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Original Research Article

Clinical profile and pattern of bacterial isolates from bile aspirates of cholecystectomy patients

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ABSTRACT

Background: Antibiotic policy and appropriate antibiotic prophylaxis cannot be designed unless data is available about bacteria colonizing the bile associated with gall bladder disease. Authors aim was to assess the clinical profile and pattern of bacterial isolates from bile aspirates of cholecystectomy patients seeking care at a tertiary care teaching hospital.
Methods: Patients who underwent cholecystectomy for various hepatobiliary ailments during year 2017-18 formed the study population. Bile aspirates were collected during cholecystectomy and sent to the microbiology laboratory. Ultrasonography, computed tomography and MRCP were done to confirm the gall bladder pathology before surgery.
Results: Out of total eighty-six patients, bacterial growth was observed in 28 (32.56%) subjects. As per division of bile samples, maximum number of study participants (39.29%) having bactibilia were seen in Group II. Group with second highest number of patients showing bactibilia was Group III with 9 subjects (32.14%). Eight subjects (28.57%) exhibited bacterial growth in bile aspirates in Group I subjects. Three patients (10.71%) showed bile infected with multiple bacteria i.e. polymicrobial infection. The gram-negative preponderance was seen in all the three groups with Escherichia coli being most common in group I and II. Pseudomonas aeruginosa was isolated in majority of the patients in group II and III.
Conclusions: It is advised that all patients undergoing cholecystectomy must have their bile aspirated during cholecystectomy and sent for microbiological examination and culture. It will help in choosing appropriate antibiotic to prevent infection.

Keywords: Bacteria, Bile, Bacteriobilia, Cholecystectomy, Gall bladder

INTRODUCTION

Probably the most commonly seen biliary pathology is related to gall stones both in developing and developed world. Disorders of gall bladder are very frequently encountered in clinical practice. Patients present with various forms like acute, chronic cholecystitis and others. Microbial flora in disease associated with Gall Bladder has been detected in up to 80% of the subjects presenting with acute symptoms and in >25% patients with chronic disease of Gall bladder.[1,2]

If bile flow is normal, then presence of bacteria in the biliary system have no clinical significance. Bacterial colonization of the bile associated with gallstones anywhere in the biliary tree is clinically very important.

Gram negative bacteria are more commonly associated with GB disorders in India.[3-5] Reports from tertiary care teaching health institutions have indicated towards polymicrobial and mixed infection.[6,7] Gram- positive and anaerobic are less commonly reported organisms. Viral and fungal agents are rare.[8]

The microbial flora probably translocate into the circulation leading to systemic infection. Antibiotic policy and appropriate antibiotic prophylaxis cannot be designed unless data is available about bacteria colonizing the bile associated with gall bladder disease. Thus, it becomes important to know the microbiological flora of the gallbladder before prophylactic antibiotics are given. Objective of this study was to assess the clinical profile and pattern of bacterial isolates from bile aspirates of cholecystectomy patients seeking care at a tertiary care teaching hospital.

METHODS

This hospital-based study was planned and executed by departments of microbiology, radiology, general surgery and community medicine of a tertiary care teaching institute of northern India. Patients who underwent cholecystectomy for various hepatobiliary ailments during year 2017-18 formed the study population. Patients undergoing cholecystectomy and those consenting for this study were included. Exclusion criteria were patients' age less than 18 year, patients with any known source of sepsis, patients with history of ascending cholangitis, patients with preoperative diagnosis of empyema of gallbladder, patients who had undergone endoscopic retrograde cholangiopancreatography (ERCP).

Baseline information, demographic profile and clinical examination of eighty-six eligible patients were studied and noted in a proforma. Detailed history was taken with physical examination and investigations were done. Ultrasonography, computed tomography and MRCP were done to confirm the gall bladder pathology before surgery. Complete blood count, urine and serum biochemistry, and chest X-ray were done. An ultrasonogram of the upper abdomen was performed in all study subjects to identify the site of obstruction, number of stones and condition of gallbladder. A CT-scan was also done in subjects if carcinoma was suspected. The open or laparoscopic cholecystectomy was performed as per patient condition. Bile aspirates were collected during cholecystectomy and sent to the microbiology laboratory in sterile syringe wrapped in a sterile gauze within an hour of collection at room temperature, 3cc bile was aspirated with use of sterile 20 No. spinal needle with 10cc sterile syringe in laparoscopic cholecystectomy. After inserting all 3 ports and before starting dissection, sample was aspirated and sterile 10cc syringe with 20 number needles in open cholecystectomy. This collected bile from gallbladder before cholecystectomy was transported to the laboratory in sterile test-tube.

