and 93.6%). Mixed cultures showed a significantly higher rate among patients with gangrenous/ perforated appendix (84.2%) compared to those with normal looking appendix and those with acutely inflamed appendix (8.7% and 2.6%). The risk of having mixed growth in patients with gangrenous/perforated appendix was 129 times higher than observed however were not significant statistically. The risk of having negative showed a higher rate among patients with normal looking appendix (13%) compared to those with gangrenous/perforated appendix and those with acutely inflamed appendix (0% and 3.8%). The differences bacterial culture in patients with normal looking appendix was 4.7 times higher than those with gangrenous/perforated or acutely inflamed appendix, table 7 and figure 2.

Table 7: Difference in relative frequency of pure, mixed and negative culture between three types of appendix.

	Type of culture							
	Negative		Pure		Mixed		Total	
Type of appendix	N	%	N	%	N	%	N	%
Acutely inflamed	3	3.8	73	93.6	2	2.6	78	100
Gengrenous /perforated	0	0	3	15.8	16	84.2	19	100
Normal	3	13	18	78.3	2	8.7	23	100
$P(\chi^2)$	0.15[NS]		<0.001		<0.001			
Total	6	5	94	78.3	20	16.7	120	100

OR for having mixed culture in gangrenous / inflamed appendix = 129
OR for having negative culture in normal looking appendix = 47

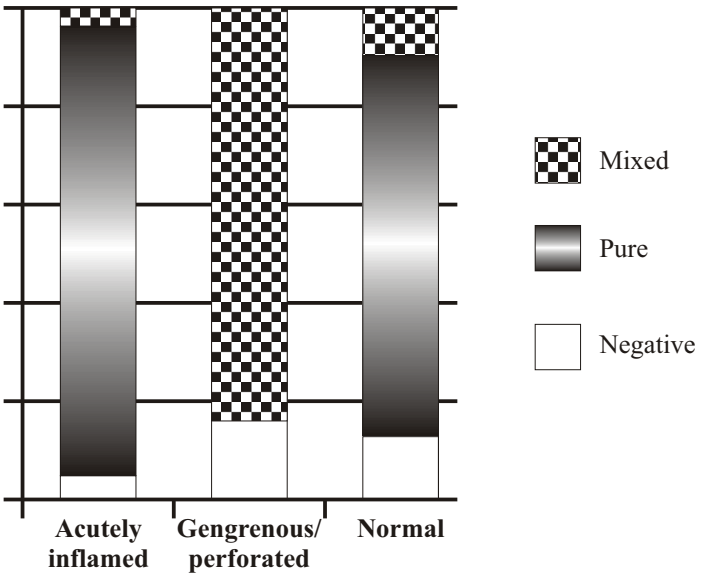

Figure 2: Stacked bar chart showing the relative frequency of different types of culture results by type of appendix.

