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RESEARCH ARTICLE

BACTERIOLOGICAL ANALYSIS OF BILE IN CHOLECYSTECTOMY PATIENTS

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ABSTRACT

Background: Cholecystectomy is currently a frequently performed operation. The most common reason for a Cholecystectomy is gallbladder stones. Usually the bile ducts are sterile. However, the presence of gallstones within either the gallbladder or biliary tree is associated with the bacterial colonization of the bile. Therefore, it is important to know the microbiological flora of the gallbladder before prophylactic antibiotics are given. **Aim and Objectives:** To evaluate the microbiological profile of bile from gall bladder in patients undergoing Cholecystectomy. **Methods:** Patients undergoing laparoscopic or open Cholecystectomy in SSG Hospital, Baroda between September 2018 and December 2019 were included in this study and bacteriological examination of bile were done of this patients bile. **Results:** In this study a total of 78 patients(54 female and 24 male) who met the inclusion crieteria were included both open as well as laparoscopic Cholecystectomy . Number of Patients with positive bile cultures were 19(24.36%) and those with negative culture were 59(75.64%). Escherichia coli was the most common isolated bacteria Among positive bile culture group (63.16%) and 15.38% Among all patients. Other organisms isolated were pseudomonas (3.85% Among all patients), Klebsiella (2.56% Among all patients), Coagulase Negative Staphylococcus and Staphylococcus Viridans (1.28%). **Conclusion:** In vast majority of patients the bile was sterile, and Escherichia coli was the most common isolated bacteria Among positive bile culture group.

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INTRODUCTION

Cholecystectomy is currently a frequently performed operation. The most common reason for a cholecystectomy is gallbladder stones. Usually the bile ducts are sterile. However, the presence of gallstones within either the gallbladder or biliary tree is associated with the bacterial colonization of the bile. Gallstones usually led to chronic dyspeptic symptoms and sometimes can manifest as acute cholecystitis. Healthy individuals with no bacteria in the gallbladder sac can still have gallstones. In particular, if the gallbladder stones are pigment stones, bacteria is more likely to be present. In cases of acute cholecystitis, microorganisms in the bile multiply and lead to bacterial infection (Scottish Intercollegiate Guidelines Network, 2000). In patients without gallbladder stone disease, previous biliary intervention is associated with high rates of bacteriobilia. Under conditions of normal bile flow, bacteria in the biliary system are of no clinical significance. Upon bile duct obstruction, bacteria proliferate within the stagnant bile while biliary pressure increases. Eventually, the bacteria presumably translocate into the circulation causing a systemic infection (Meijer, 1990).

Prophylaxis would be appropriate according to bacteria isolated from the bile and could prevent postoperative infections. To justify antibiotic prophylaxis against biliary organisms, it requires to be shown that the bile is colonized with bacteria.

PATIENTS AND METHODS

Patients undergoing laparoscopic or open cholecystectomy in SSG Hospital, Baroda between September 2018 and December 2019.

INCLUSION CRITERIA: Patients undergoing cholecystectomy in SSG Hospital, Vadodara.

EXCLUSION CRITERIA

- Immunocompromise patients.
- Patients' age less then 18 year.
- Patients with any known source of sepsis.
- Patients with history of ascending cholangitis.
- Patients with preoperative diagnosis of empyema of gallbladder.

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- Patients who had undergone endoscopic retrograde cholangiopancreatography (ERCP) and endoscopic sphincterotomy within one week before surgery.

Method of Collection of Data

- The study population was patients undergoing laparoscopic or open cholecystectomy in SSG Hospital between September 2018 and December 2019.

DATA COLLECTED INCLUDES

- Name, Age, Sex, In Patient no (IP no), Diagnosis
- Detailed routine history taking, physical examination and investigations were done.
- Antibiotics given at time of induction.
- Name of procedure and date of procedure
- Any intraoperative complications.

