

A.B. Khan¹, S.A. Salati¹, A.B. Khan², B.K.Parihar³

¹Assistant Consultant, Dept of Surgical Specialities, King Fahad Medical City, Riyadh` KSA.

²Surgical Registrar, SKIMS , SOURA Srinagar, India

³Head Department of Surgery, Government Medical College Jammu, India.

Correspondence To: Dr. Arshad Bashir Khan, Assistant Consultant
Department of Surgical Specialities, King Fahad Medical City, Riyadh
KSA-11525. Email:arshadrg007@yahoo.com Mobile:00966507797857

Background: Biliary tract disease is very common in Indian side of Jammu and Kashmir and because of this we got prompted to undertake a study to find out the association between presenting features and bactibilia.

Methods: The study was conducted prospectively on 121 patients of various biliary tract disorders in the Department of Surgery, Government Medical College Jammu, India prospectively over a period of one year from March 2003-February 2004. Details of age, history, radiology, operation and postoperative course were noted and information obtained recorded on special forms. All the patients included in this study were given single shot of preoperative antibiotic at the induction of anaesthesia and antibiotics modified postoperatively on the culture report.

Results; After recording the observations made while managing these patients of biliary tract disease the results were analysed and entered into the individual patient's protocol. The age range of our series of 121 patients was 15-74 years with a mean age of 42.5 years. There were 100 females and 21 males with male to female ratio of 4.7:1. Abdominal pain was the commonest symptom (98.3%) followed by dyspepsia (47.1%), abdominal lump (16.5%), jaundice (9.9%) and cholangitis (3.3%). Most of the patients had more than one presentation. Out of 121 cases 42(34.7%) turned out to be culture positive and rest of the cases turned out to be culture negative. In culture positive cases aerobes were identified in 38 (34.1%) and anaerobes were identified in 10 (8.2%) patients. The results of gram stain of bile were compared with bile cultures. Out of 121 patients gram staining was positive in 51(42.1%) patients. Among these patients bile cultures were positive in 42 (82.3%) patients.

Conclusion: It was concluded that the presence or absence of bactibilia can predict possibility to stratify patients in low and high risk group so that necessary pre-emptive measures be taken.

Introduction

Aspiration and culture of bile at the time of biliary surgery provides a unique opportunity to study biliary bacterial flora as this may have diagnostic ,prognostic or therapeutic implications¹. Under normal conditions bile, biliary tree and liver are sterile^{2,3} but it becomes colonised in biliary tract disease. The precise origin of biliary bacteria is not known. The possible explanations being ascending infection from duodenum, lymphatic spread and vascular spread from portal venous or hepatic arterial flow or seeding from chronically inflamed gall bladder⁴. The micro organisms most often implicated in infected bile include *Escherichia coli*, *Klebsiella* and *Enterococcus* species along with gram positive cocci and anaerobes like anaerobic streptococci ,*Bacteriodes fragilis* and *Clostridium welchii*^{5,6}. The reported incidence of bacteria in bile is highly variable ranging from 8% -42%^{7,8,9}. The reasons for these enormous variations include type of patients being studied and use of antibiotics preoperatively. The single dose of preoperative antibiotics does not render the bile sterilize¹⁰. Host of previous studies suggest that biliary surgery is attended by high risk of septic complications^{11,12}. The aim of our study was to identify the bacteria present in bile in biliary surgery and try to find the risk factors predictive of bactibilia.

Material and Methods

This prospective study was conducted in the Department of Surgery, Government Medical College Jammu, India over a period of one year from March 2003-February 2004 and included 121 patients undergoing various biliary procedures who gave an informed consent. Patients in study included those with:

- Previous acute cholecystitis
- Previous acute pancreatitis
- Obstructive biliopathy due to carcinoma or stones
- Cholelithiasis

Details of age, history, radiology, operation and postoperative course were recorded in special forms.

