INTRODUCTION

Patients in intensive care units (ICUs) have a higher risk of acquiring hospital acquired infections (HAIs) than those in non-critical care areas.1 ICU-acquired infection rate is five to ten times higher than hospital-acquired infection rates in general ward patients.2 Considerable increased mortality and major costs are associated with ICU-acquired infections.3-5 This results from the acute severity of illness of ICU patients, prolonged hospital stay, immunosuppression, increased use of antimicrobials and frequent exposure to therapeutic procedures like endotracheal intubation with mechanical ventilation, nasotracheal intubation, intravenous lines, central venous lines and urinary catheterization.6-9

The most common HAIs in ICU patients are respiratory tract infections (RTIs), urinary tract infections (UTIs) and bloodstream infections (BSIs) and are most often associated with the use of invasive devices.7 Both Gram-positive and Gram-negative bacteria and Candida spp. have been reported as cause of infection in these patients.10-12 Recently, Gram-negative bacilli have been reported more frequently than Gram-positives in this setting.10

The ICUs are an area of considerable antibiotic use in which antibiotic-resistant organisms are prevalent.13,14 Their prevalence and rates of resistance can vary enormously depending on geographic location as well as location among ICU types.15 For proper management of ICU-infections, it is important to have updated knowledge about prevalence of the causative agents and their antimicrobial susceptibility patterns in institution-specific ICUs.16 There are very few published studies on the epidemiology of HAIs in ICU patients from Pakistan. Therefore, this study was carried out to determine the prevalent microorganisms in medical ICU-patients and antimicrobial susceptibility profile of the isolates to the commonly used older and newer antibiotics.

METHODOLOGY

The study was carried out at the eight-bed medical ICU (MICU) of The Holy Family Hospital in Rawalpindi, from May 2007 to April 2008. The patients were observed for any signs of site-specific infections or fever appearing anytime after 48 hours of admission into ICU. Patients admitted into ICU with any signs of infections were
Hospital acquired infections in a medical intensive care unit

excluded from the study. Clinical specimens including tracheal aspirates, sputum, urine, urine catheter tips, central venous line tips, blood, body fluids, pus and others were collected from the patients and were processed at microbiology laboratory of the hospital. The specimens were cultured onto suitable culture media like MacConkey agar, sheep blood agar, chocolate agar and Sabouraud's agar. Plates were incubated aerobically for 24-48 hours. The isolates were identified by colonial morphology, gram-staining, biochemical tests like catalase, coagulase, oxidase, bile solubility and API-10S (Biomeroux, France).

Antimicrobial susceptibility testing was performed on Mueller Hinton agar using disc diffusion method in accordance with Clinical and Laboratory Standard Institute Guidelines."\(^\text{17}\) Zone sizes of each antimicrobial agent were recorded and interpreted as resistant, intermediate or susceptible. Intermediately susceptible isolates were considered resistant. Susceptibility was tested against following antimicrobials according to their spectrum of activity against Gram-positive or Gram-negative bacteria. Antimicrobials tested against Gram-positive bacteria were: ampicillin, co-amoxiclav, cephradine, cephalexin, cefaclor, cefotaxime, amikacin, gentamicin, tetracycline, chloramphenicol, co-trimoxazole, ofloxacin, ciprofloxacin, vancocin, and imipenem. Methicillin resistance in MRSA was tested using oxacillin discs. Antimicrobials tested against Gram-negative bacteria were co-amoxiclav, cefotaxime, cefazidime, ceftriaxone, amikacin, gentamicin, tetracycline, chloramphenicol, co-trimoxazole, ofloxacin, ciprofloxacin, vancocin, and imipenem.

Methicillin resistance in MRSA was tested using oxacillin discs. Antimicrobials tested against Gram-negative bacteria were co-amoxiclav, cefotaxime, cefazidime, ceftriaxone, amikacin, gentamicin, tetracycline, chloramphenicol, co-trimoxazole, ofloxacin, ciprofloxacin, vancocin, and imipenem. Amikacin resistance in MRSA was tested using oxacillin discs. Antimicrobials tested against Gram-negative bacteria were co-amoxiclav, cefotaxime, cefazidime, ceftriaxone, amikacin, gentamicin, tetracycline, chloramphenicol, co-trimoxazole, ofloxacin, ciprofloxacin, vancocin, and imipenem.

Microsoft Excel spread sheet was used to analyze the data in the form of percentages. Approval for the study was obtained from Research and Ethics Committee, Rawalpindi Medical College and Allied Hospitals, Rawalpindi.

