Original article

Clinical study of distribution and drug resistance of pathogens in patients with severe acute pancreatitis

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Keywords: pathogen; drug resistance; severe acute pancreatitis

Background Previous researches about necrotic pancreatic tissue infections are numerous, but the study on systemic infection related to the severe acute pancreatitis (SAP) treatment period is limited. This study aimed to investigate the distribution and drug resistance of pathogenic bacteria in patients who had hepatobiliary surgery for SAP during the past three years.

Methods A retrospective study was conducted on the distribution, category and drug resistance of pathogenic bacteria in patients who had hepatobiliary surgery for SAP from 2008 to 2011.

Results A total of 594 pathogenic bacteria samples were isolated. Among them 418 isolates (70.4%) were Gram bacteria negative, 142 isolates (23.9%) were Gram bacteria positive, and 34 isolates (5.7%) were found fungi. The most common Gram negative bacteria were Escherichia coli (19.8%), and the dominant Gram positive pathogenic bacteria were Enterococcus faecium. The distribution of SAP-related infectious pathogens was mainly in peritoneal drainage fluid, sputum, bile, and wound secretions. Almost all the Gram negative pathogenic bacteria were sensitive to carbapenem. Extended-spectrum β-lactamases (ESBLs) producing strains were more resistant to penicillins and cephalosprins than the ESBLs non-producing strains. Staphylococcus was sensitive to vancomycin and linezolid. The drug resistance of meticillin-resistant staphylococcus (MRS) to commonly used antibiotics was higher than meticillin-sensitive streptococcus (MSS). Enterococcus sp. exhibited lower drug-resistance rates to vancomycin and linezolid.

Conclusions Gram negative bacteria were the dominant SAP-related infection after hepatobiliary surgery. A high number of fungal infections were reported. Drug resistant rates were high. Rational use of antibiotics according to the site of infection, bacterial species and drug sensitivity, correctly executing the course of treatment and enhancing hand washing will contribute to therapy and prevention of SAP-related infection and decrease its mortality.

Severe acute pancreatitis (SAP) is a major critical disease of hepatobiliary surgery in the intensive care unit (ICU). Inflammatory cytokines produced by necrotic pancreatic tissue are released into the bloodstream and injure organs. Its mortality rate still remains as high as to 30% despite the current SAP therapies that have made great progress. SAP-related infections are a major factor that contribute to mortality (50%-80% mortality). In addition to necrotic pancreatic tissue infections, the categories of infections also include other intra-abdominal infections, lung infections, venous catheter infections, wound infections, urinary tract infections, and bloodstream infections. There are extensive previous researches about the necrotic pancreatic tissue infections, but the researches of systemic infection related to the SAP treatment period are still limited. On the other hand, antimicrobial agents for the treatment of SAP associated infections have an irreplaceable position along with surgical drainage. Therefore we conducted a survey of microbiological examination of 594 multiple infection specimens in severe pancreatitis cases admitted to People’s Liberation Army General Hospital during August 2008–July 2011. We studied the distribution of SAP related pathogen infection and analyzed the drug resistance in order to find the characteristics of bacterial distribution and antibiotic sensitivity. This information will have important significance for the treatment of SAP-related infections and decrease SAP-related infection mortality.

METHODS

Subjects

Ten specimens from SAP patients, including sputum,
bronchial aspirates, pus and secretions, urine, blood, etc., were collected by aseptic techniques from hospitalized patients who underwent hepatobiliary surgery from August 2008 to July 2011. Pathogenic species were cultured and isolated from selected specimens.

**Bacterial identification and susceptibility test**

Blood agar was used for culture of pathogenic bacteria. Drug susceptibility of Gram negative bacteria was detected using the Kirby-Bauer (KB) method with 13 antibiotics including amoxicillin/clavulanic acid, amikacin, ampicillin/sulbactam, cefuroxime, cefotaxime, imipenem, ceftazidime, cefepime, meropenem, ciprofloxacin, cefoperazone/sulbactam, piperacillin/tazobactam, and cotrimoxazole. Drug susceptibility of Gram-positive bacteria was detected with 12 antibiotics including gentamicin, penicillin, ampicillin/sulbactam, cefazolin, levofloxacin, vancomycin, erythromycin, clindamycin, cotrimoxazole, rifampicin, and linezolid. The standard strain of *E. coli* ATCC25922 was used for quality control. Results were determined according to guidelines published by the U.S. National Committee for Clinical Laboratory (NCCLS). Antibiotic discs and blood agar plate were purchased from Oxoid Ltd (Basingstok, UK).

**RESULTS**

Pathogen distribution

Five hundred and ninety-four bacterial samples were isolated, that included 418 (70.4%) Gram negative bacterial isolates, 142 Gram-positive bacterial isolates (23.9%), and 34 fungi isolates (5.7%). Among the isolated Gram negative pathogenic bacteria, *E. coli* was the most common (19.8%), followed by *Pseudomonas aeruginosa* (13.0%), and *Acinetobacter baumannii* (11.8%). Among the isolated Gram positive pathogenic bacteria, *Enterococcus faecium* was the most common (10.1%), followed by coagulase-negative staphylococci (5.4%), and *Enterococcus faecalis* (2.9%). *Candida tropicalis* and *Candida albicans* were the main fungi strains (Table 1). The main sources of pathogens were peritoneal drainage fluid, sputum, bile and wound secretions (Table 2).

