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The Importance of Intraoperative Culture Swabs for The Management of Patients With Complicated Appendicitis Who Undergoing Laparoscopic Appendectomy

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ABSTRACT
Background: There is a controversy about the benefits of intraoperative culture swabs (IOCS) in appendectomy patients, some studies show no efficacy of such procedures while others support its use.
Aim of the work: This study aimed to determine the epidemiological, clinical data, isolated microorganisms, and antibiotic resistance patterns in patients who underwent Laparoscopic appendectomy (LA) and evaluation of whether IOCS can modify the rate or management of post-appendectomy complications.
Methods: the study was conducted on 292 patients who underwent laparoscopic appendectomy and intraoperative culture and sensitivity were done. Patients' electronic medical records were reviewed for the relevant demographic, perioperative, and postoperative data.
Results: The most common isolated microorganisms were *E coli* 36.6%, and *Klebsiella* species 21.25% followed by *Pseudomonas* 8.33% which represents (21.27% and 5.18 % in complicated and non-complicated cases, respectively). Isolated *Pseudomonas* spp. and extended-spectrum beta-lactamase (ESBL) resistant *Enterobacteriaceae* were less frequently encountered (8.35%) but were resistant to most beta-lactam groups.
Conclusion: 2^{nd} generation cephalosporins and metronidazole were used as a first-line postoperative treatment in most uncomplicated cases. This treatment regimen was not effective against *Pseudomonas* spp and most ESBL-resistant strains. Quinolones were added to the regimen in complicated and severe cases, its use was reserved for complicated cases. A lower rate of complications was documented among studied patients in whom IOCS were used comparable to other studies' results. The treatment protocols were adjusted in a significant number of patients according to the result of IOCS, this reflects that its clinical implementation in routine surgical workup is clinically justified.
Keywords: Microbiology, Bacteria, Swab, Laparoscopy, Appendicitis, Antibiotic resistance.

INTRODUCTION

Appendicitis remains the most common cause of acute abdomen and surgical emergencies **(1).** It affects 96.5 to 100 people per 100,000 adults per year worldwide with a range from 7 to 14% **(2, 3)**. Over the last decades, there have been several trials suggested that antibiotics usage as the first approach without surgery is relatively safe **(4, 5).**

However, a systematic review with meta-analysis showed that surgical resection remains the treatment of choice for acute appendicitis (AA), although antibiotic therapy alone may be considered in some selected patients **(6).** Furthermore, relatively broad-spectrum antibiotics and combinations of multiple antibiotics were used in antibiotics alone-treated groups in most studies **(7).** Although, such treatment may be effective for major microorganisms that cause AA, it leads to multidrug resistance emerging and in fact, antibiotic resistance is a resurging problem worldwide with multiple major medical sequels **(8).**

Concerning the preoperative antibiotic treatment protocol, it is recommended to be given to all patients with acute appendicitis, whereas postoperative antibiotics are only prescribed in cases of complication **(9).** In the treatment of acute appendicitis, appropriate antibiotics have a major role in patients with infectious complications after surgery or in those receiving nonsurgical treatment. Appropriate choice of antibiotics should include agents effective against facultative, aerobic Gram-negative, and anaerobic organisms and should be based on knowledge of microbial testing **(7).**

Microbiological culture during appendicectomy is a routine medical practice. This practice was established after Altemeier. 1938 who showed that polymicrobial flora was present in the peritoneal cavity in cases of perforated appendicitis. Although many surgeons tend to rely on the proven efficacy of empirical antibiotic therapy rather than microbiological swab results **(10-12)**.

Obtaining intraoperative culture swabs (IOCS) in appendectomy patients is important to identify the pathogen and adjust the antibiotic regimen. **Peña *et al*. (13)** and **Felber *et al*. (14)** reported that IOCS can help in knowing antibiotic resistance patterns in a specific population or institution, and therefore guide the most effective empiric antibiotic regimen in patients undergoing LA rather than being blindly treated. In a cohort study conducted by **Coccolini *et al.* (15)** on 1431 AA patients, they reported that antimicrobial resistance was strictly linked to the clinical outcome of the patients and adequate empirical antimicrobial therapy being guided by microbiological swab results is fundamental to counteract bacterial resistance and cutting down postoperative complications.

Vanhatalo *et al*. (16) declared that the application of traditional culturing methods combined with new

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advanced molecular techniques for organism identification will provide extensive information about microbiological factors in the etiology of complicated and uncomplicated AA cases. Also, differentiating between etiological and non-etiological microbiota in the collected specimens**.**

There has been a major controversy raised about the benefit of microbiological swab usage in appendectomy patients last decades, some surgeons support its routine application (10, 15, 17), while other surgeons are against it (18, 19)**.** Others suggest that culture and sensitivity may be of value only in special patients such as high-risk and immunocompromised patients in whom there is an increased risk of non-resolution of intra-abdominal infections (1, 14).

So, the aim of the current study was concerned with the demographic, clinical, and bacteriological evaluation of laparoscopic appendectomies (LA) among different age groups with different nationalities and to determine the usefulness of IOCS as routine practice in the setting of complicated appendicitis and whether its use modify the rate or management of post-appendectomy complications in those patients.

MATERIALS AND METHODS

Study design: An observational prospective study was conducted at Al-Ansari Specialized Hospital, Yanbu Industrial City, Saudi Arabia through the period from May 2015 to November 2023. Two hundred and ninety-two patients diagnosed with appendicitis were selected, intraoperative culture and sensitivity were done, and patient's electronic medical records were searched for the relevant data for the study purpose. The clinical characteristics included age, sex, preoperative laboratory results, and perioperative data, which included length of hospital stay, antibiotics prescribed, microbiological profile, antibiotic resistance pattern, complication, mortality rates, and the adequacy of antibiotic prophylaxis all were retrospectively reviewed. Postoperative morbidity was defined as any deviation from the normal postoperative course until postoperative 30 days [20]**.** Patients who had surgical site infections (SSI) were classified into two groups according to the guidelines from the Centers for Disease Control and Prevention: superficial SSI and organ/space SSI [21]. we termed the latter group as Intra-abdominal abscess (IAA) group.

Surgical procedure:

As per the current study, LA was performed by conventional three-port procedures. The conversion to open was defined as any additional incision other than that initially planned for surgery [22]**,** the rate of conversion to open was reviewed and documented in the current study.

Microbiology:

The appendix was extracted laparoscopically, the lumen was opened, and a wet sterile swab was used to collect the infected material inside, luminal contents of the appendix were swabbed immediately after appendectomy as demonstrated in figure (1). A surgical swab with transport media was used to collect and transport clinical specimens to the testing laboratory. The swab was designed to maintain the viability of the collected bacteria during transport and storage. The swabs were received by the Clinical Pathology Department (Microbiology Unit) for further processing. Organism identification and antibiotic susceptibility testing (AST) according to international standard operating procedures guidelines where the specimens were inoculated on blood agar, chocolate agar, and MacConkey agar. These samples were cultivated at 37 °C for 48 hours. Pathogens were identified and extended-spectrum β-lactamase (ESBL) susceptibility testing was performed.

Preoperative laboratories workup:

It included mainly complete blood count (CBC) and C- reactive protein (CRP). CBC was measured by coulter LH 750, white blood cell (WBC) count was considered elevated above 11,000/microliters while CRP was measured by immunoturbidimetric assay, it was considered positive at a cutoff level above 6 mg/dl.