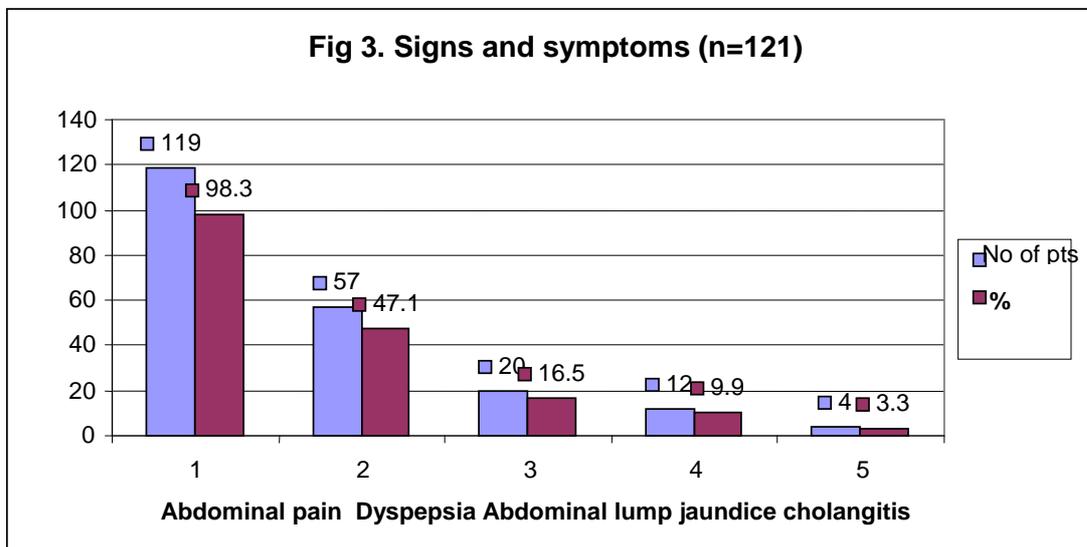
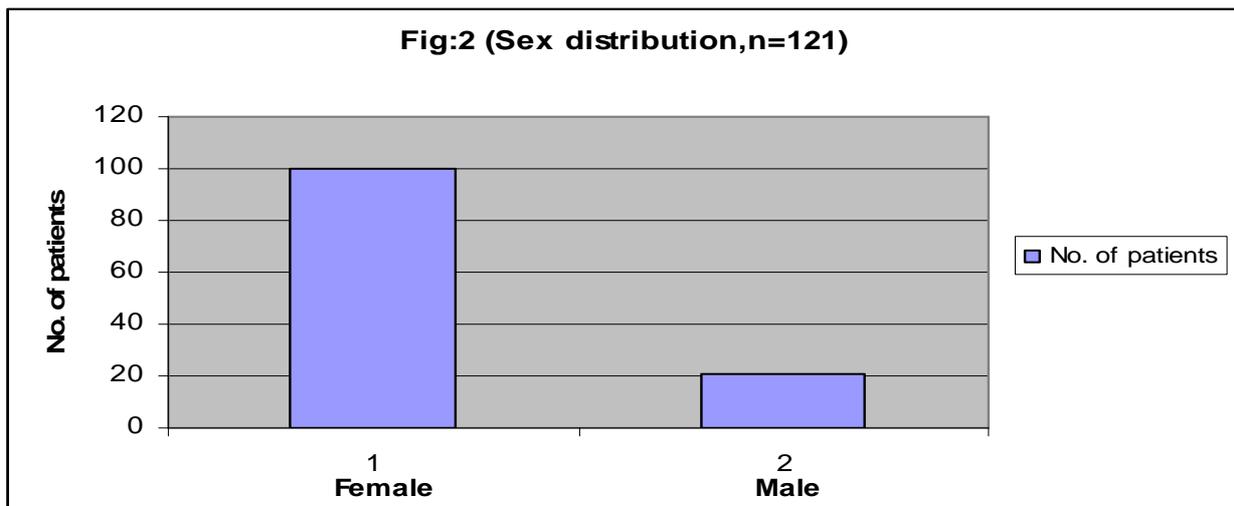
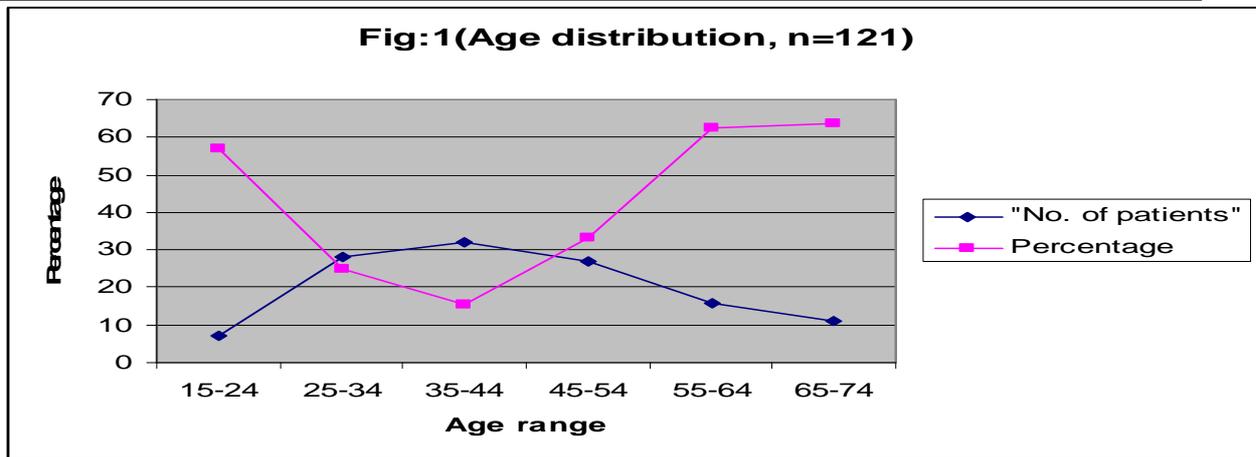
In open cholecystectomy the bile was aspirated into a syringe after removal of the viscus while in laparoscopic procedures it was bile aspirated percutaneously by spinal needle after scope was introduced. Bile from common bile duct was obtained by direct puncture. About 3-5ml of bile was drawn with needle, immediately capped and samples transported immediately to microbiology laboratory for analysis.