Microbiological examination of bile was performed. Resected gallbladder and stone were subjected to histopathological examination. Standard microbiological procedures were adopted for processing the samples. For isolation, 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.

For the analysis purpose, subjects from whom bile samples were collected were divided in three groups. Group I included acute cholecystitis with or without cholelithiasis, Group II included chronic cholecystitis with or without cholelithiasis and Group III included other hepatobiliary ailments requiring cholecystectomy viz. gallbladder carcinoma, acute emphysematous gall bladder and mucocele gall bladder. The study adhered to the tenets of the Declaration of Helsinki for research in humans. All the proforma were checked and then coded for computer entry. After compilation of collected data, analysis was done using EpiInfo software and valid conclusions were drawn.

RESULTS

In this study, data of eighty-six patients was collected and analyzed. Mean age of study subjects was 46.18±11.85 years with range of 18 to 73 years. Female subjects (74.4%, n=64) outnumbered male subjects (25.6%, n=22) with a female to male ratio of 2.9:1. Minimum of one risk factor was observed in sixty-nine (80.2%) of the total 86 subjects. Almost all the study subjects reported about presence of biliary colic presenting from as short as 2 days and ranging to three and half years. Most common clinical presentation of study subjects was tenderness ranging from mild to severe, which was graded from 1+ to 3+. Grade 1+ tenderness was seen in 37(43.02%), grade 2+ in 25(29.07%) and grade 3+ in 03(3.49%) subjects.

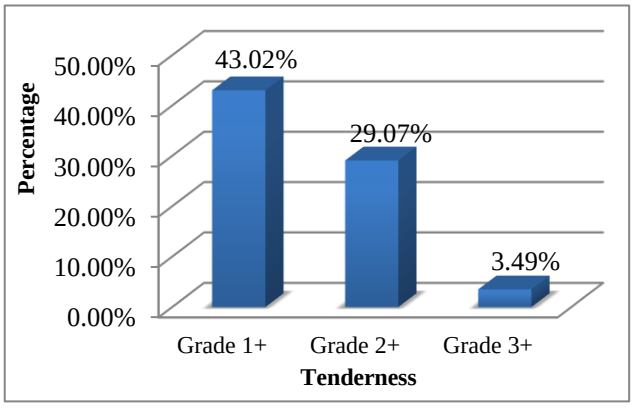

Figure 1: Distribution of most common clinical presentation among study subjects.

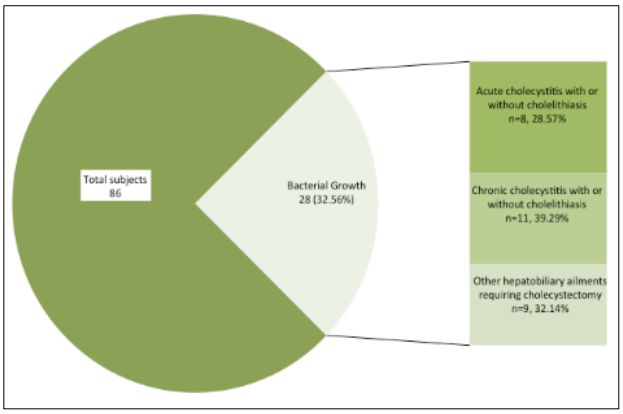

Figure 2: Group wise distribution of bacterial growth in bile aspirates.

Out of total eighty-six patients, bacterial growth was observed in 28 (32.56%) subjects. As per division of bile samples, maximum number of study participants (39.29%, n=11) having bactibilia were seen in Group II. Group with second highest number of patients showing bactibilia was Group III with 9 subjects (32.14%). Eight subjects (28.57%) exhibited bacterial growth in bile aspirates in Group I subjects. Three patients (10.71%) showed bile infected with multiple bacteria i.e. polymicrobial infection.

The gram-negative preponderance was seen in all the three groups with Escherichia coli being most common in group I and II. Pseudomonas aeruginosa was isolated in majority of the patients in group II and III (Table 1).