**Drug susceptibility results**

This study showed that the rates of antimicrobial drug resistance were significantly higher among the main Gram negative bacteria (including *E. coli* and *K. pneumoniae* strains) in the extended-spectrum β-lactamases (ESBLs) producing strains than in the ESBLs non-producing strains that came from the hepatobiliary surgery wards. But the incidence of drug resistance to carbapenem and to drugs containing enzyme inhibitors was low, thus these kinds of antibiotics were the better choice.

The drug resistant rates of *Acinetobacter baumannii* to...
common antibiotics were 81.7%–99.7%. The incidence of drug resistance to amikacin, meropenem, and ceftazidime was low in Pseudomonas aeruginosa (Table 3). Among Gram positive bacteria, the drug resistance of methicillin-resistant S. aureus (MRSA) to commonly used antibiotics was higher than in methicillin-sensitive S. aureus (MSSA). Vancomycin and linezolid resistant Staphylococcus spp. were not detected. Resistance rates to linezolid and vancomycin in enterococci were low (Table 4).

**DISCUSSION**

SAP is a serious type of acute pancreatitis, in which the mortality rate may be up to 30%.1 Features of SAP include local complications (pancreatic necrosis and pseudocyst) and systemic complications (organ dysfunction, shock, and acute respiratory distress syndrome (ARDS)), inflammatory mediators, and cytokine production by necrotic pancreatic tissue that is released into the bloodstream and affects various organs. Infection related complications are the main critical circumstances of SAP. The infection of necrotic pancreas tissue often occurs in the second phase of SAP. This phase is usually recorded in the second week (24% patients) and the fourth week of the disease (71% patients).2 Septic complications of pancreatitis develop in 40%–70% of patients with 50%–80% mortality.2,3 The mortality correlates with the extent of pancreatic necrosis. According to current studies, infection in SAP is considered to cause a fatal outcome unless treated with surgical necrosectomy and lavage,4–6 or with percutaneous catheter drainage (PCD).7,8 It has become the most important risk factor of death from necrotizing pancreatitis.9

SAP-related infections not only include infections of necrotic pancreatic tissue, but also include intra-abdominal infections, lung infections, catheter related infections, wound infections, urinary tract infections, and bloodstream infections. Infection associated with the necrotic pancreas is described in many reports.10–12 However, in the treatment of SAP, the associated multi-system infections also play an important role in the prognosis. In our literature review we found few studies about such conditions. Antimicrobial agents have an irreplaceable position in the treatment of SAP associated infections, along with surgical drainage. Even leading surgical centers have reported that mortality rates associated with early surgery are 20%–50% or even higher.13,14 Our results report the microbiological species isolated from SAP-accompanied infections. In this research, we found the main infection sites were abdominal drainage fluid, sputum, bile juice, and wound secretions. This result was related to the character of hepatobiliary disease such as complicated disease range, longer time operation and NPO (Nihil per os), lower immune system, more invasive catheter and broad-spectrum antibiotics, etc. The pancreas and the area around it were the main infection sites. Infection can occur via haematogenous spread, transmural migration through the bowel wall, via ascites to the pancreas, via the lymphatics to the circulation, or via the duodenum to the main pancreatic duct.15,16

Five hundred and ninety-five bacterial isolates were found, of which 418 were Gram negative, 142 were Gram positive, and there were 34 fungi. Among the Gram negative isolates, E. coli was the most frequently identified. P. aeruginosa and A. baumannii were the second most frequently identified strains. In the Gram positive bacteria group, the dominant pathogens were E. faecium, coagulase negative staphylococci and E. faecalis. The main fungi were C. tropicalis and C. albicans.

In this study, the results of drug sensitivity testing showed that the antimicrobial activities of carbapenems, the third generation cephalosporins plus enzyme inhibitors, and penicillin antibiotics were better against E. coli and K. pneumoniae. The drug resistance rates of Gram positive bacteria to erythromycin and penicillin were higher than other drugs. No vancomycin resistant bacterial strain was found. The drug resistance rate of P. aeruginosa and A. baumannii, which were opportunistic pathogens, showed a gradually increasing trend compared with previous studies. Because of the existence of various drug resistance mechanisms, antibiotic selection has become

