A study of bacteriological profile of ascitic fluid in suspected clinical cases of spontaneous bacterial peritonitis at a tertiary care hospital in India

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Background: Spontaneous bacterial peritonitis (SBP) is the development of a monomicrobial infection of ascitic fluid in the absence of any contiguous source of infection. It occurs most commonly in conjunction with cirrhosis of the liver and alcoholic liver diseases. Majority of the SBP cases are caused by gram-negative organisms, mostly Escherichia coli.

Objective: To isolate the various bacteriological agents from ascitic fluid from clinically suspected cases of SBP and to determine their antibiotic sensitivity pattern.

Materials and Methods: In this study, 217 ascitic fluid samples from clinically suspected cases of SBP were collected from December 2011 to November 2012. Ascitic fluid was collected by bedside tapping in blood culture bottle aseptically and immediately sent to a microbiology laboratory, Sir T Hospital, Bhavnagar, Gujarat, for microbiological examination. Bacterial examination and antibiotic sensitivity tests were carried out by standard microbiological techniques.

Results: Of 217 clinically suspected cases of SBP, 71 (43.80%) had ascitic fluid polymorphonuclear cells (PMN) count ≥ 250/mm³. Among 71 cases, 31 (43.6%) cases were culture positive and 40 (56.4%) cases were culture-negative neutrocytic ascites. From 31 culture-positive cases, E. coli was isolated from 17 (54.9%) cases; Klebsiella spp. was isolated from 5 (16.2%) cases; Staphylococcus aureus was isolated from 6 (19.3%) cases; and Pseudomonas aeruginosa was isolated from 3 (9.6%) cases. All isolates were sensitive to cefotaxime and ceftriaxone.

Conclusion: If diagnosed early, SBP can be treated with high success rate, thus ascitic fluid laboratory analysis including culture of all suspected patients will help in improving prognosis of the patients.

KEY WORDS: Ascitic fluid, spontaneous bacterial peritonitis, cirrhosis
1. Bacterascites (BA): It is defined as ascitic fluid leukocyte count <250/mm³ with positive blood culture.
2. Culture-negative neutrocytic ascites (CNNA): It is defined as ascitic fluid leukocyte count ≥500/mm³ or neutrophil count ≥250/mm³ with negative culture.

In-hospital mortality for the first episode of SBP ranges from 10% to 50%, depending on various risk factors. One-year mortality after a first episode of SBP has been reported to be 31% and 93%. In fact, the occurrence of SBP or other severe bacterial infections markedly worsens the prognosis in patients with cirrhosis, and it has been proposed that a new prognostic stage of cirrhosis not reflected in current staging systems should be defined, the so-called “critically ill cirrhotic.” Patients at this late stage have to be evaluated for the possibility of liver transplantation. It is important to stress in this context that the only factors that are modifiable in this scenario are timely diagnosis and effective first-line treatment. With the early recognition of disease and prompt and appropriate antibiotic treatment, the in-hospital mortality of an episode of SBP has been reduced to approximately 20%.

SBP is often an overlooked complication of cirrhosis. Thus, by the time it is diagnosed, the infection of ascitic fluid has become so severe that it cannot be controlled with standard antibiotic regimens and this leads to high mortality. Considering high morbidity and mortality rates of SBP, there is a need for further exploration to identify and understand SBP in detailed context. The local data on SBP frequency are scanty, and the only factors that are modifiable in this scenario are timely diagnosis and effective first-line treatment. With the early recognition of disease and prompt and appropriate antibiotic treatment, the in-hospital mortality of an episode of SBP has been reduced to approximately 20%.

This study was also conducted to document frequency and variants of SBP, causative organisms in these patients, and to compare the bacteriological profile with that of clinical features of patients with or without SBP. We conducted this study with the aim to isolate the various bacteriological agents from ascitic fluid from clinically suspected cases of SBP and to determine their antibiotic sensitivity pattern.