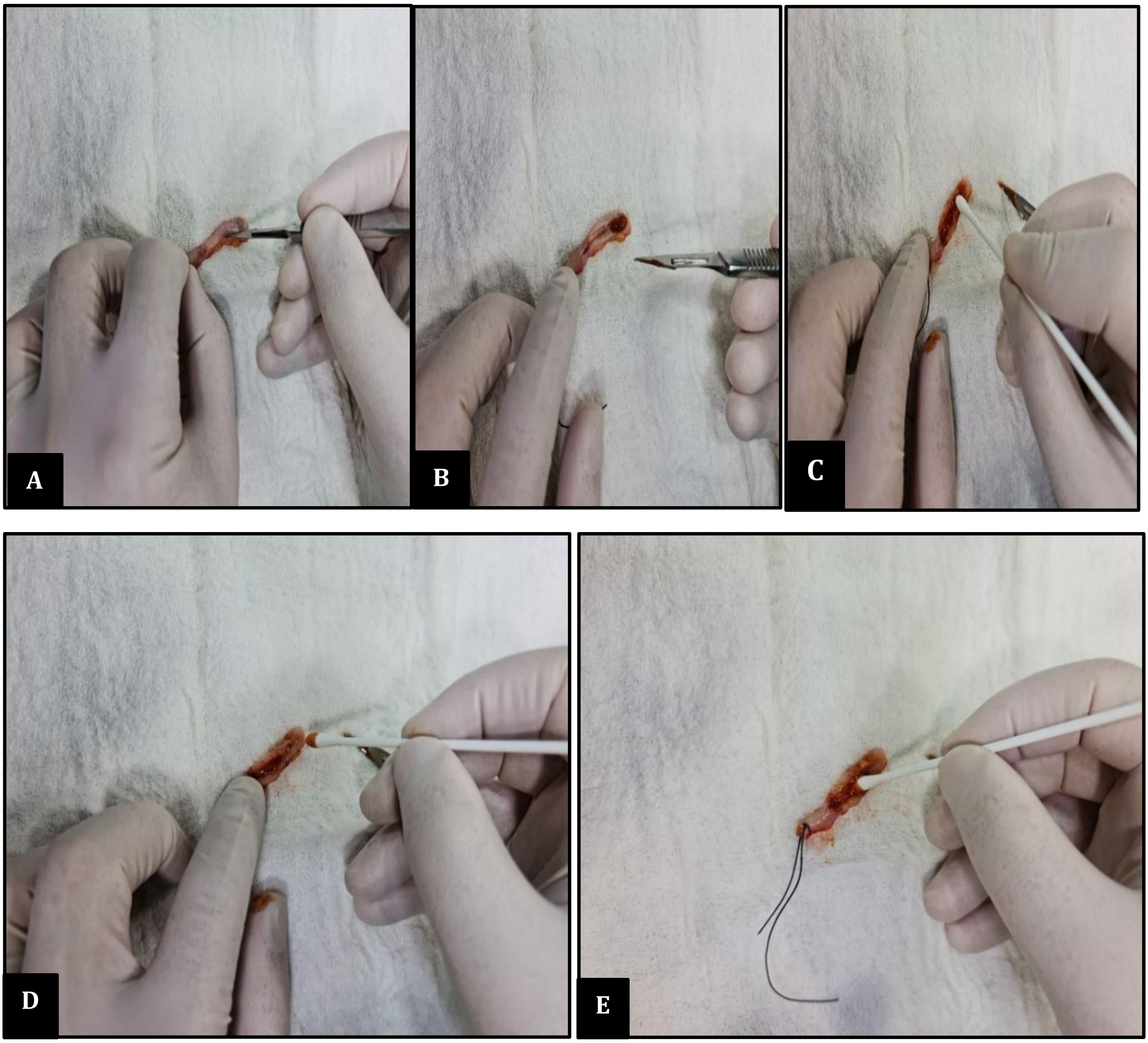

Figure (1): The steps of taking the microbiological swab from the appendicular infected material. The surgeon ensured that a good sample has been collected, by rolling the swab many times inside the infected material. The samples were taken from the peritoneal fluid only in the perforated or gangrenous appendix.

Inclusion criteria: Patients who underwent laparoscopic appendectomy (LA) in whom intraoperative abdominal culture and sensitivity were performed with available medical data upon reviewing their electronic medical records.

Exclusion criteria: Patients who underwent open appendectomy or those with insufficient data upon reviewing their medical records.

Ethical approval: The study underwent a thorough evaluation and received permission from The Research Ethics Committee, Faculty of Medicine, Ain Shams University (reference number: FM ASU R172/2023) and from the concerned institutional Research Ethics Committee. The study followed the Code of Ethics of The World Medical Association (Declaration of Helsinki) for studies done on humans.

Statistical evaluation: Data were analyzed using the Statistical Package for Social Sciences (SPSS version 25). Descriptive analyses were performed to obtain the means ± SD for quantitative data, and Numbers and frequencies for qualitative data. Bivariate analyses were performed using the independent samples “t” test, Mann- Whitney test, and the Chi-squared test for categorical variables. P value ≤ 0.05 was considered significant.

RESULTS

Two hundred and ninety-two patients diagnosed with AA who underwent LA procedure were included in the study during the period from May 2015 to November 2023. Intraoperative abdominal culture and sensitivity were performed, and patient's electronic medical records were assessed and checked for perioperative, and postoperative relevant data**.** Table (1) demonstrated patient characteristics including demographic, laboratory, and clinical data of the studied population with comparative statistics between complicated versus uncomplicated appendicitis. The age of the included patients ranged from 7 - 73 years, most of them were young adults with a mean age of 32 ± 12. Male patients were 227 and 65 were females with a male-to-female ratio of 3.5:1. The patients were screened for diabetes as a major infection risk factor, 11 patients (3.8%) were diabetic, while the remaining were nondiabetic patients with a significantly higher number of patients in complicated appendicitis versus uncomplicated group (P value 0.005). The study included patients from different nationalities, most of them were Saudi and Egyptian (figure 2). A highly significant correlation existed between the clinical presenting symptom (fever, vomiting, diarrhea, and non-localized abdominal pain) with the type of appendicitis being higher among complicated cases either in the presence of abdominal abscess or generalized peritonitis (P < 0.001). Additionally, a significant correlation existed between fever, (vomiting & diarrhea), frequency of urination, and the age of patients being higher among the younger age group (P value 0.003, 0.006, and < 0.001 respectively**).** Regarding the hospital stay, it ranged from hours (in simple appendicitis) who were discharged on the same day up to 8 days in complicated cases (P value < 0.001), the mean for the hospital stay was 2 days, which reflects rapid recovery and rapid discharge in most studied uncomplicated cases.

