Antibiotic Susceptibility of Bacterial Strains Isolated from Urinary Tract Infections in a Private Laboratory of Dhaka City

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Abstract

Background: Urinary tract infection (UTI) is one of the most common infections worldwide. Objective: The aim of the study was to identify different species of microorganisms, along with their antimicrobial susceptibility pattern, causing urinary tract infection in outpatient and indoor patients at Farazy Hospital Ltd, Dhaka, Bangladesh. Methodology: This retrospective study was conducted using urine culture and sensitivity reports collected retrospectively from records maintained in the Department of Microbiology over a one-year period from January 2020 to December 2020 in a tertiary care hospital. Results: UTI was more common in females (68.39%) than in males (31.61%). Among the uropathogens isolated Escherichia coli (67.6%) was found to be the predominant organism followed by Staphylococcus aureus (12.49%), Klebsiella species (8.23%), and Staphylococcus saprophyticus (6.83%) of total cases. The most common isolates were E. coli showed high sensitivity to ticgeyccline (97.32%), followed by meropenem (95.88%) and imipenem (95.68%). It was found to be highly resistant to ceftriaxone (62.89%) and ceftazidime (54.12%). Conclusion: In conclusion, Escherichia coli species is the most common organism causing UTI with high resistance to the commonly prescribed antimicrobial drugs.

Keywords: Urinary tract infection; bacterial isolate; antimicrobial resistance; urine culture

Introduction

Urinary tract infections (UTIs) are one of the most common bacterial infections in humans both in community and hospital setting¹. It is also one of the important cause of morbidity and the second most common cause of hospital visit². UTI can be asymptomatic or symptomatic, characterized by a wide range of symptoms ranging from mild painful voiding to bacteremia, sepsis, or even death³. Escherichia coli is responsible for community-acquired UTIs at about 80.0% to 85.0% and Staphylococcus saprophyticus accounts for 5.0% to 10.0% cases⁴. Sometimes UTIs cases are treated empirically and the antibiotic resistance patterns of the urinary pathogens determine the empirical therapy⁵. However, uncontrolled antimicrobials usage in high proportion has contributed to the emergence of microbial resistance. As a result, antimicrobial resistance is the more prevalent worldwide among urinary pathogens⁶-⁷. For this reason, having the knowledge of the changes in antibiotic resistance patterns in specific geographical locations may help clinicians to choose
the empirical antimicrobial treatment appropriately. This study was conducted to find out the prevalence of UTI and to determine the antimicrobial susceptibility patterns of commonly used antibiotics.

Methodology

Study Settings & Population: This retrospective study was conducted in the Department of Microbiology at Enam Medical College, Savar, Dhaka, Bangladesh from January 2020 to December 2020 on 6775 untreated patients with clinical symptoms of UTI referred to a private hospital, Farazy Hospital Ltd, Dhaka, Bangladesh. Ethical clearance was taken from the local Institutional Review Board (IRB) of Enam Medical College, Savar, Dhaka, Bangladesh. This study was conducted according to the Declaration of Helsinki.

Specimen Collection and Bacterial Isolates: Clean catch midstream urine samples (MSU) were collected in sterile disposable containers (4 to 5 ml) and transported immediately to the laboratory. Urine specimens were subjected to general urine examinations using direct microscopy for white blood cell (WBC) counting. Urine samples were cultured on 5.0% blood agar and MacConkey agar (Oxoid Ltd, Bashingstore, Hampire, UK) using calibrated loops for semi-quantitative method and incubated in aerobic conditions for 24 hours at 37°C. Cultures without any colony at the end of 24 hours incubation were further incubated for 48 hours. Samples with colony count equal or more than 10³ CFU/ml were considered positive. The isolates were identified and confirmed using standard microbiological methods including Gram staining, colonial morphology on media, growth on selective media, lactose and mannitol fermentation, H₂S production, catalase, oxidase, coagulase, indole, and citrate utilization, and urease test.