Direct smears of the samples were stained by gram staining (Hi media) and examined under microscope. For aerobic cultures, samples were inoculated on Sheep blood agar and MacConkeys agar medium and incubated at 37 degree centigrade overnight. For anaerobic cultures, samples were inoculated on neomycin containing blood agar medium. A disc containing metronidazole was placed over primary inoculation and the plate was then placed in gaspak jar (Hi media) and incubated at 37 degree centigrade for 48 hours. The growth on the media was identified by colony characteristics and standard biochemical identification scheme (Stokes). Post operative cultures in patients with T tubes were repeated on 4th-7th day. All the patients included in this study were given single dose of preoperative antibiotic at the induction of anaesthesia and antibiotics changed postoperatively depending on the culture and sensitivity report.

Results

The age range of our series of 121 patients was 15-74 years with a mean age of 42.5 years. There were 100 females and 21 males with male to female ratio of 4.7:1. Our patients presented with varied symptoms and signs with abdominal pain the commonest symptom(98.3%) followed by dyspepsia(47.1%), abdominal lump(16.5%), jaundice(9.9%) and cholangitis(3.3%). Most of the patients had more than one presentation.

Our series of 121 patients were distributed in following clinical groups with the largest clinical group being the chronic cholecystitis (58.6%), recent acute cholecystitis (17.3%), obstructive jaundice (9.9%), CBD stones without jaundice (8.2%), recent acute pancreatitis (2.4%) and cholangitis (3.3%). The most common operative diagnosis was cholelithiasis with chronic cholecystitis (58.6%) followed by choledocholithiasis(14.8%). Mucocoele was present in 11 cases (9%), resolving acute cholecystitis in 11 cases (9%), CBD stricture in 4 cases (3.3%) and empyema and gall bladder polyps accounted for 3 cases each, contributing 2.4% each. Gram staining of the bile smears for 121 patients showed organisms in 51 patients (42.1%) where as rest of the patients did not show any organism. Out of 121 cases 42 (34.7%) turned out to be culture positive and rest of the cases turned out to be culture negative. In culture positive cases aerobes were identified in 38 (34.1%) and anaerobes were identified in 10 (8.2%) patients. These organisms occurred in combination in various patients. The distribution of various organisms among culture positive patients is depicted below in the Table 2.