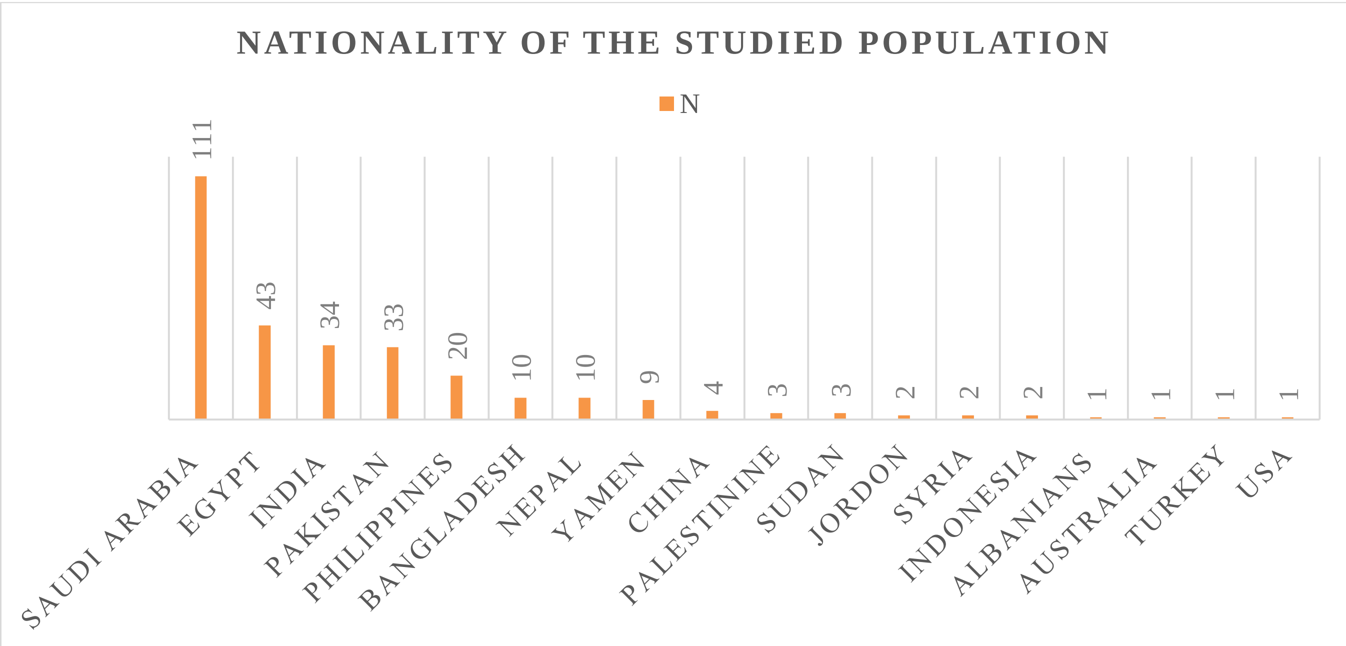

Figure 2: Diagram demonstrating the different nationalities of the studied population.

Table (1): Patient characteristics; demographic, laboratory, and clinical data of the studied population

Patients Characteristics		Total number of patients		Uncomplicated Appendicitis		Complicated Appendicitis		P value
		N	%	N	%	N	%	
		292	(100)	245	(83.9)	47	(16.1)	
Demographic variables: - Sex	Female	65	(22.3%)	56	(22.9%)	9	(19.1%)	0.54
	Male	227	(77.7%)	189	(77.1%)	38	(80.9%)	
- Age	In years Mean (SD)	32 (12) **		31 (11) **		35 (13) **		0.32
- hospital stay	(In days) Mean (SD)	2 (1) **		(2)1**		(4)1**		<0.001*
Laboratory variables: - WBC	X 10^9/ cmm3	12.7 (7.2) **		11.8 (4.5) **		17.2 (13.9) **		<0.001*
- CRP	mg/dl	48.7 (71.5) **		31.5 (42.0) **		136.6 (115.2) **		<0.001*
Clinical variables: - Fever	Negative	254	87.0%	230	93.9%	24	51.1%	<0.001*
	Positive	38	13.0%	15	6.1%	23	48.9%	
- abdominal pain	localized	243	83.2%	230	93.9%	13	27.7%	<0.001*
	Non localized	49	16.8%	15	6.1%	34	72.3%	
- vomiting & diarrhea	Negative	246	84.2%	231	94.3%	15	31.9%	<0.001*
	Positive	46	15.8%	14	5.7%	32	68.1%	
- frequency of urination	Negative	269	92.1%	230	93.9%	39	83.0%	0.011*
	Positive	23	7.9%	15	6.1%	8	17.0%	
- DM	Negative	281	96.2%	228	97.5%	39	89.4%	0.005*
	Positive	11	3.8%	6	2.5%	5	10.6 %	
Clinical Presentation: -intra-abdominal abscess	Negative	265	92.0%	241	100.0%	24	51.1%	<0.001*
	Positive	23	8.0%	0	0.0%	23	48.9%	
-Generalized peritonitis	Negative	255	88.2%	242	100.0%	13	27.7%	<0.001*
	Positive	34	11.8%	0	0.0%	34	72.3%	
Postoperative complicati - SSI	Negative	277	94.9%	238	97.1%	39	83.0%	<0.001*
	Positive	15	5.1%	7	2.9%	8	17.0%	
- post operative abscess	Negative	287	98.3%	245	100.0%	42	89.4%	---
	Positive	5	1.7%	0	0.0%	5	10.6%	
- intestinal injury/ perforation	Negative	287	98.3%	241	98.4%	46	97.9%	0.811
	Positive	5	1.7%	4	1.6%	1	2.1%	
- Readmission	Negative	285	97.6%	244	99.6%	41	87.2%	<0.001
	Positive	7	2.4%	1	0.4%	6	12.8%	
- converted to open surgery	Negative	288	98.6%	245	100.0%	43	91.5%	<0.001 *
	Positive	4	1.4%	0	0.0%	4	8.5%	
- mortality	Negative	292	100.0%	245	100.0%	47	100.0%	----
Treatment: - preoperative empirical treatment	2nd g cephalosporin	250	85.6%	245	100.0%	5	10.6%	<0.001 *
	3rd g cephalosporin	42	14.4%	0	0.0%	42	89.4%	
- postoperative treatment	1st g cephalosporin	2	0.7%	2	0.9%	0	0.0%	<0.001 *
	2nd g cephalosporin	181	65.3%	178	77.4%	3	6.4%	
	3rd g cephalosporin	21	7.6%	20	8.7%	1	2.1%	
	4th g cephalosporin	19	6.9%	11	4.8%	8	17.0%	
	quinolones	21	7.6%	6	2.6%	15	31.9%	
	penicillin's	15	5.4%	13	5.7%	2	4.3%	
	combinations	18	6.5%	0	0.0%	18	38.3%	

WBC: white blood cell count, SSI: surgical site infection DM: diabetes mellitus N= number, ** Mean (SD), * Significant P value

Note: Some data are missed from the electronic medical records, so the total number of some analysed variables were not equal to the total number of studied populations.

Concerning the laboratory parameters, CBC and CRP were ordered for the majority of patients. Most of them showed mild elevation of laboratory parameters (WBCs and CRP; 55.6% and 52% respectively). And to a lesser extent patients were presented with normal laboratory parameters (Atypical presentation of appendicitis or very early presented cases), while the moderate and marked elevation of CRP and WBCs were recorded in a minority of studied patients (Figure 3 and table 2).