Discussion: Operations on the gastro intestinal tract usually carry a potentially high risk of sepsis. The highest prevalence of acute appendicitis was in the (11-30 year) age group (52.5 %) with a female predominance, 36 (57.14 %). This may be explained by the presence of huge amount of lymphoid follicles in the appendix in this age group. While in the other age groups, < 11 & > or = 30, the males were more frequently involved, 63.33% & 55.56 % respectively. Al-Janabi I.M and Al-Tikriti F.N[10] Abdul-Hamid N.B and Al-Faddagh Z.[11] found the same age prevalence (11-30 year) and gender ratio. Female represent 49.2 % of the patients and male represent 50.8 %. Previous reports showed nearly similar gender ratio[10,11]. Per jess et al. 1981[12] demonstrated that 46.5% of patients were females and Boerem et al. 1981[13] recorded that females represent 45.8% and the commonest age group between (15-35 year). Acute appendicitis is essentially a clinical diagnosis, laboratory investigations have a supportive part to play[14]. The early surgical interference actually reduces the mortality & morbidity rate. But the problem of negative laparotomies will increase the risk and invite complications. The former opinion versus leaving a patient with suspected acute appendicitis would invite appendicular perforation & its sequel. These two opposing and conflicting observations will naturally conclude that early operation despite the negative finding would save a lot of lives and prevent the complications of perforation and its sequel[13]. In this study 23 patients (19.2%) had normal appendix. Fred Bongard et al. (1985) reported that a negative laparotomy rate of (20%) in any institution is generally considered appropriate[15]. Lewis et al 1975[16] and Adiss et al 1990[17] recorded relatively lower rates of 15% and 17% respectively. The complex bacterial flora of this part of the intestine i.e. the appendix ensures a rapid secondary invasion of the damaged tissue with the host bacteria (auto-infection)[18]. In the present study, the Escherichia coli was the most predominant aerobic microorganism (49.2%) followed by Bacteroids (25%) and Klebsiella (15.8%). Gram positive bacteria on the other hand were less frequently isolated, among them, the anaerobic Streptococci were the most frequent (8.3%) followed by Staph. Pyogenes (5%) and Bacillus spp (3.3%). This is consistent with the results of other workers. Veselyi-SV 1999[19], Bodnar-BM 1997[20] Ronchetto-F et al. 1991[21] who found that Escherichia coli represent (43%), (45%) and (48.4%) respectively. This is expected since it is the predominant aerobic bacteria in the gut[22] While Abdul-Hamid N.B et al found relatively lower rates (E. coli = 39.1 %, Bacetroids spp. = 18.3 % and Klebsiella = 10.8%)[11]. The rather low rate of isolation of anaerobic bacteria in this study is not so different from that obtained by previous workers. Veselyi-SV 1999[19], Bodnar-BM 1997[20] and Ronchetto-F et al. 1991[21] found that Bacteroid species is the most predominant anaerobic microorganism (25.5%), (22%) and (20.8%) respectively. In general, bacterial isolates were more common among patients with gangrenous/ perforated appendix group, where the highest prevalence of E. coli, Bacteroids, and Proteus spp was found to be statistically significant [P value < 0.001 each]. This finding was consistent with that of Rautio M et al. who found that the specimens from patients with gangrenous appendices yielded significantly higher numbers of anaerobic isolates per specimens than did specimens from patients with healthy appendices [11.7% vs 7.7% P < 0.01][23]. Ronchetto F. et al. found that the E. coli and Bacteroids spp represented (27.7%), (20.8%) respectively from the isolates of gangrenous and perforated appendices[21]. While in our series, the E. coli, Bacteroids, Proteus spp and Klebsiella represented a much higher prevalence among the gangrenous and perforated appendices group (89.5%, 63.2%, 36.8% and 31.6% respectively). The Gram positive anaerobic streptoccoci predominated the others in the acutely inflamed and the gangrenous/perforated groups (7.7%) and (21.1%) respectively. The prevalence rate of the Gram positive isolates was significant in the gangrenous/perforated group [P value = 0.002]. While Ronchetto - F et al found that the Gram positive aerobic streptoccoci prevails by a (6.9 %) in the gangrenous and perforated appendices[21], as compared to aerobic streptoccoi isolated from our gangrenous/perforated group (10.5%). This relatively low prevalence rate of aerobic streptoccoci in the isolates is attributed to the

infrequent presence of these organisms as normal inhabitants of the gut, the route by which this microorganism reaches the appendix is most probably haematogenous [9,24]. In 3 patients (3.8 %) with acutely inflamed appendix, no growth of bacterial pathogen was identified. The inflammation of the appendix may be attributed to other non -bacterial pathogens, like viruses[25,9].