**RESULTS**

Out of total 440 specimens received, bacteria or Candida spp. were isolated from 269/440 (60.1%) samples. The most frequent site of infection was the respiratory tract (47.95%) followed by urinary tract (25.3%), wounds (8.2%) and blood stream (7.06% BSI). *K. pneumoniae* (30.2%), *P. aeruginosa* (28.7%), and *E. coli* (19.4%) were the commonest organisms isolated from respiratory tract samples. Similarly, *K. pneumoniae* (25%), *E. coli* (23.5%), *P. aeruginosa* (16.2%) and Candida spp. (14.7%) were isolated from urinary tract samples. Organisms causing BSIs were *P. aeruginosa* (31.6%), *E. coli* (31.6%), *Streptococcus pneumoniae* (21.1%), and *K. pneumoniae* (10.5%). These BSI organisms were isolated from cultures of blood and the central venous line tips.

The isolation rate of other Gram-negative bacteria, *Staphylococcus (S.) aureus* and methicillin-resistant *S. aureus* (MRSA), was relatively low.

Details of the antibiotic resistance pattern in Gram-positive and Gram-negative isolates are shown in Table I and Table II respectively. None of the isolates, except vancomycin against Gram-positive isolates, were 100% sensitive to the antibiotics tested. Relatively low resistance was only observed against amikacin (21.3%) and imipenem (26.1%). Resistance in Gram-negative isolates was more marked than in Gram-positive isolates. Resistance to commonly used antibiotics was very high. Majority of the Gram-negative and to a lesser extent, Gram-positive isolates showed > 50% resistance to many of the antibiotics tested. Out of two fluoroquinolones tested, higher resistance was observed in ciprofloxacin (73.9% and 69.2%) than ofloxacin (65.9% and 61.5%) in Gram-negative and Gram-positive isolates respectively. Among third generation cephalosporins tested, >60% of the majority of Gram-negative isolates were resistant. Among the aminoglycosides tested, resistance was higher to gentamicin than amikacin.

**Table I**: Antibiotic resistance pattern of Gram-positive bacterial clinical isolates from patients admitted in medical intensive care unit (MICU).

<table>
<thead>
<tr>
<th>Antibiotics</th>
<th><em>S. aureus</em> (n=13)</th>
<th>MRSA (n=19)</th>
<th><em>S. pneumoniae</em> (n=07)</th>
<th>Total (n=39)</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>No resistant (%)</td>
<td>No resistant (%)</td>
<td>No resistant (%)</td>
<td>No resistant (%)</td>
</tr>
<tr>
<td>Amoxicillin</td>
<td>12 (92.3)</td>
<td>0 (0)</td>
<td>0 (0)</td>
<td>16 (40)*</td>
</tr>
<tr>
<td>Co – amoxyclav</td>
<td>09 (69.2)</td>
<td>11 (57.9)</td>
<td>03 (42.9)</td>
<td>23 (59)</td>
</tr>
<tr>
<td>Cephradine</td>
<td>03 (23.1)</td>
<td>02 (10.5)</td>
<td>02 (28.6)</td>
<td>5 (25)*</td>
</tr>
<tr>
<td>Cephalexin</td>
<td>06 (46.2)</td>
<td>04 (21.1)</td>
<td>04 (57.1)</td>
<td>10 (50)*</td>
</tr>
<tr>
<td>Cefaclor</td>
<td>08 (61.5)</td>
<td>04 (57.1)</td>
<td>04 (57.1)</td>
<td>12 (60)*</td>
</tr>
<tr>
<td>Cefotaxime</td>
<td>06 (46.2)</td>
<td>03 (23.1)</td>
<td>03 (42.9)</td>
<td>9 (45)*</td>
</tr>
<tr>
<td>Ofloxacin</td>
<td>05 (38.5)</td>
<td>04 (21.1)</td>
<td>02 (28.6)</td>
<td>24 (61.5)</td>
</tr>
<tr>
<td>Ciprofloxacin</td>
<td>08 (61.5)</td>
<td>02 (10.5)</td>
<td>02 (28.6)</td>
<td>27 (69.2)</td>
</tr>
<tr>
<td>Amikacin</td>
<td>01 (7.7)</td>
<td>01 (5.3)</td>
<td>01 (14.3)</td>
<td>03 (7.7)</td>
</tr>
<tr>
<td>Gentamicin</td>
<td>10 (76.9)</td>
<td>01 (5.3)</td>
<td>04 (57.1)</td>
<td>30 (76.9)</td>
</tr>
<tr>
<td>Tetracycline</td>
<td>08 (61.5)</td>
<td>15 (78.9)</td>
<td>05 (71.4)</td>
<td>28 (71.8)</td>
</tr>
<tr>
<td>Chloramphenicol</td>
<td>02 (15.4)</td>
<td>10 (52.6)</td>
<td>05 (71.4)</td>
<td>17 (43.6)</td>
</tr>
<tr>
<td>Co – trimoxal</td>
<td>09 (69.2)</td>
<td>18 (94.7)</td>
<td>02 (28.6)</td>
<td>29 (74.4)</td>
</tr>
<tr>
<td>Imipenem</td>
<td>07 (53.8)</td>
<td>06 (31.6)</td>
<td>02 (28.6)</td>
<td>15 (38.5)</td>
</tr>
<tr>
<td>Vancomycin</td>
<td>0 (0)</td>
<td>0 (0)</td>
<td>0 (0)</td>
<td>0 (0)</td>
</tr>
</tbody>
</table>