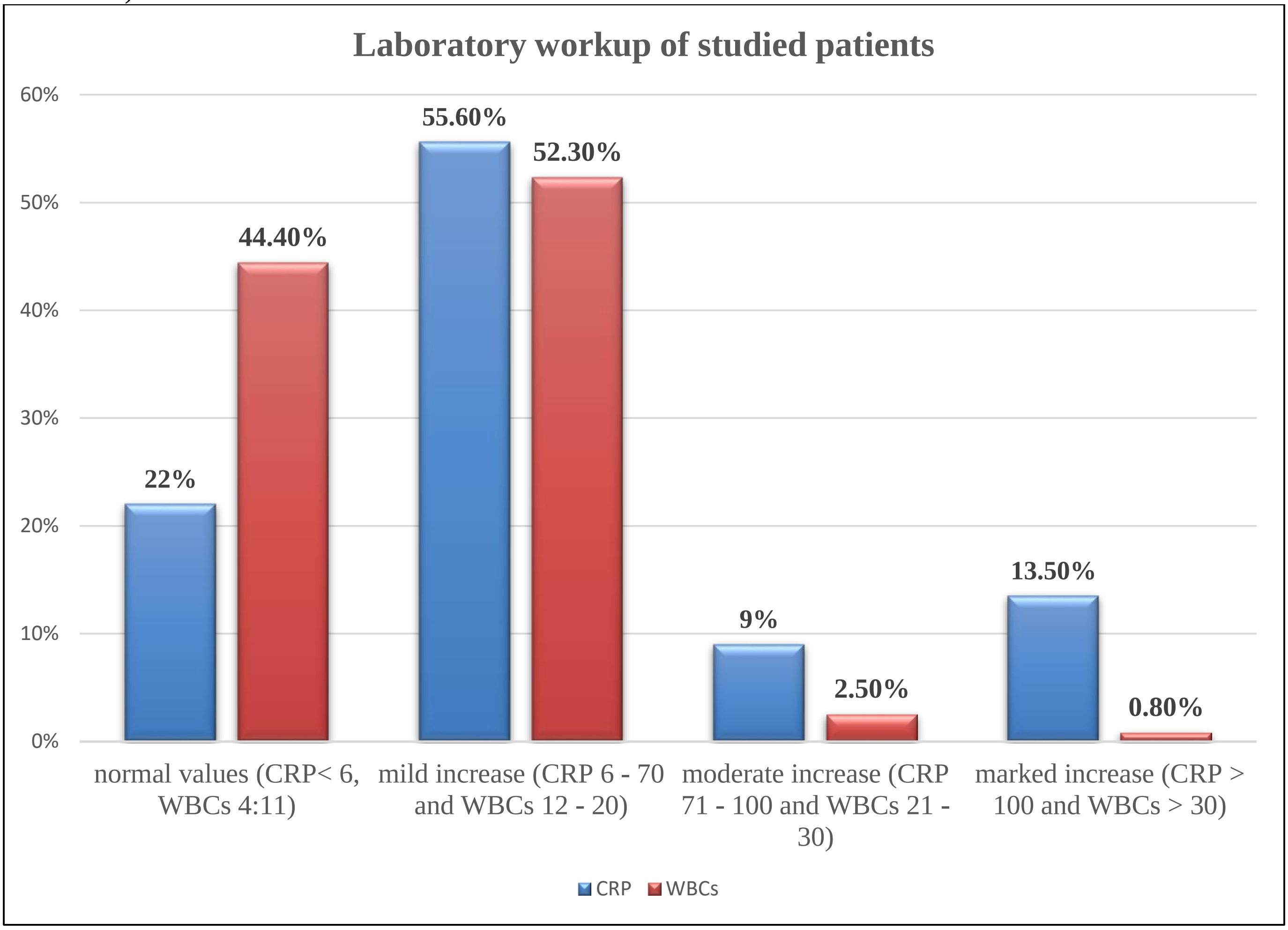

Figure (3): Laboratory workup of studied patients categorized as demonstrated diagram, most of the patients were presented by mild increase in either WBC and/ or CRP.

Table (2): Laboratory workup categorized into different groups according to the level of WBC and CRP increase in each patient.

		Number	%
CRP levels	Normal (< 6) mg/ dl	49	22.0%
	Mild (6 – 70) mg / dl	124	55.6%
	Moderate (71 – 100) mg/ dl	20	9.0%
	Marked (> 100) mg/ dl	30	13.5%
WBC levels	Normal (4 – 11) x 10^9/ cmm3	108	44.4%
	Mild (12 – 20) x 10^9/ cmm3	127	52.3%
	Moderate (21 – 30) x 10^9/ cmm3	6	2.5%
	Marked (> 30) x 10^9/ cmm3	2	0.8%

Regarding the clinical presentations and postoperative complications, 47 /292 presented by complicated appendicitis (16.1%) either in the form of generalized peritonitis [34 cases (11.8%)], or intra-abdominal abscesses IAA [23 cases (8.0%)]. The remaining patients had non-complicated presentations [245/292 (83.9%)]. By follow-up of appendectomy patients, 15 cases developed postoperative superficial infection (SSI) (5.1%), postoperative intra-abdominal abscesses IAA [5 cases (1.7 %], intestinal injury or perforation [5 cases (1.7 %)], while no mortality was documented in the current study. Comparative statistics between simple (uncomplicated) and complicated appendicitis regarding the nationality (Saudi versus non-Saudi patients) were performed, and a non-significant difference was documented (P value equal to 0.81) (Figure 4 and table 3).

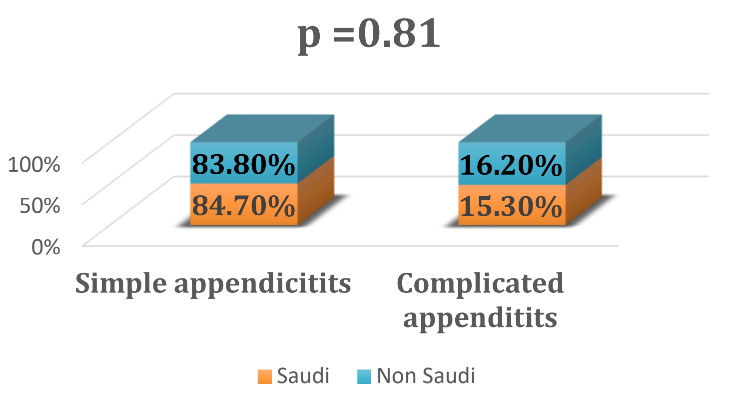

Figure (4): Comparative statistics between simple (uncomplicated) and complicated appendicitis regarding the nationality (Saudi versus non-Saudi patients), the non-significant difference was illustrated with P value equal to 0.81.

Table (3): Comparative statistics between simple (uncomplicated) and complicated appendicitis regarding the nationality (Saudi versus non-Saudi patients)

		Saudi		Non Saudi		Total		P value
		N	%	N	%	N	%	
type of appendicitis	Uncomplicated	94	84.7%	150	83.8%	244	84.1%	0.81
	complicated	17	15.3%	29	16.2%	46	15.9%	

From a microbiological point of view, 16 % of the culture results showed no growth after 48 hours of incubation, while bacterial isolates were documented from the remaining cases (84 %). Of those samples that yielded bacteria, twelve cases yielded mixed infections. The most common organisms isolated were *Escherichia coli* followed by *Klebsiella* spp. followed by *Pseudomonas* spp (Table 4 & figure 5).

Table (4): The distribution of isolated microorganisms and the result of microbiological culture in both studied groups; complicated and uncomplicated patients

		Type of appendicitis					
		Uncomplicated		Complicated		Total	
		Count	Column N %	Count	Column N %	Count	Column N %
		193		47		240	
Culture	*E coli*	78	40.41%	10	21.27%	**88**	**36.6%**
	Klebsiella	41	21.2 %	10	21.27%	51	21.25%
	Pseudomonas	10	5.18 %	10	21.27%	20	8.33%
	E. coli (ESBL)	8	4.1 %	7	14.89 %	15	6.25%
	Anaerobic	8	4.1%	5	10.63%	13	5.41%
	Klebsiella (ESBL)	2	1.03 %	2	4.25 %	4	1.7%
	Proteus	4	2.07 %	0	0.0%	4	1.7%
	Acinetobacter	2	1.0%	2	4.25%	4	1.7%
	Proteus (ESBL)	0	0.0%	1	2.12%	1	0.4%
	Staph CONS	1	0.5%	0	0.0%	1	0.4%
	No growth	39	20.2%	0	0%	39	16.25%
P value		<0.001*#		0.004*#		<0.001*$	

E Coli = Escherichia coli ESBL = Extended spectrum beta lactamase $ p value of comparison between the two groups # p value of comparison within groups