Table 1: Pattern of distribution of bacterial isolates in various samples of bile aspirates.

Bacteria in bile aspirates	Group I Acute cholecystitis with or without cholelithiasis	Group II Chronic cholecystitis with or without cholelithiasis	Group III Other hepatobiliary ailments requiring cholecystectomy
N=31 (28+3*)	N=9 (8+1*)	N=13 (11+2*)	N=9
Escherichia coli	4	6	3
Klebsiella oxytoca	1	4	2
Pseudomonas aeruginosa	1	5	3
Citrobacter freundii	-	1	-
Acinetobacter baumannii	-	-	2
Enterobacter aerogenes	2	1	-
Staphylococcus epidermidis	1	2	1
Total	9	19	11
*polymicrobial infection			

DISCUSSION

The most common reason for a cholecystectomy is gallbladder stones. However, the presence of gallstones within either the gallbladder or biliary tree is associated with the bacterial colonization of the bile. In patients without gallbladder stone disease, previous biliary intervention is associated with high rates of bacteriobilia. Ascending infection from the duodenum is thought to be the primary mechanism by which bacteria enter the bile. 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.

Acute cholangitis spans a continuous clinical spectrum and can progress from a local biliary infection to advanced disease with sepsis and multiple organ dysfunction syndrome. Different microbes in the bile may be cause to post- cholecystectomy infections. Positive bile cultures are significantly more common in elderly (>60 years) patients with symptomatic gallstones than in younger patients (45% versus 16%).[9] Studies document a strong association between the presence of bacteria in bile cultures taken at surgery and the occurrence of subsequent infection.[10]

In our study, mean age of study subjects was 46.18±11.85 years with range of 18 to 73 years. In Alaattin Öztürk et al, study a total of 114 patients were included in this study.[11] Ages ranged from 17 to 86 years with a mean age of 48.6±16.8. We observed in our study that female subjects (74.4%, n=64) outnumbered male subjects (25.6%, n=22) with a female to male ratio of 2.9:1. Similar findings have been observed in prior studies conducted in North India and Pakistan.[12,13]

In this study, bacterial growth was observed in 28 (32.56%) subjects. As per division of bile samples, maximum number of study participants (39.29%, n=11) having bactibilia were seen in Group II. Group II included subjects with chronic cholecystitis with or

without cholelithiasis. Findings of this study are similar to observations of another study from North Carolina on chronic cholecystitis patients.[14] However, other studies from other countries have reported higher rates of isolation in acute cholecystitis patients as well.[15,16] It could be due to bacterial overgrowth in gallbladder deformed and fibrosed by repeated infections and healing. Such organ favors bacteria to persist and grow especially when repeated ineffective doses of antibiotics are given to these patients during acute exacerbations.

We observed preponderance of gram-negative bacteria in all the three groups with Escherichia coli being most common in group I and II. Pseudomonas aeruginosa was isolated in majority of the patients in group II and III. The significance of *E. coli* dominance is also supported by previous reports indicating a potential role for *E. coli* glucoronidase enzymatic activity in formation of calcium bilirubinate gall stone.[12,15]

In this study, bacterial growth was observed in 28 (32.56%) subjects. Van Leeuren PA et al, study total 840 patients from which 138 patients showed positive bile culture (16.4%) and 72 patients showed wound infection (9%).[17] Bile culture showed *E. coli* was most common organism isolated (36%). But there was no correlation between positive bile culture and subsequent wound culture. Valazquez-Mendoza JD et al, study total 80 patients' study, 40 patients with bile culture positive and 40 patients with wound culture positive.[18]

In this investigation, 10.71% subjects showed bile infected with multiple bacteria i.e. polymicrobial infection. The association of polymicrobial infection with obstruction due to cholelithiasis or previous manipulation of the biliary tract has been widely reported by various authors.[19,20]

CONCLUSION

Bacterial growth was observed in the bile of 32.56% subjects. Maximum number of study participants (39.29%) having bactibilia were seen in subjects with chronic cholecystitis with or without cholelithiasis. The gram-negative preponderance was seen in all the groups. It is advised that all patients undergoing cholecystectomy must have their bile aspirated during cholecystectomy and sent for microbiological examination and culture. It will help in choosing appropriate antibiotic to prevent infection.

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Ethical approval: The study was approved by the Institutional Ethics Committee

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