A Prospective Study of Bacteriological Profile of Bile in Patients with Biliary Stones

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Abstract: INTRODUCTION: Cholelithiasis is the most common digestive tract disorder. Treatment of acute episodes requires antibiotics. But pathogens and sensitivity of the pathogens to antibiotics vary widely. Bacteriological profile helps to prevent bacterial septicemia. This study was conducted to study the frequency of bacterial growth in bile samples from patients undergoing surgery for bile stones, identify the common microorganisms which are found in the bile samples from patients undergoing surgery for bile stones & to analyze the antibiotic sensitivity pattern of the microorganisms cultured.

METHODS: prospective study in a sample of 60 patients with cholecystitis who underwent cholecystectomy from June 2015 to August 2016. A structured questionnaire was used to collect patient details and symptoms. Microbiological and radiological investigations were done. Intraoperatively bile was collected and culture & sensitivity testing was done. Results were analysed using SPSS software.

RESULTS: majority of the patients were elderly female with abdominal pain as presenting symptom and tenderness in right hypochondrium as major clinical sign. 21% of the sample was culture positive with gram negative enterobacteriaceae as major organism and maximum sensitivity to aminoglycosides.

CONCLUSION: Culture of the bile showed gram negative enterobacteriaceae mainly klebsiella and E.coli. These bacteriae were sensitive to Aminoglycosides & other broad spectrum antibiotics.

KEYWORDS: Amino glycosides, Bacteriological profile, Cholelithiasis, Gram negative enterobacteriaceae, Klebsiella, Septicaemia.

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I. Introduction:

Cholelithiasis is the most common gastro-intestinal condition requiring hospitalization. With an overall prevalence of 10-15%, geographic specific prevalence is from 0-10% in Africans to 60-70% in Pima Indians. Even though the treatment of choice in cholelithiasis is surgical decompression of biliary system and extraction of stones, the role of antibiotics for the control of biliary infection is undisputed. Development of resistant strains and decreased susceptibility of gram negative bacteria and anaerobes to antibiotics have made the treatment of biliary tract infections difficult. This points to the importance of culture of bile at the time of cholecystectomy, so that sensitive antibiotics can be given post-operatively to prevent gram negative septicemia which can be fatal.

Objectives:
(1) To study the frequency of bacterial growth in bile samples from patients undergoing surgery for bile stones
(2) To identify the common microorganisms which are found in the bile samples from patients undergoing surgery for bile stones
(3) To analyze the antibiotic sensitivity pattern of the microorganisms cultured

II. Methods:

(a) Study type: Prospective study
(b) Study setting:
• Department of General surgery and Surgical gastroenterology, Government Mohan Kumaramangalam Medical college hospital, Salem, Tamilnadu
• Department of Microbiology, Government Mohan Kumaramangalam Medical college hospital, Salem, Tamilnadu
(c) **Study period:**
Eighteen months – from June 2015 – August 2016

(d) **Research subjects:**
Inclusion criteria:
All patients with biliary stones who are undergoing surgery for the same
Exclusion criteria:
Patients who are not willing to take part in the study

(e) **Sampling:**
Sample size: 60 patients with cholelithiasis.
Sampling technique: Every consecutive patient eligible for the study is included.

(f) **Data management and analysis:**
Data was analysed using SPSS software version 7. Chi-square test was used for analysing the variables

(a) **Structured questionnaire** which includes the following
Variables:
1. **Patient details** including sociodemographic data and details of hospital admission
2. **Details of presenting complaints**
3. **Past history** of similar illness
4. **Family history** of biliary stones
5. **Clinical examination findings** like jaundice, murphy’s sign, palpable gallbladder
6. **Investigation reports** of Liver function tests, X Ray abdomen, ultrasound scan of abdomen

**Microbiological study tool:** 2 ml bile samples were collected in sterile capped bottles from the gallbladder of all patients intraoperatively and sent for culture and sensitivity.

