THE DISTRIBUTION AND DRUG-RESISTANCE OF PATHOGENS AMONG LIVER TRANSPLANT RECIPIENTS WITH GRAM-POSITIVE BACTEREMIAS

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ABSTRACT

Introduction: To study the species distribution and frequency of antimicrobial resistance among Gram-positive bacteria responsible for bacteremias in liver transplant recipients.

Materials and methods: Blood samples were processed by the BACTEC 9120 blood culture system. Species identification was performed using the Vitek-2 system. The drug susceptibility of pathogens was performed using the ATB FUNGUS 3 system.

Results: Sixty-eight episodes of Gram-positive bacteremias occurred in 51 liver recipients between January 31, 2003 and September 1, 2014. The median time to the onset of Gram-positive bacteremias was 6 days after liver transplantation and 92.6% of Gram-positive bacteremias were early-onset. The most common site of primary infection was intra-abdominal/biliary, followed by lungs. Of these 51 liver recipients with Gram-positive bacteremias, 25 (49%) died. The most common bacteria were Staphylococcus aureus (29.4%) and Enterococcus faecium (27.9%). Of these 20 Staphylococcus aureus, 90% were methicillin-resistant. The Gram-positive bacteria were highly resistant to erythromycin, penicillin and trimethoprim-sulfamethoxazole (resistance rate > 80%) and relatively susceptible to glycopeptides and oxazolidone antibiotics (resistance rate < 10%). There were 21 (30.9%) episodes of Gram-positive bacteremias without receiving appropriate antibiotic therapy.

Conclusion: In liver transplantation, Gram-positive bacteria led to severe bacteremias with a high mortality rate and were resistant to the majority of commonly used antibiotics.

Keywords: Liver transplantation, Gram-positive, bacteremia, drug resistance.

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Introduction

Bacteremias remain the most important infectious complications in liver transplantation. Gram-positive bacteria were sometimes the main organisms in liver transplant recipients with bloodstream infections (BSIs)⁵⁻⁹. Our previous study revealed that 46.8% of the pathogens causing BSIs were Gram-positive bacteria after liver transplantation⁹. Some researchers have found high rates of methicillin-resistant Staphylococcus aureus (MRSA), coagulase-negative Staphylococcal (CoNS) or methicillin- or vancomycin-resistant enterococcal isolates in liver recipients²⁻⁶,¹₂⁻¹⁶. The growing antibiotic-resistant Gram-positive bacteria are a significant factor that influences the prognosis and survival in liver recipients. Therefore, in this retrospective study, we aimed to investigate the species distribution and frequency of drug resistance of Gram-positive bacteria to frequently used antibiotics among liver recipients with BSIs and to provide the evidence for clinical prevention and therapy.
Materials and methods

Study population and clinical isolates

This study was conducted at the Third Xiangya Hospital, Central South University, Changsha and Zhongnan Hospital, Wuhan University, Wuhan, China, between January 1, 2003 and September 1, 2014. Immunosuppressants including calcineurin inhibitor (cyclosporine or tacrolimus) and corticosteroids with or without mycophenolate mofetil were administered to all enrolled recipients. The present study was approved by the two hospitals’ ethics committees.

Definitions

Bacteremias were defined according to Centers for Disease Control and Prevention criteria (17): The isolation of a bacterium other than normal skin flora (Diphtheroids, Bacillus spp., or Coagulase-negative Staphylococcus) in one culture with signs of infection or the isolation of a bacterium from at least two consecutive cultures correlated with signs of infection.

Microbiologic studies

A 10-mL blood sample drawn under sterile conditions was injected into each bottle of a set of aerobic and anaerobic blood culture bottles. Blood samples were processed by the BACTEC 9120 blood culture system (Becton Dickinson, Cockeysville, MD, USA). Species identification for the bacteria and fungi was performed using the Vitek-2 system (bioMérieux, Marcyl’Etoile, France). For bacteria, antimicrobial susceptibility was determined by the Kerby-Bauer disk diffusion method and the minimum inhibitory concentration (MIC) was measured by agar dilution.