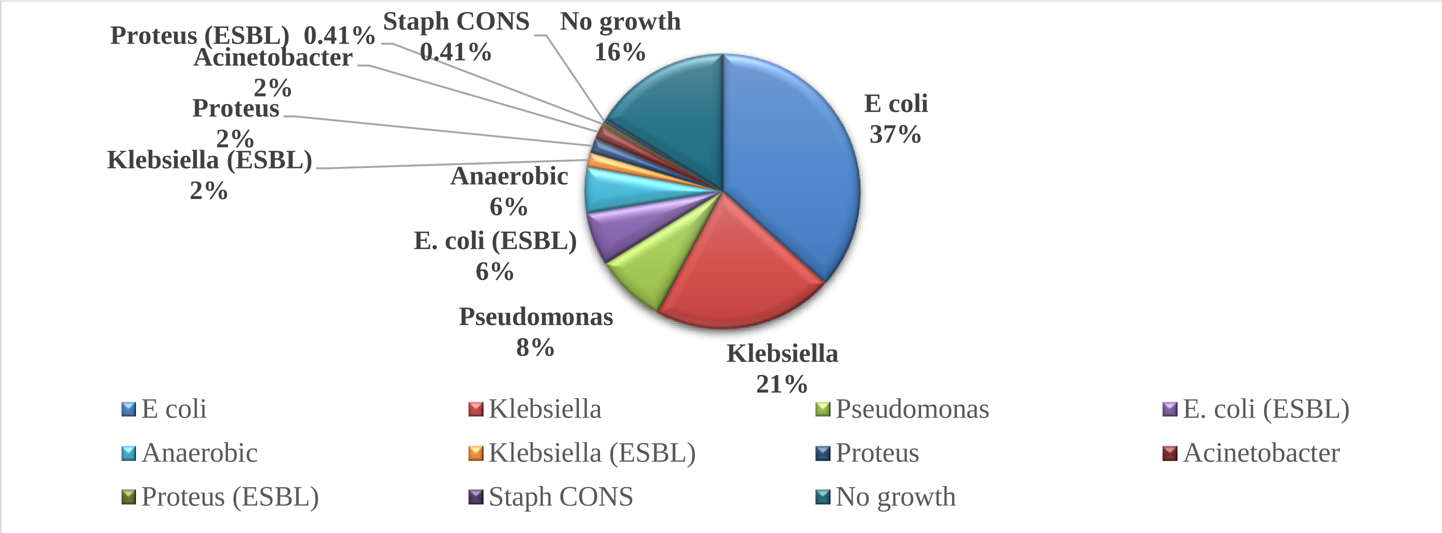

Figure (5): The result of the intra-abdominal culture swab and the most common microorganisms isolated.

Concerning the antibiotic-resistant pattern of isolated microorganisms, 62 % of the reported results were positive for resistant strains to penicillin's group regardless of the name of the microorganisms isolated. For the cephalosporins group, the resistance was encountered in 44 %. Regarding the quinolones group, the documented resistance was less frequent than both penicillins and cephalosporins groups with reported resistance in 26.9 % of cases. For the carbapenem group, the documented resistance was 11.5%, for aminoglycosides 8.7 %, for tetracyclines 45.6%, and for Sulfamethoxazole and trimethoprim group 50.5 %**.** The antibiotic-sensitivity pattern for different antibiotic groups studied in the current study corresponding to each microorganism is demonstrated in table (5).

Table (5): frequency of antibiotic susceptibility testing for each individual antibiotic and its general group

	Total	***E coli***	***E coli (ESBL)***	***Klebsiella***	***Klebsiella (ESBL)***	***Pseudomonas***	***Proteus***	***Proteus (ESBL)***	***Acineto-bacter***	***Staph. CONS***
Penicillin's	**38.0%**	52.3%	0.0%	56.4%	0.0%	0.0%	50.0%	0.0%	0.0%	0.0%
Amoxicillin/ clavulanic acid	50.0%	70.9%	0.0%	66.7%	0.0%	0.0%	100.0%	0.0%	0.0%	0.0%
ampicillin/ sulbactam	47.8%	66.3%	0.0%	69.2%	0.0%	0.0%	50.0%	0.0%	0.0%	0.0%
Piperacillin/ tazobactam	85.9%	88.4%	93.3%	92.3%	100.0%	65.0%	100.0%	0.0%	75.0%	0.0%
Cephalosporins	**56.0%**	76.7%	0.0%	79.5%	0.0%	**5.0%**	100.0%	0.0%	0.0%	0.0%
2nd G cephalosporin	58.7%	79.1%	**0.0%**	82.1%	0.0%	10.0%	100.0%	0.0%	0.0%	0.0%
3rd G cephalosporin	69.0%	83.7%	0.0%	89.7%	0.0%	55.0%	100.0%	0.0%	50.0%	0.0%
4th G cephalosporin	77.2%	91.9%	0.0%	89.7%	0.0%	75.0%	100.0%	0.0%	50.0%	0.0%
Carbapenams	**88.5%**	91.9%	**57.1%**	94.9%	50.0%	**90.0%**	100.0%	100.0%	75.0%	100.0%
Imipenem/ cilastatin	89.6%	93.0%	57.1%	94.9%	75.0%	90.0%	100.0%	100.0%	75.0%	100.0%
Meropenem	91.3%	94.2%	64.3%	94.9%	50.0%	95.0%	100.0%	100.0%	75.0%	100.0%
Aminoglycosides	**91.3%**	91.8%	**66.7%**	94.9%	100.0%	**100.0%**	100.0%	100.0%	75.0%	100.0%
Amikacin	97.3%	96.5%	93.3%	97.4%	100.0%	100.0%	100.0%	100.0%	100.0%	100.0%
Gentamicin	91.8%	91.9%	71.4%	94.7%	100.0%	100.0%	100.0%	100.0%	75.0%	100.0%
Quinolones	**73.1%**	74.4%	**33.3%**	81.6%	25.0%	**85.0%**	75.0%	100.0%	66.7%	100.0%
Levofloxacin	87.4%	94.2%	46.7%	94.7%	25.0%	95.0%	75.0%	100.0%	66.7%	100.0%
ciprofloxacin	77.5%	79.1%	40.0%	89.5%	25.0%	85.0%	75.0%	100.0%	66.7%	100.0%
Moxifloxacin	76.4%	79.1%	33.3%	84.2%	50.0%	85.0%	75.0%	100.0%	66.7%	100.0%
Tetracyclines group	**54.4%**	57.0%	**20.0%**	65.8%	25.0%	**55.0%**	0.0%	100.0%	100.0%	100.0%
Tetracyclines	57.1%	60.5%	26.7%	65.8%	25.0%	60.0%	0.0%	100.0%	100.0%	100.0%
Doxycycline	60.4%	62.8%	33.3%	71.1%	25.0%	60.0%	0.0%	100.0%	100.0%	100.0%
Sulfamethoxazole and Trimethoprim	**49.5%**	59.3%	**26.7%**	55.3%	0.0%	**35.0%**	25.0%	100.0%	66.7%	100.0%

In context with the patient's clinical condition and microbiological swab results, 26.66 % of patients had their course of antibiotics adjusted in correlation to the result of the microbiological swab (IOCS) and changed accordingly to the appropriate antibiotic regimen.

The isolated microorganisms {especially *Pseudomonas* spp. and *Enterobacteriaceae* resistant strains were frequently not covered by the empirical antibiotics chosen and associated with a post-operative complicated course of the disease, SSIs and prolonged hospital stay (Table 6).

Table (6): Microorganisms encountered from IOCS in patients who developed surgical site infection (SSI) & post-operative intra-abdominal abscess later in the postoperative follow-up period (30 days post-operative)

Microorganisms in patients who had SSI.	Total number of patients = 15 (5.13%) *(Mixed infection encountered in 5 cases)*	
	Number	(%)
Pseudomonas	9	(60%)
Klebsiella	5	(33.3%)
E coli	2	(13.3%)
Anaerobic	2	(13.3%)
Proteus	1	(6.66%)
Acinetobacter	1	(6.66%)
Microorganisms in patients who developed Postoperative intra-abdominal abscess	Total number of patients = 5 (1.7%) *(Mixed infection encountered in 3 cases)*	
Pseudomonas	3	(60%)
E coli	2	(40%)
Proteus	1	(20%)
Acinetobacter	1	(20%)

DISCUSSION

Acute appendicitis is one of the most common diseases and medical emergencies affecting particularly young but also older people. Antimicrobial therapy in appendicitis patients is essential for both empirical and postoperative treatment**.** For many surgeons, the standard has been resection alone with single-shot antibiotics, while real antibiotic treatment is only in cases of perforation or peri typhlitic abscess **[1, 12, 14]**.