Materials and Methods

From December 2011 to November 2012, we collected 217 ascitic fluid samples from clinically suspected cases of SBP who were admitted to medical ward of Sir T. General Hospital, Bhavnagar, Gujarat. Patients in this study were of mixed ages and consisted of both men and women; all patients met clinical criteria for suspicion of SBP including fever, ascites, and abdominal pain. Ascitic fluid was collected by bedside tapping in blood culture bottle aseptically using standard and universal precautions to ensure that a sterile sample was collected before delivery to the microbiology laboratory, Sir T. General Hospital, Bhavnagar, for examination. Bacterial examination and antibiotic sensitivity tests were carried out by standard microbiological techniques. PMN count data were collected retrospectively.

Inclusion criterion was adult patients of both genders with clinical cirrhosis of liver with ascites. Exclusion criteria were ascites due to renal, cardiac, tubercular, malignant pathology secondary peritonitis, pregnant women, patients who were not willing to participate in the study, and patients unable to communicate. Statistical analysis was conducted using MS Excel software and p-value <0.5 was considered as statistically significant.

Results

During the entire study period, 217 cases were admitted with cirrhotic ascites. Out of these, 71 (32.7%) cases were suspected having SBP. Out of these 71 cases, 31 (43.6%) cases were culture positive and 40 (56.4%) cases were CNNA. Frequency of SBP was maximum in the age group of 50–59 years followed by that of 60–69 years whereas SBP was more common in males than females. The total number of males with SBP was 49 (69%) while that of females with SBP was 22 (31%) [Table 1]. Among the total 31 culture-positive cases, men were 23 (74.1%) and women were 8 (25.9%). The culture-positive SBP was more common in age group of 60–69 years followed by that of 50–59 years, and the number of men was more than that of women [Table 2].

Of the 31 culture-positive cases, *Escherichia coli* was isolated from 17 (54.9%) samples; *Klebsiella* spp. was isolated from 5 (16.2%); *Staphylococcus aureus* was isolated from 6 (19.3%) samples; and *Pseudomonas aeruginosa* was isolated from 3 (9.6%) samples.

Tables 3 and 4 show the antibiotic susceptibility of the bacterial isolates from culture-positive ascitic fluid samples. Numbers suggest percentage of total particular bacterial isolates susceptible to particular antibiotic.

### Table 1: Age and gender distribution of patients with spontaneous bacterial peritonitis

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<th>Age group (years)</th>
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<th>Gender</th>
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<tbody>
<tr>
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<tr>
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<td>70–79</td>
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</table>

### Table 2: Age and gender distribution of patients with culture-positive spontaneous bacterial peritonitis

<table>
<thead>
<tr>
<th>Age group (years)</th>
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<th>Gender</th>
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<tbody>
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<td>20–29</td>
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SBP can be a serious, fatal complication for individuals with ascites and cirrhosis, with high mortality and recurrence rates and poor long-term prognosis. Early identification of patients that are at high risk for the development of SBP has been shown to be critical for prognostic improvement. As mentioned earlier, although gram-negative bacteria are predominantly responsible for SBP, with increasing antibiotic prophylaxis, exposure to hospital environment, and frequent invasive procedures, recent studies have shown a trend toward an increase in infections of gram-positive bacteria, particularly Enterococci, Staphylococci, and Streptococci.

Early recognition of SBP by detection of eubacterial presence in otherwise sterile ascitic fluid and identification of the causative organisms involved could influence clinical decisions regarding timely initiation of therapy and appropriate antibiotic selection to ensure sufficient coverage. Furthermore, SBP may be asymptomatic or have minor symptoms only. With the early diagnosis of the disease and prompt and appropriate antibiotic treatment, the in-patient mortality of an episode of SBP has been reduced to approximately 20%, which is much lower in comparison to that found in our study. Llach et al. reported the occurrence of the first episode of SBP in cirrhotic patients with ascites; followed for a long period of time was relatively low at 11% after 1 year and 15% after 3 years of follow-up. The reason that could explain for this variation in comparison to our study is that they included patients with only moderately advanced liver disease. They included patients who were on oral nonabsorbable antibiotics during upper gastrointestinal (UGI) bleeding, which on follow-up might have reduced the risk of SBP in their patients.