### Table 4. The resistance rates of main Gram-positive bacilli to antimicrobial drugs (%)

<table>
<thead>
<tr>
<th>Drugs</th>
<th>Enterococcus faecium (n=60)</th>
<th>Coagulase-negative staphylococi (n=32)</th>
<th>Enterococcus faecalis (n=17)</th>
<th>Staphylococcus aureus (n=11)</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>MSSCoN (n=11)</td>
<td>MSSCoN (n=21)</td>
<td>MSSA (n=5)</td>
<td>MRSA (n=6)</td>
</tr>
<tr>
<td>Gentamicin</td>
<td>77.5</td>
<td>9.2</td>
<td>45.2</td>
<td>49.3</td>
</tr>
<tr>
<td>Penicillin</td>
<td>92.7</td>
<td>65.2</td>
<td>100.0</td>
<td>12.1</td>
</tr>
<tr>
<td>Cefotaxim</td>
<td>0</td>
<td>0</td>
<td>100.0</td>
<td>0</td>
</tr>
<tr>
<td>Ampicillin/sulbactam</td>
<td>0</td>
<td>0.5</td>
<td>100.0</td>
<td>0</td>
</tr>
<tr>
<td>Cefazolin</td>
<td>0</td>
<td>0</td>
<td>100.0</td>
<td>0</td>
</tr>
<tr>
<td>Levofloxacin</td>
<td>0.8</td>
<td>11.7</td>
<td>58.3</td>
<td>4.5</td>
</tr>
<tr>
<td>Vancomycin</td>
<td>6.8</td>
<td>0</td>
<td>0</td>
<td>4.5</td>
</tr>
<tr>
<td>Erythromycin</td>
<td>0</td>
<td>76.2</td>
<td>91.4</td>
<td>0</td>
</tr>
<tr>
<td>Clindamycin</td>
<td>0</td>
<td>35.6</td>
<td>63.8</td>
<td>0</td>
</tr>
<tr>
<td>Cotrimoxazole</td>
<td>0</td>
<td>28.4</td>
<td>65.1</td>
<td>0</td>
</tr>
<tr>
<td>Rifampicin</td>
<td>0</td>
<td>3.3</td>
<td>76</td>
<td>0</td>
</tr>
<tr>
<td>Linezolid</td>
<td>2.9</td>
<td>0</td>
<td>0</td>
<td>6.4</td>
</tr>
</tbody>
</table>

MSSCoN: methicillin-sensitive staphylococcus coagulase-negative; MRSCoN: methicillin-resistant staphylococcus coagulase-negative; MSSA: methicillin-sensitive Staphylococcus aureus; MRSA: methicillin-resistant Staphylococcus aureus.
more difficult. It also accounts for the occurrence of uncontrollable multi-site infections in patients after an operation and increases mortality. In the study by Tsui et al., cultured organisms were usually multidrug-resistant, including Gram negative bacteria, Gram positive bacteria, and fungi. These organisms were mainly included, for example, *P. aeruginosa*, *Xanthomonas maltophilia*, enterococcus, methicillin-resistant *S. aureus*, *C. albicans* and *C. krusei*. A high isolation rate of *A. baumannii* was found in another study, which was considered to correlate with the high rate of intensive care unit (ICU)-acquired infection in patients with severe sepsis. Fungi, which are considered to be caused by extensive use of extended-spectrum antibiotics, are frequently isolated in cases of severe sepsis. The incidence of invasive fungal infection is reported to be 28.3% in the severe sepsis patients. But Tsui et al. reported a relatively low incidence of fungal infections. They considered it to still be unclear whether the use of long-term antibiotic treatment promoted fungal infections.

All of the Gram negative pathogenic bacterial strains were sensitive to carbapenem in this investigation. So for experimental treatment of infection, carbapenems, the third generation cephalosporins plus enzyme inhibitors, and penicillin should be chosen first. Peng et al. reported that almost all of the Gram negative pathogenic bacteria were sensitive to carbapenem. Aminoglycosides should be used cautiously because of their ototoxicity and nephrotoxicity in spite of their good antibacterial activity. ESBLs producing strains were more resistant to penicillins and cephalosporins than ESBLs non producing strains. Staphylococcus was sensitive to vancomycin and linezolid. The drug resistance of meticillin-resistant staphylococcus (MRS) was higher to commonly used antibiotics than meticillin-sensitive streptococcus (MSS). Vancomycin and linezolid showed lower drug-resistance rates against *Enterococcus sp*. In order to avoid or slow the development of the drug resistant bacterial strains, combinations of two kinds of antibiotics were necessary for complicated and critical patients. The study also indicated that surgeons should increase the sample test rate of surgical infection patients so as to improve the specificity of the prescription. Before the pathogen cultured results are available, antibiotics against both Gram negative and Gram positive bacteria could be selected for treatment. After getting results, we should quickly adjust antibiotics according to bacteria culture results and the disease progression. This could reduce the occurrence and dissemination of drug resistant bacterial strains.

On the other hand, the occurrence of SAP associated infections is the result of complex interactions probably involving patients’ underlying conditions, the characteristics of the patient population, the extensive use of extended-spectrum antibiotics, low attention to sanitary precautions, the invasive interventions, and the quality of care provided. Thus more attention should be paid to sanitary precautions for decreasing the occurrence of drug resistant bacterial strains, such as hand washing in the ICU.

In conclusion, for pathogen distribution of SAP-related infection in hepatobiliary surgery, Gram negative bacteria were dominant, and an increased number of fungal infections were reported. Drug resistant rates were high. Rational use of antibiotics according to the site of infection, bacterial species and drug sensitivity, correctly monitoring the course of treatment and enhancing hand washing will contribute to therapy and prevention of SAP-related infections and decrease their mortality.

REFERENCES


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