**ETHICAL CLEARANCE:** The study was conducted after obtaining ethical committee clearance from the institutional ethical committee

### III. Results:

**Sex Distribution**
Out of 60 patients studied 94 were females (69.6%) and 41 were males (30.4%)

Male: female ratio is **1:2.3**

![Sex Distribution Chart](image)

**Fig 1 sex distribution**
Age Distribution
In this study the youngest patient was 25 years of age and the oldest patient was 78 years of age. Maximum prevalence was in the 40-49 year age group (29.6%) 

<table>
<thead>
<tr>
<th>Age group</th>
<th>No. of patients</th>
<th>Percentage</th>
</tr>
</thead>
<tbody>
<tr>
<td>20-29</td>
<td>6</td>
<td>10.4%</td>
</tr>
<tr>
<td>30-39</td>
<td>12</td>
<td>19.3%</td>
</tr>
<tr>
<td>40-49</td>
<td>18</td>
<td>30.3%</td>
</tr>
<tr>
<td>50-59</td>
<td>13</td>
<td>22.2%</td>
</tr>
<tr>
<td>60-69</td>
<td>9</td>
<td>14.8%</td>
</tr>
<tr>
<td>70-79</td>
<td>2</td>
<td>3.7%</td>
</tr>
</tbody>
</table>

Locality
Out of the 60 patients 36 were from urban areas (59.3%) and 24 were from rural areas (40.7%). Gall stone disease is more prevalent in the urban areas and the rising trend in the rural areas may be attributed to the change in life habits in these areas.

<table>
<thead>
<tr>
<th>Area</th>
<th>No of patients</th>
<th>Percentage</th>
</tr>
</thead>
<tbody>
<tr>
<td>Urban</td>
<td>36</td>
<td>59.3%</td>
</tr>
<tr>
<td>Rural</td>
<td>24</td>
<td>40.7%</td>
</tr>
</tbody>
</table>

Diet
55 patients who took part in the study were non vegetarians (91.1%) and 5 patients were vegetarians (8.9%) clearly showing the increased prevalence of the disease among non vegetarians.

<table>
<thead>
<tr>
<th>DIET</th>
<th>No. of patients</th>
<th>Percentage</th>
</tr>
</thead>
<tbody>
<tr>
<td>NON VEGETARIAN</td>
<td>55</td>
<td>91.1%</td>
</tr>
<tr>
<td>VEGETARIAN</td>
<td>5</td>
<td>8.9%</td>
</tr>
</tbody>
</table>

IV. Socio Economic Status
The disease was found to be more prevalent among the middle and upper class population when compared to the lower class. 23 patients belonged to middle socio economic status (38.5%), 20 patients belonged to upper socioeconomic status (33.3%) and 17 patients to low socio economic status (28.2%).

<table>
<thead>
<tr>
<th>S.E.S</th>
<th>No. of patients</th>
<th>Percentage</th>
</tr>
</thead>
<tbody>
<tr>
<td>upper</td>
<td>20</td>
<td>33.3%</td>
</tr>
<tr>
<td>middle</td>
<td>23</td>
<td>38.5%</td>
</tr>
<tr>
<td>lower</td>
<td>17</td>
<td>28.2%</td>
</tr>
</tbody>
</table>

Presenting Complaints
Majority of patients had abdominal pain as one of their presenting complaints (95.6%), followed by dyspepsia (76.3%) and vomiting (45.9%). Less common symptoms included jaundice (2.9%) and fever (1.5%). 36 patients (60.7%) gave a history of recurrent episodes of one of the above mentioned symptoms which were treated conservatively. 4 patients (6.7%) gave a history of gall stone disease in a first degree relative.

Clinical Examination
33 patients (55.6%) showed some clinical signs suggestive of gall stone disease while the remaining 27 patients (44.4%) were free of any clinical signs at the time of examination. 32 patients had tenderness in right hypochondrium, murphy sign was present in 2 patients and jaundice was present in 2 patients.
Culture Positivity
The study sample was 60. A total of 13 positive cultures were obtained yielding a total culture positivity of 21.5%.

All bacteria isolated were gram negatives. The common bacteria isolated were the coliforms which accounted for more than 90% of the total isolates. The most common bacteria was Klebsiella (48.3%), followed by E.coli (34.5%). Less common bacteria isolated were Enterobacter species (10.3%) and pseudomonas species (6.9%).