In the past few years, exclusive antibiotic treatment has become more popular. No matter, which method a clinician adopts, the appropriate use of antibiotics is inevitable in the treatment of AA patients, based on accurate bacteriology. The initial antibiotic therapy is generally empiric because the patients need immediate treatment, but the culture results need up to 24–72 hours to become available **[7].** We aimed in our study to determine the usefulness of IOCS as routine practice in AA patients and whether its use can modify the rate or management of post-appendectomy complications in those patients.

Since *Enterobacteriaceae* are the most common pathogens isolated from AA patients in our current study, so empirical antibiotics have been directed against these organisms primarily. However, resistance to cephalosporins and quinolones has been reported recently at an increasing rate among isolated *E coli* strains, besides emerging other resistant microorganisms like *pseudomonas* species, which is resistant to most cephalosporin. Therefore, microbiology and antibiotic susceptibility studies in AA patients is sound to be essential when selecting appropriate antibiotics in this clinical situation, as antibiotic resistance patterns are directly linked to multiple major medical consequences affecting the overall patient outcome with a higher rate of postoperative complications **[7, 23, 24]**.

In the current study, when simple uncomplicated appendicitis was suspected on physical examination and abdominal/pelvic computed tomography, a second-generation cephalosporin combination with metronidazole was given intravenously. With patients suspected of having more severe infections, broad-spectrum antibiotics were chosen, including 3rd or 4th generation cephalosporin while in complicated severe cases (IAA/ generalized peritonitis) quinolones, piperacillin/tazobactam, or meropenem were added to the treatment regimen. Follow-up of the cases after three days post-operative is the rule of the management to correlate with the microbiological result and antibiotic sensitivity testing and tailor the management accordingly.

Concerning the microbial growth recovered, the most identified aerobic microorganisms were *E. coli*, *klebsiella* spp., followed by *Pseudomonas*, then gram-positive bacteria. These results are not surprising since coliforms and anaerobes were the most frequently isolated bacteria, as reported by other studies **[7, 13, 25]**. *E. coli* was the most common pathogen isolated in a considerable number of published studies **[13, 17, 24]**. While, **Plattner *et al.*** **[26]** reported that *Bacteroides fragilis* was the most common intra-abdominal microbes for perforated appendicitis followed by *E coli* positive results.

On the other side, 16 % of culture results were negative for either aerobic or anaerobic microbial growth, which is expected to be encountered in mild and chronic cases that are already under antibiotic coverage. These results are comparable to other studies that reported 20% of acute appendicitis patients in whom IOCS were performed, the culture results were negative for any bacterial isolates **[13]**. Although, this percentage was higher than other series of cases with complicated appendicitis **[27, 28]**. **Bancke *et al*.** **[3]** reported that the bacterial isolation rate in the obtained intraoperative swabs was 51%, with a significantly higher rate observed in patients with complicated appendicitis compared to those with uncomplicated appendicitis, which comes in the same way as our study.

The cause of this variability between the studies depends mainly on the appendix inflammatory status (i.e., simple versus complicated appendicitis) and the collection technique used. The site of microbiological swab collection is a contributing factor whether it is collected directly from the appendicular infected material or blinded peritoneal swabs, besides to the processing steps that differ from one study to another according to the different standard operating procedures (SOPs) for each laboratory [18, 27].

As per the current study, the microbiological swabs from all patients were taken from the appendicular lumen as supported by other studies [7, 17, 24]. The appendicular swab was used rather than blinded peritoneal swabs to ensure a high level of accuracy and exclude contamination from any source of infection that could be concomitantly present. On the other side, some surgeons reported that they used appendicular swabs in most appendectomy patients, while in patients with peri-appendiceal abscess, the specimens were obtained from abscess fluid [7]**.** Additionally, some surgeons performed intraoperative swabs from other sites such as the abdominal cavity and appendix fossa [12], but generally, the studies with luminal swabs presented a higher rate of culture positivity, compared to the studies that performed swabs from other sites [13]**.** This might be explained by significant bacterial translocation that was not identified in noncomplicated appendicitis [7] and a higher load of the causative microorganisms at the primary site of infection.

Concerning the demographic data of the studied population, the mean age was 32 ± 12 years, which is comparable to **Akingboye *et al*.** [29] who reported that most of the presenting AA cases were in the young adult age group with the mean age 37.2 years. By comparing the complicated versus uncomplicated AA patients, in the current study, we failed to show a significant difference between both groups regarding the age of patients (P value 0.32). Unlike **Bancke *et al*.** [3] who reported a higher age group in the complicated cases with a mean of 51 ± 28 (P value < 0.001) between complicated versus simple cases. Regarding gender, the male/female ratio in the current study was 3.5:1, the male gender was more than the females. This comes in line with other studies [29], as they reported that the male/female ratio was 1.35:1. **Song *et al*.** [7] reported that male/female ratio was 1.55:1, but the male percentage was little bit higher in the current study than reported by others as our hospital is in industrial city so most of attending patients were male workers.

The comorbid risk factors for postoperative appendicitis complications were studied and correlated with patient complications, it is well known that a higher rate of complications can be encountered among old age, obese, diabetic, isolation of *pseudomonas*, and ESBL-resistant *Enterobacteriaceae*. In the current study, a highly significant difference was documented regarding diabetes as a risk factor for postoperative complications (P value < 0.001) and isolation of *pseudomonas* and ESBL-resistant *Enterobacteriaceae*. These results come in line with **Peña *et al*.** [13] and **Bancke L *et al*.** [3] who reported that obesity and diabetes are common risk factors for post-operative surgical wound complications.

No mortality could be documented in the current study, while postoperative significant infective complications (IAA) were documented in 1.7 % of studied cases. On the other side, it is reported by one study that 5% of patients developed significant infection postoperatively, with the omission of patients with 'minor' wound infections managed in the community [30].

Concerning the clinical presenting data of studied patients, general and localized clinical symptoms were reviewed and documented including the site of abdominal pain, presence of fever, vomiting, diarrhea, and frequency of urination. It is evident from the statistical analysis that a higher rate of fever, non-localized abdominal pain, vomiting, and diarrhea were documented more commonly in complicated cases rather than simple cases and among the paediatric age group rather than adult patients (P value < 0.001).

Regarding the surgical procedure, as per the current study, laparoscopic appendectomy (LA) was performed in all studied patients by conventional three-port procedures as well as that performed in other previous studies. The conversion to open rate was 1.4%, which is comparable to other studies' results [7].

As regards the laboratory workup of studied patients, most of them were presented with a mild increase in either WBC and/or CRP; 55.6% & 52.3% respectively. In the current study, highly significant differences were documented between complicated versus uncomplicated appendicitis regarding both WBCs and CRP (P value < 0.001). These results come in line with other studies [3] that showed similar results.

Concerning the antibiotic resistance pattern, organisms resistant to broad-spectrum antibiotics were isolated from a significant proportion of the patients in whom swabs were taken, the documented percentage of resistance to penicillin's group was 62%, cephalosporines was 44 %, quinolones group was 26.9 %, carbapenem group was 11.5%, aminoglycosides was 8.7 %, and tetracyclines was 45.6%**.** There is a wide variation between the studies regarding the concerned issue, **Peña *et al*.** [13] reported that the broad-spectrum antibiotic resistance rate was evident among 36% of the patients, while these proportions are higher than 2–16% resistance to broad-spectrum antibiotics reported by other studies [15, 18]**.** In line with our study, **Coccolini *et al*.** [15] reported that microbiological analysis of isolated bacteria in acute appendicitis revealed a surprisingly elevated rate of antimicrobial-resistant pathogens.