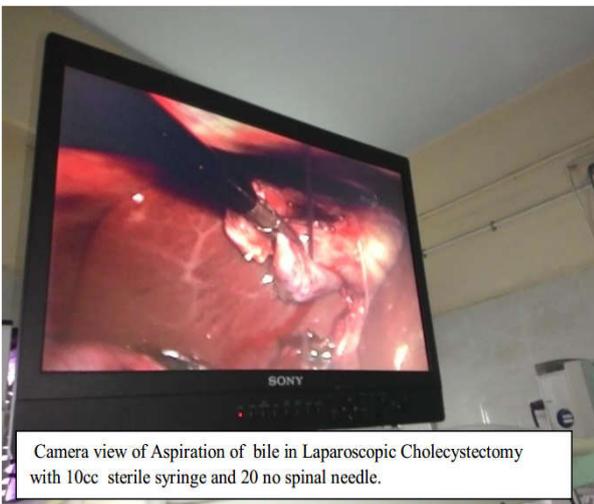
SAMPLE COLLECTION AND TRANSPORT

3cc Bile was aspirated with use of:

- In laparoscopic cholecystectomy with use of Sterile 20 no spinal needle with 10cc sterile syringe.
- In open cholecystectomy : Sterile 10cc syringe with 20 no needle.

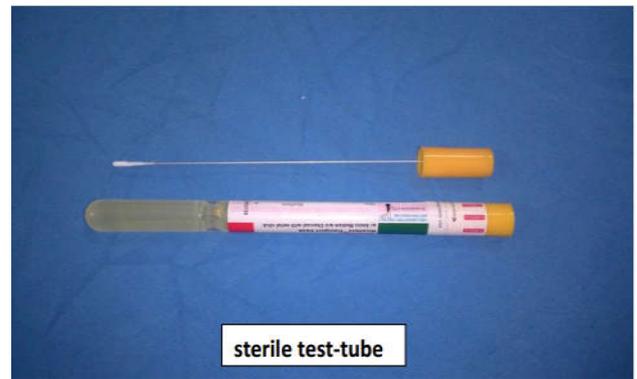


Aspiration of bile in Laparoscopic Cholecystectomy with 10cc sterile syringe and 20 no spinal needle.



Camera view of Aspiration of bile in Laparoscopic Cholecystectomy with 10cc sterile syringe and 20 no spinal needle.

- This collected bile from gallbladder before cholecystectomy was transported to the laboratory in sterile test-tube



SAMPLE CULTURE & SENSITIVITY

- The specimen was evaluated to find out whether it is sterile or has any bacteria present.
- The types of bacteria are determined and whether the amount of isolate is significant or not. And sensitivity to antibacterial agents determined.
- For that: Bile was first examined for gram smear then inoculation of bile for a culture in culture media like Brain Heart infusion Agar, Mac-Conkey Agar and Blood Agar, and then isolated organism identified by Biochemical Tests. Antibiotic Sensitivity Testing was performed for the same.

Statistical methods: Data analyzed for finding the microbiological profile of bile in cholecystectomy patients by using chi square test. P value less than 0.05 considered to be statistically significant.

RESULTS

A total of 78 patients admitted to Surgery Department ,of Baroda Medical College and S.S.G Hospital, were included in the study. 19 patients whose bile cultures were positive and 59 patients whose bile cultures were negative. An evaluation was undertaken to study Bacteriological analysis of bile in cholecystectomy patients and sensitivity of isolated organism to antibiotics was observed.