Antimicrobial Susceptibility Testing: Antimicrobial susceptibility testing was performed on Mueller-Hinton agar (Oxoid Ltd, Bashingstore, Hampire, UK) using disk diffusion (Kirby Bauer’s) technique according to Clinical and Laboratory Standards Institute (CLSI) guidelines. The antibiotic discs and their concentrations consisted of Ceftriaxone (CTX, 30μg), Imipenem (IPM, 10μg), Ciprofloxacin (Cip, 5μg), Ceftazidime (CAZ, 30μg), Gentamicin (GM, 10μg), Amikacine (AK, 10μg), Trimethoprim-sulfamethoxazole (SXT, 30μg), Nitrofurantoin (FM, 50μg) and, Nalidixic acid (NA, 30μg) for Gram negative isolates, and Chloramphenicol (C, 30μg), Ampicillin (AM, 10μg), Erythromycin (E, 15μg), Norfloxacin (NOR, 10μg), Gentamicin(GM, 10μg), Vancomycin (V, 30μg), Nitrofurantoin (FM, 50μg), Tetracycline (TE, 30μg), and Trimethoprim-sulfamethoxazole (SXT, 30μg) for Gram positive isolates all obtained from Oxoid Ltd, Bashingstore, Hampire, UK. E. coli (ATCC 25922) and Staphylococcus aureus (ATCC 25923) were used as quality control strains.

Statistical Analysis: Statistical analysis was performed by Statistical Package for Social Sciences (SPSS) version 22.0. Qualitative data were expressed as frequency and percent. The quantitative data were expressed as mean with standard deviation.

Ethical Clearance: All procedures of the present study were carried out in accordance with the principles for human investigations (i.e., Helsinki Declaration) and also with the ethical guidelines of the Institutional research ethics. Formal ethics approval was granted by the local ethics committee.

Results

A total of 6775 urine samples were analyzed for culture and sensitivity during the study period, of which 1433 (21.15%) had significant bacteriuria. The rate of positive culture was 980 (68.39%) for female subjects and 453 (31.61%) for male subjects (Figure 1).

Figure 1: Sex wise distribution of UTI in percentage

In this study, the Gram-negative bacilli which accounts for 79.7% and Gram positive cocci 20.3% of the total positive isolates have been isolated respectively. Among the gram-negative bacilli, the predominant isolate was the Escherichia coli (n=970, 67.6%) followed by Klebsiella spp. (n=118, 8.23%), and Acinetobacter spp. (n=22, 1.53%) among the major isolates. Among the gram-positive bacteria, the main
organism identified was *Staphylococcus aureus* (n=179, 12.49%) (Table 1).

Table 1: Distribution of Uropathogens Isolated in the Study

<table>
<thead>
<tr>
<th>Name of the isolates</th>
<th>Frequency</th>
<th>Percent</th>
</tr>
</thead>
<tbody>
<tr>
<td><em>Escherichia coli</em></td>
<td>970</td>
<td>67.7</td>
</tr>
<tr>
<td><em>Klebsiella spp.</em></td>
<td>118</td>
<td>8.23</td>
</tr>
<tr>
<td><em>Pseudomonas spp.</em></td>
<td>27</td>
<td>1.88</td>
</tr>
<tr>
<td><em>Acinetobacter spp.</em></td>
<td>22</td>
<td>1.54</td>
</tr>
<tr>
<td><em>Citrobacter spp.</em></td>
<td>05</td>
<td>0.35</td>
</tr>
<tr>
<td><em>Staphylococcus aureus</em></td>
<td>179</td>
<td>12.49</td>
</tr>
<tr>
<td><em>Staphylococcus saprophyticus</em></td>
<td>98</td>
<td>6.84</td>
</tr>
<tr>
<td><em>Enterococcus spp.</em></td>
<td>14</td>
<td>0.97</td>
</tr>
</tbody>
</table>

Table 2: Antimicrobial Resistance of Gram Negative Uropathogens isolated in this study