- Incidence of bactibilia as per age is shown in Figure 5.
- We examined the incidence of bactibilia in different clinical groups in these 121 cases. Highest incidence of culture positivity was in patients with cholangitis (75.0%). The distribution in different groups is depicted in Table 3.

We examined the incidence of bactibilia as per operative diagnosis in these patients. Highest incidence of positive cultures was found in patients with operative diagnosis of resolving acute cholecystitis (81.8%) followed by bile duct stricture (75%). The distribution is shown in Table 4.

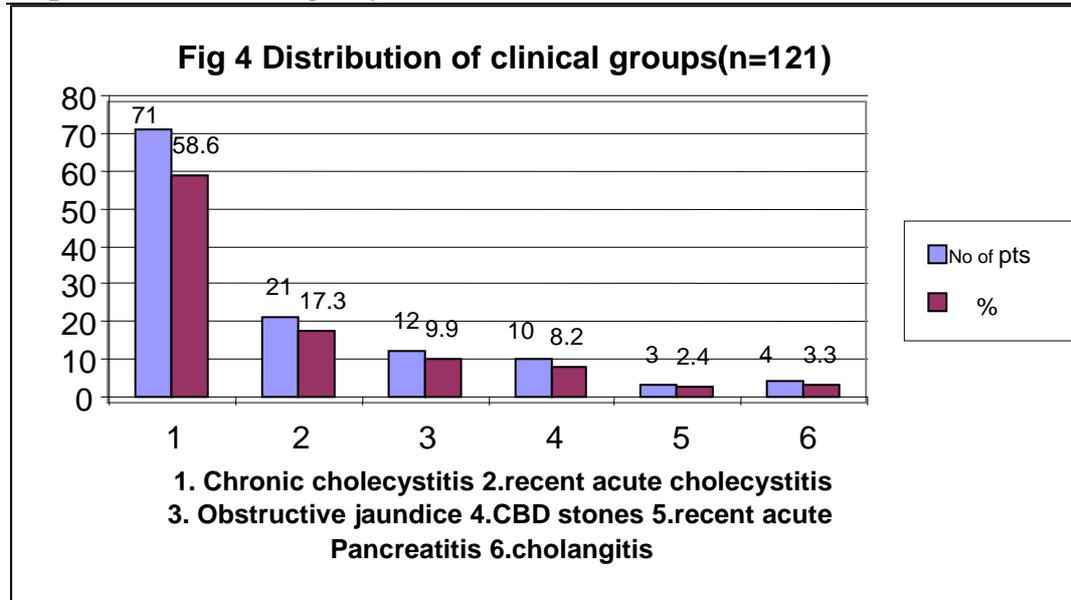


Table 1. Organisms isolated (n=121)

Organisms	No. of patients	Percentage
1.Aerobes	38	31.4
2.Anaerobes	10	8.2
<i>a. mixed with aerobes</i>	6	4.9
<i>b.pure anaerobes</i>	4	3.3

Table 2. Organisms Recovered in 42 Culture Positive Patients

Aerobes	No. of patients	Percentage
1E coli	17	40.4
2.Enterococcus	8	19.0
3.Klebsiella	6	14.2
4.Proteus	2	4.7
5.Citrobacter	2	4.7
6.Staphylococcus	2	4.7
7.Yeast	1	2.3
Anaerobes	No. of patients	Percentage
1.Bacteroides fragilis	6	14.2
2.Anaerobic streptococcus	2	4.7
3.Clostridium welchii	2	4.7