Quality control was performed by Staphylococcus aureus 25923, which belonged to American Type Culture Collection (ATCC) strains. The results were evaluated according to the NCCLS manual (18). Intermediate susceptibility to the antibiotics was considered as resistance.

Antibiotics for bacteria

Amikacin (AN), levofloxacin (LVF), and trimethoprim-sulfamethoxazole (SXT), penicillin (PEN), erythromycin (ERY), clindamycin (CC), vancomycin (VAN), rifampicin (RA) and teicoplanin (TEC), and linezolid (LZD) were products of Oxoid, England. The powder of vancomycin and teicoplanin was purchased from Tongtai Co., Guangzhou, China.

Results

One hundred and fifty one episodes of BSIs occurred in 81 liver recipients between January 31, 2003 and September 1, 2014. Of them, 68 (45%, 68/151) episodes of Gram-positive bacteremias occurred in 51 patients involving 40 male and 11 female liver recipients aged 12 to 67 years (the average age 44.8 ± 13.4 years). Approximately 97% of episodes of Gram-positive bacteremias were nosocomial infection.

| Table 1: Classification and percentage of Gram-positive bacteria. |
The median time to develop Gram-positive bacteremias was 6 days (interquartile ratio: 3-15) after liver transplantation and 92.6% (63/68) of Gram-positive bacteremias were early-onset (within one month of transplantation). The most common site of primary infection was intra-abdominal/biliary (48.5%, 33/68), followed by lungs (32.4%, 22/68). There were 27 episodes of bacteremias accompanied with body temperature of 39°C or greater. A total of 25 (49%) liver recipients died due to infection, within one month posttransplantation, after being diagnosed with Gram-positive bacteremias.

The most common species was *Staphylococcus* spp., followed by *Enterococcus* spp. of *Staphylococcus* spp., *Staphylococcus aureus* (69%, 20/29) was the predominant bacterium. *Enterococcus faecium* (70.4%, 19/27) was the predominant bacterium among *Enterococcus* spp. The most common bacterium were *Staphylococcus aureus* (29.4%), followed by *Enterococcus faecium* (27.9%), *Staphylococcus epidermidis* (7.4%) and *Enterococcus faecalis* (7.4%). The species distribution of Gram-positive bacteria is illustrated in Table 1.

All but one of *Staphylococcus aureus* and *Enterococcus faecium* were susceptible to vancomycin. However, of those 20 *Staphylococcus aureus*, 18 (90%) were MRSA. Table 2 showed the drug resistance of Gram-positive bacteria to 10 kinds of commonly used antibiotics.

### Table 2: Gram-positive cocci on antibiotic resistance rate of ten kinds of commonly used antibiotics.

<table>
<thead>
<tr>
<th>Antibacterial agents</th>
<th>S.aureus -10</th>
<th>Enterococcus -10</th>
<th>The other staphylococcus -10</th>
<th>The other enterococcus -10</th>
<th>The other positive bacteria -10</th>
<th>Total drug tolerance rate (%)</th>
</tr>
</thead>
<tbody>
<tr>
<td>ERY</td>
<td>18(90.0)</td>
<td>8(44.4)</td>
<td>16(84.2)</td>
<td>7(85.7)</td>
<td>11(91.7)</td>
<td>88.2</td>
</tr>
<tr>
<td>VAN</td>
<td>1(5.0)</td>
<td>0(0.0)</td>
<td>1(5.3)</td>
<td>0(0.0)</td>
<td>4(33.3)</td>
<td>8.8</td>
</tr>
<tr>
<td>CC</td>
<td>14(70.0)</td>
<td>5(60.0)</td>
<td>16(84.2)</td>
<td>8(100.0)</td>
<td>9(75.0)</td>
<td>76.5</td>
</tr>
<tr>
<td>LVF</td>
<td>10(50.0)</td>
<td>6(55.6)</td>
<td>14(73.7)</td>
<td>8(100.0)</td>
<td>10(83.3)</td>
<td>72.1</td>
</tr>
<tr>
<td>SXT</td>
<td>15(75.0)</td>
<td>6(55.6)</td>
<td>17(87.5)</td>
<td>8(100.0)</td>
<td>9(75.0)</td>
<td>80.9</td>
</tr>
<tr>
<td>AN</td>
<td>6(30.0)</td>
<td>3(33.3)</td>
<td>14(73.7)</td>
<td>4(50.0)</td>
<td>8(66.7)</td>
<td>51.5</td>
</tr>
<tr>
<td>PEN</td>
<td>19(95.0)</td>
<td>9(100.0)</td>
<td>11(57.9)</td>
<td>7(100.0)</td>
<td>10(83.8)</td>
<td>82.4</td>
</tr>
<tr>
<td>RA</td>
<td>2(10.0)</td>
<td>2(22.2)</td>
<td>15(78.9)</td>
<td>5(62.5)</td>
<td>8(66.7)</td>
<td>47.1</td>
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<tr>
<td>TEC</td>
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<td>0(0.0)</td>
<td>0(0.0)</td>
<td>0(0.0)</td>
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<tr>
<td>LZD</td>
<td>0(0.0)</td>
<td>0(0.0)</td>
<td>0(0.0)</td>
<td>0(0.0)</td>
<td>0(0.0)</td>
<td>0.0</td>
</tr>
</tbody>
</table>