Table 1. Age distribution

Age in years	Organism Isolated	Organism not Isolated
19-30	08	13
31-40	04	15
41-50	04	18
51-60	01	09
61-70	02	03
>70	00	01
Total	19	59

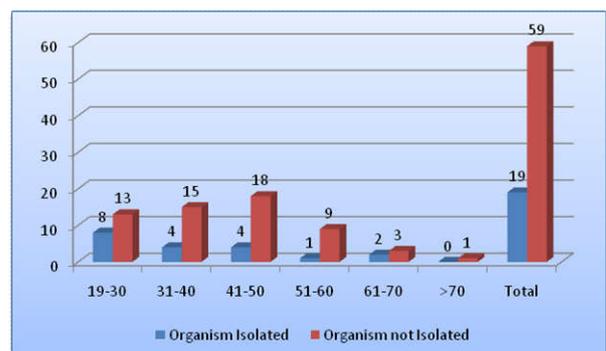


Table 1: Shows age distribution of the study population. Maximum number of patients belonged to 41-50years. Organism isolation from bile found maximum in 19-30years age group.

Table 2. Gender distribution

Gender	Organism Isolated (n=19)		No Organism Isolated (n=59)	
	No	%	No	%
Male	08	10.26	16	20.51
Female	11	14.10	43	55.13
Total	19	24.36	59	75.64

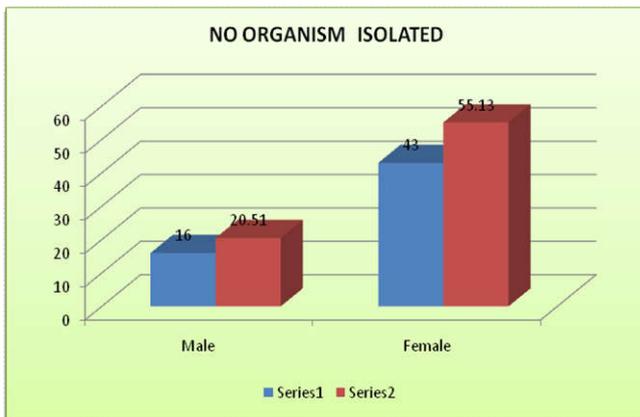
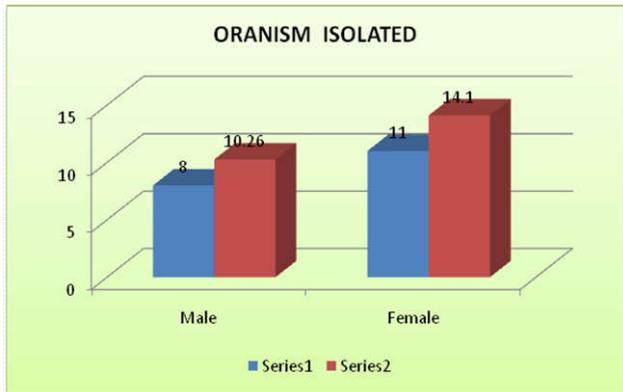


Table 2: Shows gender distribution of study population. In both groups, females predominated.

Table 3. Relation to diagnosis

Diagnosis	Organism Isolated (n=19)		No Organism Isolated (n=59)	
	No	%	No	%
Acute cholecystitis	05	50	05	50
Chronic cholecystitis	14	20.59	54	79.41

Table 3: P value is 0.1034. So difference is not statistically significant in isolation of organism in acute and chronic cholecystectomy.

Table 4. Relation to Surgical Procedure

Surgical Procedure	Organism Isolated (n=19)		No Organism Isolated (n=59)	
	No	%	No	%
Laparoscopic Cholecystectomy	14	21.21	52	78.79
Laparoscopic converted to open Cholecystectomy	05	55.56	04	44.44
Open Cholecystectomy	00	00.00	03	100.00
Total	19		59	