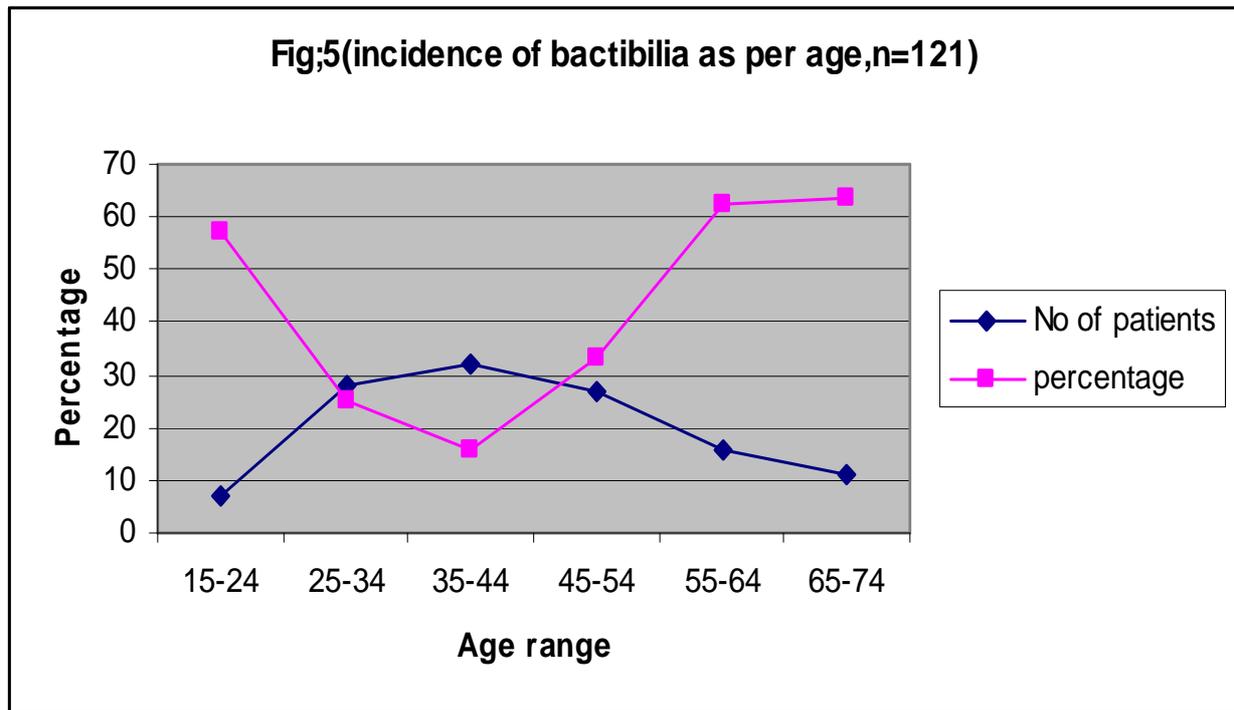


Table 3. Incidence of Bactibilia as Per Clinical Groups (n=121)

Clinical group	No. of Patients	Positive Bile cultures	Percentage
Cholelithiasis	71	14	19.7
Recent acute Cholecystitis	21	10	47.6
Obstructive Jaundice	12	8	66.6
CBD stones Without Jaundice	10	6	60.0
Recent acute Pancreatitis	3	1	33.3
Cholangitis	4	3	75.0
Total	121	42	34.7

- The results of gram stain of bile were compared with bile cultures. Out of 121 patients gram staining was positive in 51(42.1%) patients. Among these patients bile cultures were positive in 42 (82.3%) patients. However in 9 patients (17.6%) no organisms were cultured. Gram staining of bile correctly identified organisms in 37(72.5%) cases, where as in 5 cases (9.8%) it was not able to correctly identify organism.