### Discussion

BSIs are still an important complication threatening liver recipients so far. In the present study, the main pathogens causing BSIs were Gram-positive bacteria after liver transplantation in the both hospitals investigated over eleven years and 92.6% of Gram-positive bacteremias were early-onset, in accordance with the study reporting that the majority of the Gram-positive bacteremias occurred within 30 days among liver recipients. In the present study, the drug susceptibility test showed that the susceptibility of Gram-positive bacteria to the ten kinds of commonly used antibiotics was low. These Gram-positive bacteria were only susceptible to glycopeptide and oxazolidinone antibiotics, which was consistent with a previous report. The present study found no episodes of teicoplanin-resistant *Staphylococcus aureus* and *Enterococcus* spp., and all but one episode of *Staphylococcus aureus* were susceptible to vancomycin. However, 90% (18/20) of *Staphylococcus aureus* isolates were confirmed to be MRSA, in line with other studies which stressed that, among bacteremic liver recipients, more than 90% of *Staphylococcus aureus* were MRSA. Therefore, MRSA represents a serious problem among liver recipients with bacteremias. Only 5% (1/19) of *Enterococcus faecium* isolates was resistant to vancomycin, in contrast to the study demonstrating that 38.0% of *Enterococcus faecium* were vancomycin-resistant. The predominance of Gram-positive cocci in bacteremias has been associated with CoNS (93.3% of *Staphylococcus* spp., and 71.4% of the Gram-positive bacteria) in a study among liver recipients with BSIs. However, our present study found CoNS was accounted for only 31% of *Staphylococcus* spp and 13.2% of the Gram-positive bacteria. The results from the current study, therefore, demonstrated that these CoNS pathogens represented less of a problem than *Staphylococcus aureus*. The Gram-positive bacteria were highly resistant to erythromycin, penicillin and trimethoprim-sulfamethoxazole (resistance rate > 80%) and were relatively susceptible to glycopeptides and oxazolidinone antibiotics (resistance rate < 10%). There were 21 (30.9%) episodes of Gram-positive bacteremia without receiving appropriate antibiotic therapy. None of *Staphylococcus aureus* and *Enterococcus* spp., was resistant to teicoplanin and none of the Gram-positive bacteria was resistant to linezolid.
The reasons for the distinction between the two studies included a different occurrence rate of nosocomial catheter-related BSIs and the use of different antimicrobial prophylaxis after liver transplantation.

The present study also revealed that Gram-positive bacteria causing bacteremias after liver transplantation had high drug resistance. The possible explanations to this finding include the nosocomial infection and the long-term use of broad-spectrum antibiotics and immunosuppressants after liver transplantation. Physicians can choose glycopeptide and oxazolidinone antibiotics for empirical therapy in highly suspected patients with Gram-positive bacteremias when they are waiting for the bacterial identity and susceptibility pattern.

References

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