Since there was a high incidence of cephalosporin resistance especially among complicated AA cases (44%) comparable to quinolones resistance (26.9%), so we changed the protocol of complicated patients' management, accordingly, shifting the antibiotic from the 3rd or 4th generation cephalosporins to the quinolones antibiotic group, which resulted in better patients' management. This is supported by **Song *et al*.**

[7] study in which they reported that *E. coli* was found to be 82–85% susceptible to quinolones and proposed that quinolones can be used to treat community-acquired complicated intra-abdominal infections. On the other side, **Jeon *et al*.** [24] documented that the use of quinolone as a first-line antibiotic is not recommended. While, in practical settings, it is not easy to exclude these drugs as first-line therapy, especially in complicated and severe cases, because they are highly effective against other microorganisms, especially *pseudomonas aeruginosa* and ESBL resistance *Enterobacteriaceae*.

Regarding the usage of penicillin's group as empirical treatment in the era of antibiotic resistance, amoxicillin/clavulanic acid and ampicillin/sulbactam were not effective against most isolated microorganisms in the present study with overall rates of resistance 50% and 52.4% respectively. Additionally, *pseudomonas aeruginosa* is naturally resistant to ampicillin/sulbactam and only piperacillin/tazobactam can be used to treat these isolates with low resistance rate 14.1%.

Most *Enterobacteriaceae* and *pseudomonas* strains were susceptible for the aminoglycosides group (either gentamycin or amikacin) with an overall rate of susceptibility of 91.3 %, but concerns remain due to nephrotoxicity and ototoxicity side effects, especially in age extremities. These results are supported by other study results [7].

Concerning the complications documented in our study, the postoperative complicated cases represented 8.5 % of all studied patients. These complications lead to prolonged hospital stays, increased medical costs, and compromised overall patient outcomes. SSI was not a rare complication after the AA procedure, as per the current study, the overall SSI was encountered in fifteen patients/292 (5.1%), which was lower than reported by other comparable studies (4.5–14%) [7, 31, 32]. Moreover, the presence of *Pseudomonas* spp. was a significant predictive factor associated with SSI in the current study. **Chen *et al*.** [17] reported similar results as *Pseudomonas* spp. was significantly associated with SSI after appendectomy because it was frequently not covered by empirical antibiotics. **Song *et al*.** [7] documented that *Pseudomonas* spp. was the only significant microorganism associated with SSI according to multivariate analysis adjusting for other clinical factors (P = 0.030). **Pena *et al*.** [13] reported that the rate of postoperative complication was documented among 16.5% of included patients. 33% belonged to the group where intra-abdominal culture swab (IOCS) was used, and 67% of patients where IOCS couldn't be performed.

In our current study, it was evident that microbiological culture is an important step in proper surgical patient management, the importance of microbiological culture, is not only for tailoring the treatment regimen for each patient specifically according to the pattern of antibiotic resistance but also for infection control purposes and changes of antibiotic protocols of treatment according to the antibiogram of isolated microorganisms in each health care facility accordingly. This is supported by other studies results [7, 13, 14] who reported that IOCS can help knowing gut flora resistance in a specific population or institution, and therefore guide the most effective antibiotic regimen in patients undergoing LA**. Felber *et al*.** [14] added that the need for a standard protocol of swabs in investigating AA microbiology is highly recommended to ensure better diagnostic efficacy. As regards the treatment protocol change according to the culture results, 26.6% of studied patients had their course of antibiotics adjusted in response to the result of the microbiological swab and changed according to the appropriate antibiotic regimen as the isolated microorganisms (especially *Pseudomonas* spp. and ESBL-resistant *Enterobacteriaceae* strains) were frequently not covered by the empirical antibiotics chosen and was associated with a complicated course of the disease, SSI and prolonged hospital stay. These results are supported by **Peña *et al*.** [13] who declared that the initial empiric antibiotic course was modified due to bacterial resistance in 36% of the studied patients, while **Bancke *et al*.** [3] reported that the analysis of swab samples obtained during appendectomy for AA can help identify patients at a higher risk of a worse postoperative outcome. However, the frequency of antibiotic regimen changes based on the swab analysis was low.

in the present study, the readmission rate was 2.4 %, this low rate could be explained by a lower rate of complications that necessitate readmission, which could be attributed to many factors including available IOCS culture result for included patients, the procedure of removal of infected appendix was performed by ensuring non-touch technique to decrease the rate of infection spread to the surgical wound (fishing technique for simple cases, inverted glove technique for moderate severity cases, and endo-pack usage in the perforated and severe cases).

The diagnostic value of IOCS can be increased by the proper sample collection, appropriate sample transport, and proper sample processing, followed by comprehensive microbiological work (aerobic and anaerobic culturing) from the microbiologist side and the presence of SOPs for each step guided by updated internationally approved guidelines. And in case of limited feasibility for performing IOCS from all cases, the surgeon can decide to take or not take microbiological swabs selectively from each patient depending on the general clinical condition of the patient being recommended in immunocompromised individuals (Patients with chronic diseases, immunodeficiency patients, and those under immunosuppressive medications, besides to extremes of ages). Additionally, the staging of appendicitis during the time of surgery can guide the medical decision, being recommended to take microbiological swabs from moderate and severe cases either complicated or not. These discussed measurements absolutely improved the patient's health care, lowered hospital stay, and decreased overall morbidities.

In summary, despite intraoperative culture swab usage being more fitting for the routine application as a part of the daily postoperative evaluation, it will add a financial burden and a medical workload, but in fact, catching the right antibiotic with less unnecessary antibiotic side effects, changing the antibiotics prescribed, in addition to saving the cost of treatment complications, antibiotic resistance, nursing care, and bed charges. This as a whole will absolutely make the expense of routine intraoperative culture swab implementation is more than compensated for by the potential savings generated.

CONCLUSION

Post-appendectomy patients' morbidity and complications depend mainly on appendicitis staging at the time of presentation, the severity of the case, and microbiological culture results besides the general comorbidity risk factors. *E. coli* was the most identified microorganism in patients with acute appendicitis, followed by *Klebsiella*.

2nd generation cephalosporins and metronidazole combination can be used as a first-line of treatment regimen in most uncomplicated cases. However, treatment protocols should be changed to a higher level of antibiotics being broad spectrum to be more effective against resistant microorganisms including *Pseudomonas* spp. and most ESBL-resistant Enterobacteriaceae. As per the current study, quinolones were more effective than the beta-lactam group in the management of complicated cases. Its use was reserved for complicated cases. *Pseudomonas* spp. was a significant predictive factor associated with SSI and IAA. Routine intra-operative cultures during laparoscopic appendectomies have relevant clinical advantages and a valuable role in patient management. It is highly recommended to tailor the treatment regimen for each patient according to antimicrobial-resistant specific patterns to cut down patients' complications, patient hospital stays, and readmission rates, and for better clinical outcomes.

Limitation of the study:

The limitation of this study included firstly the retrospective nature of part of the study adds some limitations, although this study was conducted on a large, single-center retrospective cohort to ensure more accurate and reliable results. 2nd, various prophylactic and treatment antibiotics were used in the included patients while this heterogeneity was intended to be included to be more reliably reflecting the routine daily practice. 3rd, some of the antibiotics tested for susceptibility were different from those actually used, so it was difficult to evaluate appropriate antibiotic selection for treatment of some cases. 4th, the result of microbiological culture swab was not available for some studied patients, those patients were included in the study for different clinical variables, but they were not included in the evaluation of diagnostic importance of microbiological swab in the treatment decision.

List of abbreviations:

AA: acute appendicitis **AMR:** Anti-microbial resistance **CBC:** Complete blood count
CRP: C - reactive protein
E. coli: Escherichia coli **ESBL:** Extended-spectrum beta-lactamase **IAA:** Intra-abdominal abscess **ICU:** intensive care unit
IOCS: Intraoperative culture swabs **LA:** Laparoscopic appendectomy **SD:** standard deviation **SOPs:** Standard operating procedures **SPSS:** Statistical Package for Social Sciences **SSI:** Surgical site infections **WBCs:** white blood count.