Table 4. Incidence of bactibilia as per operative diagnosis, (n=121)

Operative Diagnosis	No. of Patients	Positive Bile Cultures	
		No. of Patients	Percentage
Chronic cholecystitis	71	14	19.7
Choledocholithiasis	18	11	61.1
CBD stricture	4	3	75.0
Resolving acute cholecystitis	11	9	81.8
Mucocoele	11	3	27.2
Empyema	3	2	66.0
Gallbladder polyp	3	0	00.0

Discussion

The main goal of this study was to assess the prevalence of micro organisms in bile in patients suffering from various biliary tract diseases so that a categorisation of the patients could be done as to who require prophylactic antibiotics. As evident from the results of the study the overall prevalence of positive bile cultures was 34.7% and the most common isolate grown from bile was Ecoli (40.4%) followed by Enerococcus faecalis (19.0%) and Klebsiella (14.2%). Anaerobic yield was 8.2% with the most common isolate being Bacteriodes fragilis. Our findings concur with the previous findings^{10,13,14,17} but number of other studies are at variance with the results of our study^{15,16} and this difference could be explained by different patient characteristic of study population

The use of prophylactic antibiotic at the time of induction of anaesthesia in the present study did not sterilize the bile and did not reduce the incidence of bactibilia as the yield of micro organisms is similar to host of other studies^{18,19,20}. In present study gram staining correctly identified the presence and type of organisms in 72.5% of cases where as in 5% cases it was not able to identify organism correctly this is as per the previous study¹⁴ but at discord with another study²¹ which showed poor correlation between microscopy and culture. The result of the present study strengthen our belief that if significant number of bacteria are present in bile, they should be identified readily on uncentrifuged sample of bile where they may also be more easily distinguished from epithelial debris. We believe that the concept of using gram staining of bile should be similar to the concept of using frozen sections in breast disease. This has lent us to the belief that Intraoperative bile smears may be used as an early indicator to institute appropriate antibiotic therapy at the time of surgery²².

The results of the present study lend us to the view that preoperative characteristics of the patients are highly predictive of Intraoperative bactibilia. In present study the incidence of bactibilia was significantly higher in certain clinical groups which could be categorised as age>65, recent acute cholecystitis, common duct stones with and with out jaundice, recent acute pancreatitis and cholangitis. This is in accordance with number of previous studies^{10,16,23,24,25}. So we consider it worthwhile to divide patients with one or more of these risk factors in high risk category and patients without these risk factors in low risk category. This has a clinical significance as in high risk group bile cultures are frequently positive where as in low risk group they are infrequently positive. Thus if high risk factors are identified preoperatively appropriate antibiotics could be instituted preoperatively.

In accordance with previous studies^{8,15,26} we found that the operative findings provide a useful guide as to likelihood of bactibilia. In a previous study⁸ biliary sepsis was common in patients with jaundice and stones of common bile duct but unusual in stones confined to gall bladder undergoing elective cholecystectomy. In addition to this in the present study we got higher percentage of positive bile culture in resolving acute cholecystitis (81.8%), empyema (66.6%) and CBD stricture (75%).

Conclusion

In conclusion patients with biliary tract disease harbour bacteria in their bile. Patients could be stratified according to the presence of risk factors such as age >65, recent acute cholecystitis, recent acute pancreatitis, CBD stones with or without jaundice, CBD stricture and cholangitis into high risk category and in absence of these risk factors into low risk category. Low risk category patients should receive limited prophylactic antibiotics whereas high risk patients should receive full therapeutic antibiotics to reduce the incidence of postoperative sepsis. Intraoperative gram staining is an important tool to identify presence of bactibilia early before the results of culture are available and the culture should be done routinely and antibiotics modified according to the sensitivity report.