References:

1. Russell R.C.G, Williams N.S, Bulstrode C.J.K.: The vermiform appendix, chap 59; Bailey & Love's Short Practice of Surgery, ed 23. London, Arnold, 2000, p 1076.
2. Irvin TT. Abdominal pain: a surgical audit of 1190 emergency admissions. Br.J. surg. 1989; 76: 1121- 1125.
3. Kevin PL, Charles SCJ, Richard JA: Appendix, chap 45; Sabiston Textbook of Surgery, ed 16. Philadelphia, WB Saunders Company, 2001, p919.
4. Burkitt D P. Effect of dietary fiber on stools and transit-times, and its role in causation of disease. Lancet, 1972; ii: 1408- 1412.
5. Brender J D, Marcuse E K et al. Appendicitis in children: Am Dis child 1985; 139: 338 - 340.
6. Martin DL, Gustafson TL. A cluster of true appendicitis cases. Am J sury, 1985; 150: 554-557.
7. Jawetz E, Melilick J, and Adelberg E.A: Enteric Gram- Negative Rods (Enterobacteriaceae), Chap 16; Review of medical microbiology, ed 21, Appleton & Lange, Norwalk. 1998, p 218 - 230.
8. Ganong W F: Regulation of Gastro-intestmal Functions, chap 26; Review of medical physiology, ed 15. Norwalk, Appleton & Lange, 1991, p 474.
9. Galloway WH. Appendicitis in the course of Measles. Br Med J 1953; ii: 1412 - 1414.
10. Al-Janabi I.M, Al-Tikriti F.N: Acute Appendicitis, Clinical and Histopathological Diagnosis, A thesis submitted to the Iraqi Commission of Medical Spesialization, 1992.
11. Abdul Hamid M.B, Al-Faddagh Z: Clinicobacteriological Study of Acute Appendicitis, A thesis submitted to the Iraqi Commission of Medical Spesialization, 1996.
12. Jess P, Bjerragard B, Brynitz. Acute appendicitis: prospective trial concerning diagnostic accuracy and complications. Am. J. surg. 1981; 141:232-234.
13. Boerem W J, Fitzpatrick Rl et al. Acute appendicitis. Aust. N. J. Surg. 1981; 51: 165 - 168.
14. Harold E, Nathanson LK: Appendix and Appendectomy, chap 39; Maingofs Abdominal Operations, ed. 10. Stamford Plaza, Appleton & Lange, 1997,p 1199.
15. Bongard F, Daniel V. Differential diagnosis of appendicitis and pelvic inflammatory disease. AmJ surg. 1985; 90-96.
16. Lewis F R, Holeroft J W, Boey J. Appendicitis: Article review in diagnosis and treatment of 1000 cases. Arch surg. 1975; 110: 677-684.
17. Addiss D C; Shatter N, Fawler BS and Touxe R V. The epidimiology of appendicitis and appendectomy in the United State. Am J epidim. 1990; 132: 910-925.
18. Litovskii IV, Savich El. Anaerobic -aerobic infection in acute appendicitis. Khirurgiia Moskl998; 1:26-29.
19. Veselyi S V. The abdominal cavity microflora of children with appendicular peritonitis. Mikrobiol - Z. 1999 May-Jun.;61(3):46-51.
20. Bodnar B M. Characteristics of peritoneal exudate micro flora in children with appendicular peritonitis. Klin-Khir. 1997 (11-12): 64.
21. Ronchetto F, Milan: P, stacchini E, Guascoc. Bacteriology of abdominal pus in 43 cases of acute appendicitis and appendical absscess at the Iverea - castellamonte. G - Bacterial Virol - Immunol. 1991 Dec.; 84(1-12): 77-86.
22. Fujita W, Shigemoto H, Nishimota T. Acute appendicitis a study in 118 patients. Nippin Geka 1985; 86: 464-469.
23. Rautio M, Saxen H, siitonen A, Somer H. Bacteriology of histopatho logically defined appendicitis in children. Pediatr. Infet. Dis. J 2000 Nov; 19(11): 1078-1083.
24. EL - Fadil, Omer E Y, Abdin M E. The role of bacterial pathogen in preoperative sepsis of acute appendicitis. East African Med. J 1985; 397:85-89.
25. Tobe T. In apparent virus infection as a trigger of appendicitis. Lancet 1965; I: 1343 - 1346.