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REFERENCES

1. **Obsa M, Adema B, Shanka G *et al.* (2020):** Prevalence of acute appendicitis among patient admitted for acute abdomen in Ethiopia: systematic review and meta-analysis. International Journal of Surgery Open, 26: 154-160.
2. **Moris D, Paulson E, Pappas T (2021):** Diagnosis and management of acute appendicitis in adults: a review. Jama, 326 (22): 2299-2311.
3. **Bancke L B, Maak M, Langheinrich M *et al.* (2023):** The role of intraoperative swab during appendectomy in patients with uncomplicated and complicated appendicitis. International Journal of Colorectal Disease, 38 (1): 272.
4. **Svensson J, Patkova B, Almström M *et al.* (2015):** Nonoperative treatment with antibiotics versus surgery for acute nonperforated appendicitis in children: a pilot randomized controlled trial. Ann Surg. ,261(1):67-71.
5. **Salminen P, Paajanen H, Rautio T *et al.* (2015):** Antibiotic therapy vs appendectomy for treatment of uncomplicated acute appendicitis: the APPAC randomized clinical trial. Jama, 313 (23): 2340-2348.
6. **Podda M, Cillara N, Di Saverio S *et al.* (2017):** Antibiotics-first strategy for uncomplicated acute appendicitis in adults is associated with increased rates of peritonitis at surgery. A systematic review with meta-analysis of randomized controlled trials comparing appendectomy and non-operative management with antibiotics. The surgeon, 15 (5): 303- 314.
7. **Song D, Park B, Suh S *et al.* (2018):** Bacterial culture and antibiotic susceptibility in patients with acute appendicitis. International Journal of Colorectal Disease, 33: 441-447.
8. **Frieri M, Kumar K, Boutin A (2017):** Antibiotic resistance. Journal of infection and public health, 10 (4): 369-378.
9. **Van Rossem C, Schreinemacher M, van Geloven A *et al.* (2016):** Antibiotic duration after laparoscopic appendectomy for acute complicated appendicitis. JAMA surgery, 151 (4): 323-329.
10. **Sippola S, Haijanen J, Grönroos J *et al.* (2021):** Effect of Oral Moxifloxacin vs Intravenous Ertapenem Plus Oral Levofloxacin for Treatment of Uncomplicated Acute

Appendicitis: The APPAC II Randomized Clinical Trial. JAMA ,325(4):353-362.
11. **Kakar M (2023):** Clinical, Molecular Biological, and Microbiological Integrated Investigation in the Case of Paediatric Acute Complicated and Uncomplicated Appendicitis: Doctoral Thesis: Sub-Sector – Surgery (Paediatric Surgery). Rīga: Rīga Stradiņš University. https://doi.org/10.25143/prom-rsu_2023-23_dt.
12. **Teng T, Thong X, Lau K *et al*. (2021):** Acute appendicitis–advances and controversies. World Journal of Gastrointestinal Surgery, 13 (11): 1293.
13. **Peña M, Sadava E, Laxague F *et al*. (2020):** Usefulness of intraoperative culture swabs in laparoscopic appendectomy for complicated appendicitis. Langenbeck's Archives of Surgery, 405: 691-695.
14. **Felber J, Gross B, Rahrisch A *et al*. (2023):** Bacterial pathogens in pediatric appendicitis: a comprehensive retrospective study. Frontiers in Cellular and Infection Microbiology, 13: 1027769.
15. **Coccolini F, D'Amico G, Sartelli M *et al*. (2016**): Antibiotic resistance evaluation and clinical analysis of acute appendicitis; report of 1431 consecutive worldwide patients: A cohort study. International Journal of Surgery, 26: 6-11.
16. **Vanhatalo S, Munukka E, Sippola S *et al*. (2019):** Prospective multicentre cohort trial on acute appendicitis and microbiota, aetiology and effects of antimicrobial treatment: study protocol for the MAPPAC (Microbiology APPendicitis ACuta) trial. BMJ open, 9 (9): e031137.
17. **Chen C, Chen Y, Pu H *et al.* (2012):** Bacteriology of acute appendicitis and its implication for the use of prophylactic antibiotics. Surg Infect., 13(6):383–390.
18. **Teng T, Thong X, Lau K *et al*. (2021):** Acute appendicitis–advances and controversies. World Journal of Gastrointestinal Surgery, 13 (11): 1293.
19. **Montuori M, Santurro L, Gianotti L *et al*. (2020):** Uselessness of microbiological samples in acute appendicitis with frank pus: to collect or not to collect? European Journal of Trauma and Emergency Surgery, 46: 835-839.
20. **van Boekel R, Warlé M, Nielen R *et al*. (2019):** Relationship between postoperative pain and overall, 30-day complications in a broad surgical population: an observational study. Annals of surgery, 269 (5): 856-865.
21. **Sliepen J, Onsea J, Zalavras C *et al*. (2021):** What is the diagnostic value of the Centers for Disease Control and Prevention criteria for surgical site infection in fracture-related infection.Injury, 52 (10): 2879-2885.
22. **Park B, Park J, Ryoo S *et al*. (2015):** Effect of visceral obesity on surgical outcomes of patients undergoing laparoscopic colorectal surgery. World journal of surgery, 39: 2343- 2353.
23. **Teklu D, Negeri A, Legese M *et al*. (2019):** Extended-spectrum beta-lactamase production and multi-drug resistance among Enterobacteriaceae isolated in Addis Ababa, Ethiopia. Antimicrobial Resistance & Infection Control, 8: 1-12.
24. **Jeon H, Ju H, Kim G *et al*, (2014):** Bacteriology and changes in antibiotic susceptibility in adults with community-acquired perforated appendicitis. PLoS One, 9(10): e111144. https://doi.org/10.1371/journal.pone.0111144.
25. **Montuori M, Santurro L, Gianotti L *et al*. (2020):** Uselessness of microbiological samples in acute appendicitis with frank pus: to collect or not to collect?. European Journal of Trauma and Emergency Surgery, 46: 835-839.
26. **Plattner A, Newland J, Wallendorf M *et al*. (2021):** Management and microbiology of perforated appendicitis in pediatric patients: A 5-year retrospective study. Infectious Diseases and Therapy, 10: 2247-2257.
27. **Bastiaenen V, Allema, W, Klaver C *et al*. (2020):** Routine histopathologic examination of the appendix after appendectomy for presumed appendicitis: Is it really necessary? A systematic review and meta-analysis. Surgery, 168 (2): 305-312.
28. **Boueil A, Guégan H, Colot J *et al*. (2015):** Peritoneal fluid culture and antibiotic treatment in patients with perforated appendicitis in a Pacific Island. Asian Journal of Surgery, 38 (4): 242-246.
29. **Akingboye A, Davies B, Tien T (2019):** Pus samples in complicated appendicitis: an important investigation or a waste of resources: a prospective cohort study. Scandinavian Journal of Surgery, 108 (1): 55-60.
30. **Sartelli M, Baiocchi G, Di Saverio S *et al*. (2018):** Prospective observational study on acute appendicitis worldwide (POSAW). World Journal of Emergency Surgery, 13: 1-10.
31. **Xiao Y, Shi G, Zhang J *et al*. (2015):** Surgical site infection after laparoscopic and open appendectomy: a multicenter large consecutive cohort study. Surgical Endoscopy, 29: 1384- 1393.
32. **Kim J, Kim H, Park S *et al*. (2015):** Single-incision laparoscopic appendectomy versus conventional laparoscopic appendectomy. Annals of Surgery, 262 (6): 1054-1058.