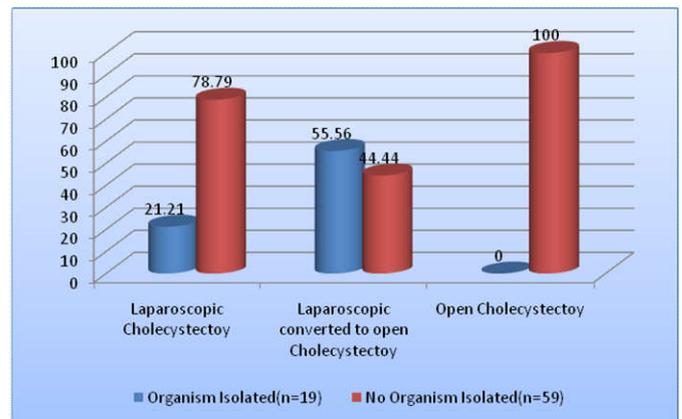
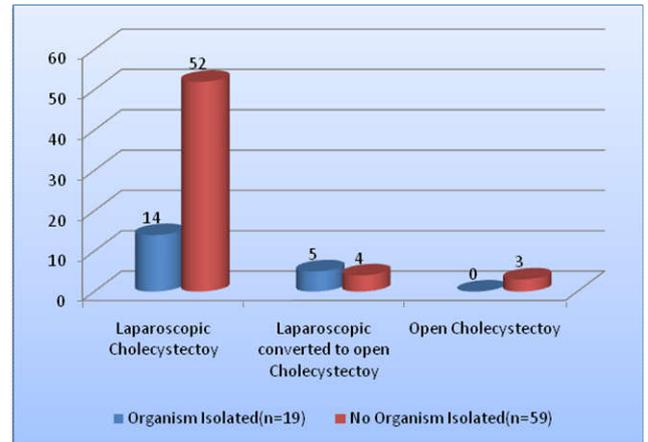
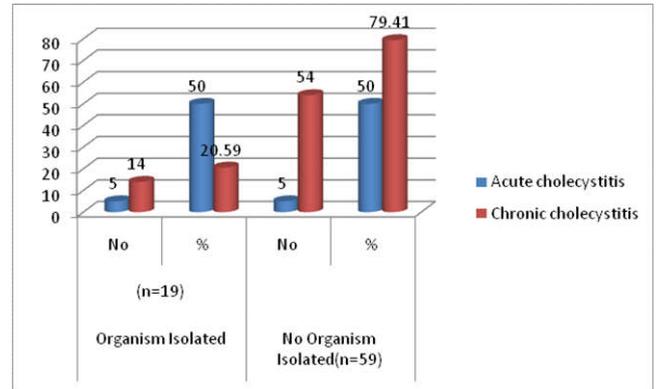


Table 5. Relation with Duration of surgery

Duration of Surgery	Organism Isolated (n=19)	No Organism Isolated Cases (n=59)
0.30-1.30 hours	02	11
1.31-2.30 hours	15	48
2.31-3.30 hours	02	00
Total	19	59

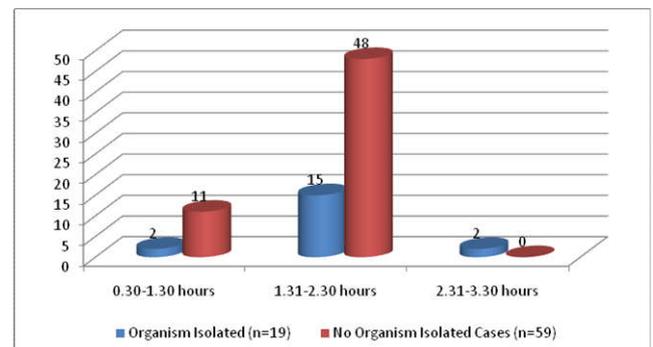


Table 5: In most of the patients duration of surgery was 1.31-2.30 hours.

Table 6. Bacteria isolated in bile culture

Bile culture in patients	Number of patients	Percentage (%)
• E.Coli	12	15.38
• Pseudomonas	03	3.85
• Klebsiella	02	2.56
• Coagulase Negative Staphylococcus	01	1.28
• Staphylococcus Viridance	01	1.28
No growth	59	75.64