*No (%) of isolates resistant to the tested antibiotic out of total of 20 (S. aureus=13 and S. pneumoniae=07).*

**DISCUSSION**

HAIs are seen worldwide but are less studied and are given less emphasis in developing countries like Pakistan. This study reports a high rate of HAIs, as well as high antibiotic resistance, in ICU settings of a tertiary care hospital in Rawalpindi. RTIs (47.95%) and UTIs (25.3%) were the commonest infections from these...
patients and this is in agreement with other studies from Pakistan\textsuperscript{11,12,18} and other countries.\textsuperscript{3,9} In a study conducted at Karachi, the frequency of RTIs and UTI was 21% and 44.6% respectively.\textsuperscript{12} In another study carried out at Hyderabad, the frequency of RTIs and UTIs was 30.1% and 39.1% respectively. This difference could possibly be due to the difference in antibiotic prescribing practices and variations in sample collection, culture and susceptibility testing practices. Most RTIs were associated with mechanical ventilation (82.2%) as the later is a major factor for RTIs in ICUs.\textsuperscript{1,19} Higher rate of UTIs (25.3%) in these patients is obvious due to chronic illnesses like stroke and diabetes. This was associated with urinary catheterization (67.6%) and prolonged urinary catheterization is a major risk factor for UTIs in ICU patients.\textsuperscript{15} These findings are in close agreement with those reported by the U.S. National Nosocomial Infections Surveillance (NNIS) system and local studies.\textsuperscript{7,12,18} Relatively lower rate of BSIs (7.06%) in this study as compared to other local studies 23.7\% and 27%,\textsuperscript{12,18} may partly be due to the reason, that blood cultures were manually processed or primary BSIs are less prevalent in our setting. The relatively high incidence of HAIs observed in this study may be a reflection of the higher severity of illness, poor nutritional status, more interventions, fewer staff, poor adherence to aseptic measures and high proportion of mechanical ventilation.\textsuperscript{4,9}

Gram-negative bacteria i.e. \textit{P. aeruginosa}, \textit{K. pneumoniae} and \textit{E. coli} were the commonest organisms isolated from RTIs, UTIs and BSIs. This is in agreement with previous studies from Pakistan\textsuperscript{12,20} and other countries.\textsuperscript{9,10} Prevalence of other Gram-negative bacteria, \textit{S. aureus} and MRSA is relatively low as in general Gram-negative bacterial infections are increasing in ICUs than Gram-positive bacterial infections.\textsuperscript{10,21} A markedly higher isolation rate of \textit{Candida} spp. from UTIs is in agreement with other report from US NNIS,\textsuperscript{22} but in present study this was not associated with urinary catheterization.