References

1. Piersol CM, Bokus HL. A study of bile obtained by non surgical biliary drainage, with special reference to bacteriology. *Am J M Science* 1923;165:486
2. Csendes A, Fernandez M, Uribe P. Bacteriology of gall bladder bile in normal subjects. *Am J Surgery* 1975; 129:629-631.
3. Scott AJ. Progress report: Bacteria and diseases of biliary tract. *Gut* 1971; 12:487-492.
4. Scott AJ, Khan GA. Origin of bacteria in bile duct bile. *The Lancet* 1967; October 14:790-791.
5. Keighly MRB, Drysdale RB, Burdon DW et al. Antibiotic treatment of biliary sepsis. *Surgical clinics of North America* 1975; 55:1379-1390.
6. Zakko SF, Afdhal NH. Clinical features and diagnosis of acute cholecystitis. *Up to date: Gastroenterology and Hepatology* 2002; 11.1:1-3.
7. Anderson RE, Priestly JT. Observations on bacteriology of choledochal bile. *Annals of Surgery* 1951; 133:486-489.
8. Edlund YA, Mollstedt PO, Ouchterlony. Bacteriological investigations of the biliary system and liver in biliary tract disease correlated to clinical data and microstructure of gall bladder and liver. *Acta Chir Scand* 1959;116:461-467
9. Flemma RJ, Lewis M, William W et al. Bacteriologic studies of biliary tract infection. *Annals of Surgery* 1967; 563-570.
10. Chetlin SH, Elliot DW. Biliary bacteria. *Arch Surg* 1971; 102:303-307.
11. Cox JL, Helfrich LR, Pass ILL et al. The relationship between biliary tract infection and postoperative complication. *Surg Gynaecol Obstet* 1978; 146:233-236.
12. T Nomura, Y Shirai, K Hatakeyama. Impact of bactibilia on the development of postoperative abdominal septic complications in patients with malignant biliary obstruction. *Int Surg* ; 84(3):204-208.
13. Pyrtek LJ, Bartus SH. Clostridium welchii infection complicating biliary tract surgery. *New Eng J Medicine* 1962; 266:689-693.
14. Keighly MRB, Mc leish AR, Bishop M et al. Identification of presence and type of biliary micro flora by immediate gram stains. *Surgery* 1977; 81:469-472.
15. Pitt HA, Postier RG, Cameron JL. Biliary bacteria: significance and alteration after antibiotic therapy. *Arch Surg* 1982; 117:455-459.

16. Morris-stiff G.J, ODonohue P.O, Ogunbyi S, Sheridan W.G. Microbiological assessment of bile during cholecystectomy: Is all bile infected? Journal of the international Hepato Pancreato Biliary Association (HPB) 2007;9:225-228.
17. Dellikaris P.G, Michail P.O, Klonis G.D et al. Biliary bacteriology based upon Intraoperative bile cultures. Am J Gastroenterol 1997; 68: 51-55
18. Chetlin S.H, Elliot D.W. Preoperative antibiotics in biliary surgery. Arch Surg 1973; 107: 319-323.
19. Bergen T, Dobloug I, Liarag I. Bacterial isolates in cholangitis and cholelithiasis. Scand J Gastroent 1979; 14: 625-631.
20. Pitt H.A, Course N.R. Biliary sepsis and toxic cholangitis. In Moody F.G(Ed) Surgical treatment of digestive diseases 1990; 2nd edition. Year book medical publishers, Chicago: 332-350.
21. Gallagher P, Ostick G, Jones D et al. Intraoperative microscopy of bile, is it useful. Br J Surg 1982; 69: 473-474
22. Beinfield M.S, Hayes R.L. Use of Intraoperative gram staining during cholecystectomy. Am J Surg 1979; 137: 773-774.
23. Lewis R.T, Godall R.G, Marien B et al. Biliary bacteria, antibiotic use and wound infection in surgery of gall bladder and common bile duct. Arch Surg 1987; 122; 44-47.
24. Wells G.R, Taylor W, Lindsay G et al. Relationship between bile colonisation, high risk factors and postoperative sepsis in patients undergoing biliary tract operations while receiving prophylactic antibiotics. Br J Surg 1989; 76; 374-377.
25. Targorana E.M, Carls M.R, Roset F et al. Single dose antibiotic prophylaxis in patients at high risk for infection in biliary surgery: a prospective and randomized study comparing cefonicid and mezlocillin. Surgery 1990; 107: 327-334.
26. Wolloch Y, Feigenberg Z.V, Zer M et al. Influence of biliary infection on postoperative course after biliary tract surgery. Am J Gastroenterol 1977; 67: 456.