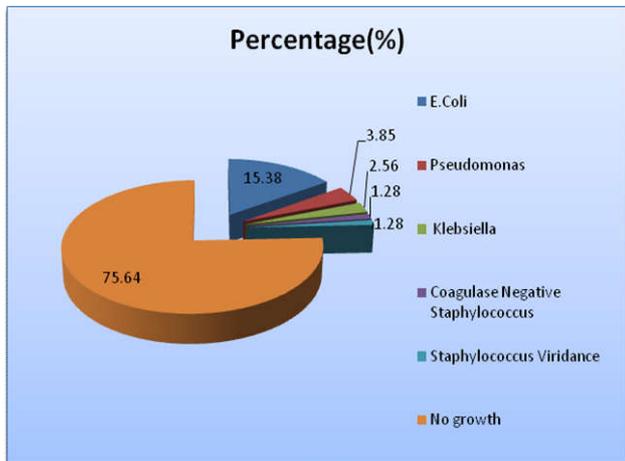
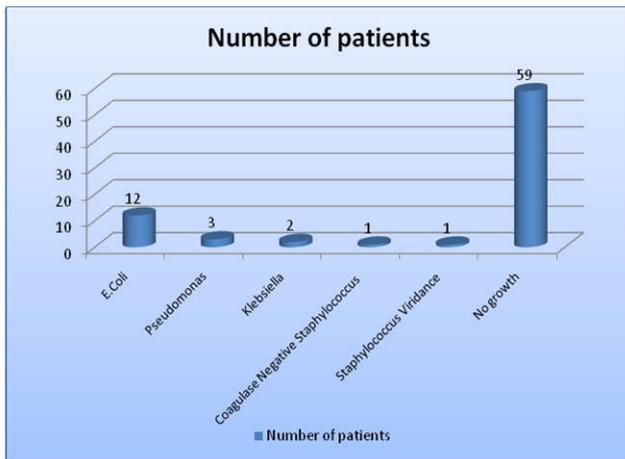


Table 6: The most common organism isolated from bile was E.coli (15.38%) (63.16% Among positive bile culture), Other organism pseudomonas (3.85%), Klebsiella (2.56%), Other organisms isolated are as shown in the table.

Table 7. Sensitivity Patterns of common Antibiotics used

ANTIBIOTICS	SENSITIVE	RESISTANT
Amikacine	11	05
Gentamycin	07	10
Cefotaxime	06	09
Cefipime	10	06
Cefoparazone+Sulbactum	16	00
Piperacilin+Tazobactum	16	00
Levofloxacin	13	00
Meropenem	16	00

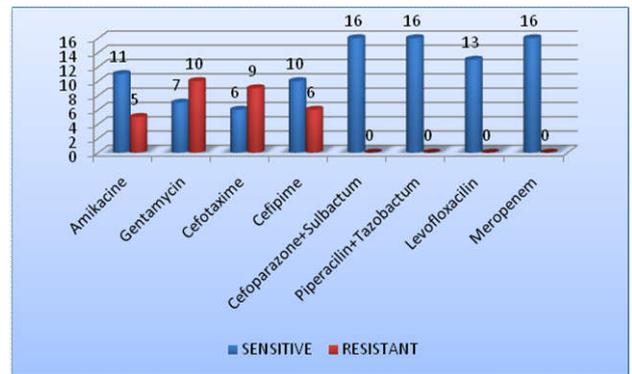


Table 7: Most of the organism shows sensitivity against Cefoparazone, Sulbactum, Piperacilin, Tazobactum, levofloxacin & Meropenem. Multiple antibiotics resistant organisms are tested sensitivity against netillin, Imipenam, Cefepime+tazobactam, Aztreonam, Cefuroxime, Ceftazidime, Amoxycillin+Clavulinic acid and Doxyccline. One organism is multiresistant to all antibiotics except Netillin, Imipenam, Cefepime+Tazobactam, Aztreonam.