In this study the most commonly used antibiotics belonging to penicillins, cephalosporins, fluoro-quinolones, aminoglycosides, co-trimoxazole and carbapenems were tested against the bacterial isolates to have an update current status of the resistance pattern. The higher resistance in these patients is probably due to different antibiotic prescribing practices as it varies enormously.\textsuperscript{23} Very high resistance rate to the most commonly used antibiotics including; fluoroquinolones is most likely due to misuse, overuse and over-the-counter availability of these antibiotics. HAIs particularly in ICUs admitted patients who are at higher risk get primary infection with multi-resistant organisms. This makes the treatment of these infections progressively more complicated\textsuperscript{16} and is also associated with increased patient charges or hospital costs and mortality.\textsuperscript{5} Furthermore, a major contributing factor to this increased resistance is use of the antibiotics without culture and susceptibility testing as even basic facilities for culture and antibiotic susceptibility testing are not available at majority of healthcare settings in our set up. Relatively lower resistance rate to imipenem and amikacin is probably due to lesser use of these antibiotics and is in agreement with another local study.\textsuperscript{12} Vancomycin remains the only option for MRSA as most of the other antibiotics tested appear ineffective; however, chances of development of vancomycin-resistant \textit{S. aureus} are there.

In view of this present situation one approach is to opt for newer and more expensive antibiotics like linezolid, dalfopristin-quinupristin and tigecycline for Staphylococcal infections and cefoperazone-sulbactam for Gram-negative infections. Better and a more cost effective approach will be, use of the antibiotics after culture and antibiotic susceptibility testing particularly in ICU.

\begin{table}
\centering
\begin{tabular}{|c|c|c|c|c|c|c|}
\hline
\textbf{Antibiotics} & \textbf{E. coli (n=53)} & \textbf{K. pneumonia (n=72)} & \textbf{Citrobacter spp. (n=05)} & \textbf{Proteus spp. (n=09)} & \textbf{P. aeruginosa (n=74)} & \textbf{Total (n =211)} \\
\hline
Co-amoxiclav & 45 (84.9) & 60 (83.3) & 03 (100) & 08 (88.9) & 53 (71.6) & 169 (80.1) \\
Cefotaxime & 33 (62.3) & 43 (59.7) & 02 (66.7) & 05 (55.6) & 49 (66.2) & 132 (62.6) \\
Ceftriaxone & 38 (71.7) & 34 (47.2) & 02 (66.7) & 06 (66.7) & 63 (85.1) & 143 (67.8) \\
Amikacin & 13 (24.5) & 21 (29.2) & 02 (66.7) & 02 (22.2) & 07 (9.5) & 45 (21.3) \\
Oxacillin & 73 (100) & 73 (100) & 01 (100) & 08 (88.9) & 53 (71.6) & 146 (69.2) \\
Tetracycline & 38 (71.7) & 44 (61.1) & 02 (66.7) & 09 (100) & 53 (71.6) & 146 (69.2) \\
Chloramphenicol & 34 (64.2) & 42 (58.3) & 02 (66.7) & 05 (55.6) & 44 (59.5) & 125 (59.2) \\
Co -trimoxazole & 34 (64.2) & 64 (88.9) & 02 (66.7) & 08 (88.9) & 51 (68.9) & 159 (75.4) \\
Imipenem & 08 (15.1) & 15 (20.8) & 01 (33.3) & 01 (11.1) & 30 (40.5) & 55 (26.1) \\
Aztreonam & 17 (32.1) & 05 (6.9) & 02 (66.7) & 05 (55.6) & 45 (60.8) & 74 (35.1) \\
\hline
\end{tabular}
\caption{Antibiotic resistance pattern of Gram-negative bacterial clinical isolates from patients admitted in medical intensive care unit (MICU).}
\end{table}
patients. The best approach would be to improve and implement optimal infection prevention practices in ICUs of low resource countries and expertise as it is believed that one third of these hospital-acquired infections can be prevented by optimal infection prevention practices.25

CONCLUSION

Bacteria or Candida spp. were isolated from 60.1% samples taken from supposedly infected patients admitted in medical ICU. The most frequent site of infection was the respiratory tract followed by urinary tract. P. aeruginosa, K. pneumoniae and E. coli were the commonest organisms followed by MRSA and S. aureus. The high frequency of HAIs and high bacterial resistance rate in ICU patients indicate the importance to study HAIs and suggest, that more strict measures regarding infection control practices along with prescription of antibiotics according to antibiotic susceptibility testing should be implemented to limit the emergence of antibiotic resistant organisms.

Acknowledgement: This study is a part of research project funded by Higher Education Commission of Pakistan, Islamabad. Thanks to my colleagues Mr. Yusuf Rajput, Dr. Umar Mahmood, Dr. Amber Habib, Dr. Abubakr, Dr. Waqas, Mr. Tariq Baig, Mr. Arthur Manzoor, Mr. Muhammad Mushtaq and Mr. Sajjad for their facilitation in this study in their capacity.

REFERENCES


.....★.....