<table>
<thead>
<tr>
<th>Organism</th>
<th>Number</th>
<th>Percentage</th>
</tr>
</thead>
<tbody>
<tr>
<td>Klebsiella</td>
<td>6</td>
<td>48.3%</td>
</tr>
<tr>
<td>E.coli</td>
<td>5</td>
<td>34.5%</td>
</tr>
<tr>
<td>Enterobacter sp</td>
<td>1</td>
<td>10.3%</td>
</tr>
<tr>
<td>Pseudomonas</td>
<td>1</td>
<td>6.9%</td>
</tr>
</tbody>
</table>

Table 4 Culture Positivity

<table>
<thead>
<tr>
<th>Antibiotic</th>
<th>Klebsiella</th>
<th>E.coli</th>
<th>Enterobacter</th>
<th>Pseudomonas</th>
</tr>
</thead>
<tbody>
<tr>
<td>Ciprofloxacin</td>
<td>60%</td>
<td>71.4%</td>
<td>100%</td>
<td>100%</td>
</tr>
<tr>
<td>Gentamycin</td>
<td>90%</td>
<td>71.4%</td>
<td>100%</td>
<td>100%</td>
</tr>
<tr>
<td>Ceftriaxone</td>
<td>60%</td>
<td>64.3%</td>
<td>100%</td>
<td>-</td>
</tr>
<tr>
<td>Cefazidime</td>
<td>-</td>
<td>-</td>
<td>-</td>
<td>100%</td>
</tr>
<tr>
<td>Cefoperazone</td>
<td>70%</td>
<td>71.4%</td>
<td>66.6%</td>
<td>50%</td>
</tr>
</tbody>
</table>

Table 5 Antibiotic Sensitivity Test Results
The observations following the culture and sensitivity testing

- The most effective antibiotics against the coliforms were Aminoglycosides-Gentamycin (87%) and Amikacin(80%).
- Ciprofloxacin (77%) and Imipenem(81%) were also found to be effective against coliforms.
- Among the third generation Cephalosporins,Ceftriaxone (75%) and Cefaperazone+sulbactam(73%) were found to be effective.
- Aztreonem (43%), though not very effective against coliforms, was found to be effective against certain resistant Klebsiella spp.
- The Pseudomonas spp. were highly sensitive to Aminoglycosides (Gentamycin/Amikacin) and Fluoroquinolones(ciprofloxacin).
- Among the 3rd gen cephalosporins,Ceftazidime was highly effective.

V. Discussion:
In this study we have obtained a culture positivity rate of 21.5% which is comparable to the previous study conducted by the Department of Surgery,Shahrekord University of medical science Iran, where the culture positivity rate was 20%

Culture positivity rates were in the range of 23-46% in symptomatic gall stone disease in other parts of the world

In surgical practice in our institute, acute cholecystitis is managed conservatively with antibiotics followed by elective cholecystectomy, so in acute cases culture was difficult to obtain. Since antibiotics were startedpreoperatively, it might have reduced our culture positivity rates. There was practical difficulty in culturing the anaerobes, which was a limitation of our study might have affected the positivity rates, though we were able to get 10%, which was obtained in many previous studies.

All bacteria isolated were gram negatives. The commonbacteria isolated were the coliforms which accounted for more than 90% of the total isolates. The most common bacteria was Klebsiella(48.3%),followed by E.coli (34.5%).In the previous study results, it was shown that, among the isolates majority(91%) were gram negative enteric organisms, with Escherichia coli in 40% and Klebsiella in 35% forming the majority.

Aminoglycosides (amikacin,gentamycin) and Ciprofloxacin showed good coverage ranging between 75%-90% against both coliforms and non coliforms.The 3rd generation cephalosporins Ceftriaxone and Cefaperazone+ sulbactam showed good coverage against coliforms while Ceftazidime was highly effective against the non coliforms (pseudomonas)

VI. Conclusion:
- Our study results are comparable with similar studies done around the globe.
- Commonest bacteria associated with gallstone disease were found to be gram negative enterobacteriaceae.
- A combination of Ciprofloxacin, Aminoglycoside and Metronidazole may be started empirically without waiting for bacteriologic cultures
- Ceftriaxone/cfepaperazone+sulbactam or Imipenem can be used in cases of sepsis as cholecystitis is a common cause of intra abdominal sepsis.
- This study shows significant culture positivity and hence underlines the importance of obtaining bile for culture at the time of cholecystectomy. This will help in administering appropriate and sensitive antibiotics to patients, for preventing gram negative septicemia.

References

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