Table 8: Post-op wound infection

Post-op wound infection	Organism Isolated (n=19)		No Organism Isolated Cases (n=59)	
	No	%	No	%
No	16	84.21	58	98.30
Yes	03	15.79	01	1.70
Total	19	100.00	59	100.00

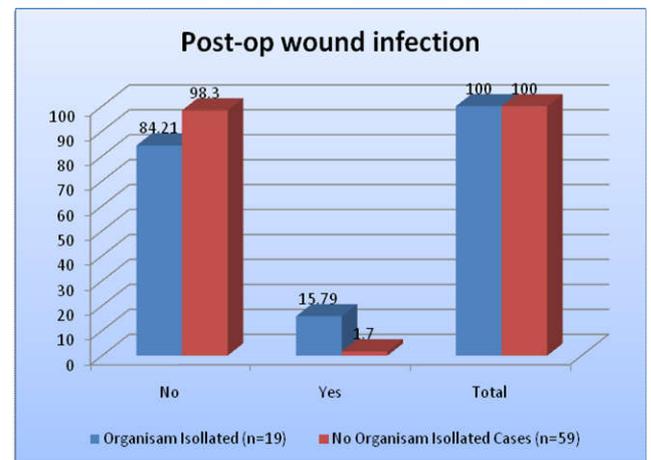
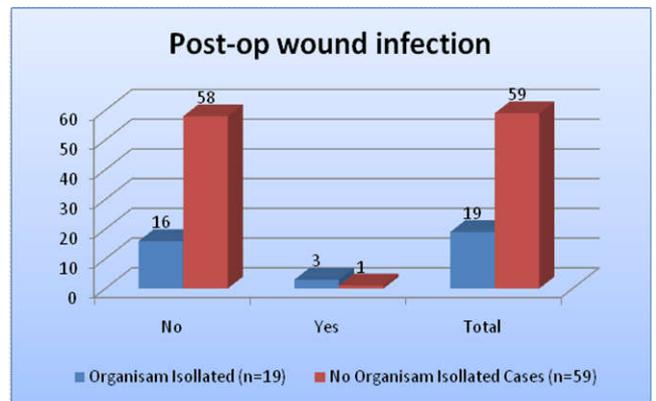


Table 8: Post operative wound infection is more common (15.79%) in group of patients with isolated organism from bile.

Table 9: Wound culture

Post-op Wound culture	Number of patients (n=04)	%
1.E.Coli	1	25.00
2.Enterococcus spp	1	25.00
3.Klebsiella	1	25.00
4.No Organism	1	25.00

Table 10: Correlation of bile culture and Wound culture

Patient	Bile culture	Wound culture
1	Ecoli	Ecoli
2	Ecoli	Enterococcus spp.
3	Klebsiella	Klebsiella
4	No organism	No organism

Study	Alaattin ÖZTÜRK et al.(n=114) (8)	Acharya Suri et al.(n=150) (7)	Muhammed Sameer et al. (n=59) (8)	In this study (N=78)
Organism isolated				
E.coli	3 (20%)	14(53.84%)	19(54.8%)	12(15.38%)
Pseudomonas	2 (13.3%)	7(26.92%)	4(12.9%)	3 (3.86%)
Enterococ supp.	3 (20%)	-	3(9.7%)	-
Klebsiella	-	-	9(29.1%)	2(2.56%)
Staphylococcus aureus	-	5(19.23%)	-	-
Staphylococcus Viridans	-	-	-	1(1.28%)

Cephalosporins	Acharya Suri et al.(n=150) ⁽⁷⁾		In this study (n=78)	
	Sensitivity	Resistance	Sensitivity	Resistance
Cefoparazone	19	07	16	00
Cefepime	18	08	15	01
Cefuroxime	25	01	-	-
Cefotaxime	-	-	06	09

DISCUSSION

In this study a total of 78 patients(54 female and 24 male) who met the inclusion criteria were included both open as well as laparoscopic cholecystectomy. Number of Patients with positive bile cultures were 19(24.36%) and those with negative culture were 59 (75.64%).