Evan et al. reported the prevalence of SBP in the population of 427 cirrhotic outpatients to be 3.5%. SBP in outpatients is less frequent; occurring in patients with less advanced liver disease and may have a better outcome than its counterpart in hospitalized patients with SBP.

We found that mean age of the patients in our study was 53.5 years, which matches the findings of study conducted by Dinis et al. In our study most common presenting symptoms were jaundice (94%) followed by UGI bleeding (83%) and fever (45%). Great variations in symptoms and signs have been reported. Mihas et al. reported fever in 54%,
pain in abdomen in 57%, and hepatic encephalopathy in 67% patients. In another study by Pelletier et al.,[22] 89% patients were having fever, 42% had UGI bleeding, 53% patients had pain abdomen, and 50% cases had hepatic encephalopathy. Completely asymptomatic cases have been reported between 14% and 100%.

In our study, of 71 cases with SBP, bacteria were isolated in 31 cases (43.6%). Most of them were gram-negative bacteria, mainly *E. coli* in 17 (54.8%) and *Klebsiella pneumonia* in 5 (16.1%) cases. The only gram-positive organism was *S. aureus*, which was found in 6 (19.3%) cases. *E. coli* was found as the most common organism in most of the other studies, being found in approximately 60% of all positive culture, whereas Jain et al. found *Staphylococcus* as the most common organism.[23]

Few of the studies have reported a predominance of gram-positive organisms in ascitic fluid cultures. David et al.[24] found that 53% of the organisms were *Streptococcus*. The striking feature of our study was *P. aeruginosa* isolates from 3 (9.6%) cases, which is not a common isolate in SBP. Other studies have reported similar findings with prevalence of *P. aeruginosa* varying from 3% to 9% in culture-positive cases.[24]

We have found cultures positive only in 43.6% of our cases. This low proportion of positive ascitic fluid is probably due to the relatively low concentration of bacteria in ascitic fluid. The low rate of culture positivity can be attributed to prior antibiotics intake by the patients.

We also studied antibiotic susceptibility and resistance patterns of all the culture-positive SBP cases. *E. coli* and *Klebsiella* were susceptible to all the antibiotics except fluoroquinolones where only 70% and 80% of total isolates were susceptible. *E. coli* and *Klebsiella* were also less susceptible to tetracycline, in 80% and 70% of cases, respectively. All the *P. aeruginosa* isolates were susceptible to all antibiotics except only 50% isolates were susceptible to tetracycline, 67% were susceptible to ampicillin/sulbactum, and 60%–80% were susceptible to different fluoroquinolones [Table 3]. *S. aureus* was the only gram-positive isolate found in our study. All isolates were susceptible to all the antibiotics listed in Table 4, except only 80% isolates were susceptible to amoxicillin, 78% isolates to erythromycin, and 40% isolates were susceptible to penicillin.

All the patients were treated with IV cefotaxime 2 g BD for 5 days irrespective of their culture results. Of total 71 cases, 66 (93%) SBP cases were cured as determined by ascitic fluid culture carried out after 5 days of treatment and daily clinical evaluation and improvement in symptoms.

The treatment response to SBP with injection cefotaxime was 85% in a study by Navasa et al.[25] Our study also showed almost similar response rate. Thanopoulos et al.[26] showed that resolution of SBP was achieved in 90% patients with cefotaxime or quinolone. In another study, Franca et al.[27] found resolution rate on day 5 of treatment to be 73%. In our study, the response rate after complete treatment was 93%.[27]

There were few limitations of our study. First, the sample size was too small to have a generalized conclusion, so similar studies have to be conducted at different centers. Second,
therefore, PMN ≥250/mm³ in ascitic fluid should be considered as standard criteria to treat patients. All the organisms were susceptible to third-generation cephalosporins with favorable outcome. SBP, if diagnosed early, can be treated with high success rate; thus, ascitic fluid laboratory analysis including culture of all suspected patients will help in improving prognosis of the patients.

**References**


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