Comparison of Micro-organism isolated in various studies

Table shows in all study Escherichia Coli is most common organism isolated. In Acharya Suri et al. (7); On gram staining, no organism was found microscopically in the bile of 24 (82.6%) patients. Later on, it was confirmed that such bile was sterile because no growth appeared on culture plates, both aerobic and anaerobic. In the present study only aerobes were cultivated. Escherichia. coli (53.84%) was one of the most common isolated bacteria followed by Pseudomonas aeruginosa (26.92%) and Staphylococcus aureus (19.23%).

In none of the cultures Streptococcus, Clostridium or Klebsiella was present. In this study, E.coli(15.38%) was the most common organism isolated from bile cultures other organisms were Pseudomonas(3.85%) , Klebsiella (2.56%), Coagulase Negative Staphylococcus (1.28%) and Staphylococcus Viridans (1.28%). These findings are similar to as observed in other studies. Majority of the organisms were sensitive to third generation cephalosporins followed by aminoglycoside group of drugs. 1 patients had multi resistant organisms. 03 (15.79%) patients in the bile culture positive group developed post operative wound infection. Only 1 (1.70%) patients in negative bile culture group developed wound infection.

Sensitivity pattern to antibiotics of isolated micro-organisms: Sensitivity Patterns of Cephalosporins and its comparison. In Acharya Suri et al. ⁽⁷⁾ ;The sensitivity of the organisms grown in our analysis of 26 out of 150 patients was tested against cefuroxime, cefoperazone and cefepime, and it was found that sensitivity to third- and fourth-generation cephalosporins was higher as compared to second-generation cephalosporins in acute as well as chronic cholecystitis. The resistance to second-generation cephalosporins has increased while third- and fourth-generation cephalosporins show better promise and may be used as the first line of preoperative prophylaxis in operations for Cholecystectomy.

In this study, The sensitivity of the organisms grown in our analysis of 19 out of 78 patients was tested against Amikacine, Gentamycin, Cefotaxime, Cefepime, Meropenem, Cefoparazone+Sulbactam, Piperacilin+Tazobactam, Levofloxacin and it was found that sensitivity to third- and fourth-generation cephalosporins (Cefoparazone, Cefepime) was higher as compared to aminoglycoside in acute as well as chronic cholecystitis. In this study Levofloxacin (Fluoroquinolones) also shows good sensitivity against isolated organism from bile. Piperacilin and tazobactam (Ureidoopenicillins) also shows good sensitivity against isolated organism from bile and they are more effective against pseudomonas. The resistance to second-generation cephalosporins and aminoglycoside has increased while third generation, fourth generation cephalosporins and Levofloxacin (Fluoroquinolones) show better promise and may be used as the first line of preoperative prophylaxis in operations of cholecystectomy.

Conclusion

Following conclusion were drawn in our study

- In vast majority of patients the bile was sterile, Patients with negative bile culture were 59 (75.64%).
- Escherichia coli was the most common isolated bacteria Among positive bile culture group (63.16%) and 15.38% Among all patients.
- Other organisms isolated were pseudomonas (3.85% Among all patients), Klebsiella (2.56% Among all patients), Coagulase Negative Staphylococcus and Staphylococcus Viridans (1.28% Among all patients).
- The third generation, fourth-generation cephalosporins and Levofloxacin (A Fluoroquinolones) show better promise and may be used as the first line of preoperative prophylaxis in operations for Acute and Chronic cholecystitis patients undergoing Cholecystectomy Surgery.
- Positive bile culture was a more common finding (50% of patients were bile culture positive) in patients with acute cholecystitis in this study. Although this difference was statistically not significant because P value is more than 0.005 (P value was=0.1034).
- Post operative wound infection is more common (15.79%) in group of patients with isolated organism from bile. There is a strong correlation between bile culture and wound culture(75%).

Acknowledgement

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List Of Abbreviations

ACC:Acute Cholecystitis
 CCC:Chronic Cholecystitis
 LC:Laparoscopic Cholecystectomy
 LOC:Laparoscopic Converted to Open Cholecystectomy
 OC:Open Cholecystectomy
 E.Coli:Escherichia coli

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