<table>
<thead>
<tr>
<th>Organism</th>
<th>AMC</th>
<th>Cip</th>
<th>Imp</th>
<th>MEM</th>
<th>GM</th>
<th>AK</th>
<th>F</th>
<th>CT</th>
<th>CXM</th>
<th>CFM</th>
<th>CTX</th>
<th>CAZ</th>
<th>CRO</th>
<th>NET</th>
<th>TZP</th>
<th>TGC</th>
</tr>
</thead>
<tbody>
<tr>
<td><em>E. coli</em></td>
<td>9.48</td>
<td>33.5</td>
<td>4.32</td>
<td>4.12</td>
<td>44.85</td>
<td>42.78</td>
<td>18.56</td>
<td>2.57</td>
<td>18.76</td>
<td>33.51</td>
<td>42.47</td>
<td>54.12</td>
<td>62.89</td>
<td>29.07</td>
<td>11.55</td>
<td>2.68</td>
</tr>
<tr>
<td><em>Klebsiella spp.</em></td>
<td>9.32</td>
<td>52.54</td>
<td>8.47</td>
<td>8.47</td>
<td>51.69</td>
<td>54.23</td>
<td>58.47</td>
<td>13.56</td>
<td>42.37</td>
<td>52.54</td>
<td>44.92</td>
<td>50</td>
<td>58.47</td>
<td>10.17</td>
<td>5.08</td>
<td>8.47</td>
</tr>
<tr>
<td><em>Pseudomonas spp.</em></td>
<td>37.03</td>
<td>77.78</td>
<td>29.63</td>
<td>29.63</td>
<td>59.26</td>
<td>66.67</td>
<td>74.07</td>
<td>18.52</td>
<td>51.85</td>
<td>62.96</td>
<td>70.37</td>
<td>77.77</td>
<td>81.48</td>
<td>22.22</td>
<td>29.63</td>
<td>14.81</td>
</tr>
<tr>
<td><em>Acinetobacter spp.</em></td>
<td>54.55</td>
<td>95.45</td>
<td>45.45</td>
<td>54.55</td>
<td>90.90</td>
<td>90.90</td>
<td>90.90</td>
<td>9.09</td>
<td>90.90</td>
<td>81.81</td>
<td>86.36</td>
<td>86.36</td>
<td>86.36</td>
<td>86.18</td>
<td>54.55</td>
<td>31.82</td>
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<tr>
<td><em>Citrobacter spp.</em></td>
<td>20</td>
<td>40</td>
<td>0</td>
<td>40</td>
<td>40</td>
<td>60</td>
<td>0</td>
<td>40</td>
<td>60</td>
<td>60</td>
<td>60</td>
<td>20</td>
<td>0</td>
<td>20</td>
<td>0</td>
<td>20</td>
</tr>
</tbody>
</table>

Table 3: Antimicrobial Resistance of Gram Positive Uropathogens isolated in this study

<table>
<thead>
<tr>
<th>Organism</th>
<th>AMC</th>
<th>Cip</th>
<th>Imp</th>
<th>MEM</th>
<th>GM</th>
<th>AK</th>
<th>F</th>
<th>CT</th>
<th>CXM</th>
<th>CFM</th>
<th>CTX</th>
<th>CAZ</th>
<th>CRO</th>
<th>NET</th>
<th>TZP</th>
<th>TGC</th>
</tr>
</thead>
<tbody>
<tr>
<td><em>Staph. aureus</em></td>
<td>6.7</td>
<td>26.81</td>
<td>3.35</td>
<td>2.79</td>
<td>12.29</td>
<td>16.20</td>
<td>14.53</td>
<td>2.79</td>
<td>29.05</td>
<td>34.64</td>
<td>40.22</td>
<td>43.02</td>
<td>51.39</td>
<td>5.58</td>
<td>4.47</td>
<td>5.59</td>
</tr>
<tr>
<td><em>Staph. saprophyticus</em></td>
<td>6.12</td>
<td>22.45</td>
<td>2.04</td>
<td>2.04</td>
<td>10.20</td>
<td>10.20</td>
<td>8.16</td>
<td>0</td>
<td>12.25</td>
<td>18.37</td>
<td>19.38</td>
<td>23.47</td>
<td>29.59</td>
<td>12.25</td>
<td>4.08</td>
<td>4.08</td>
</tr>
<tr>
<td><em>Enterococcus spp.</em></td>
<td>42.85</td>
<td>64.29</td>
<td>14.29</td>
<td>14.29</td>
<td>57.14</td>
<td>57.14</td>
<td>50</td>
<td>7.14</td>
<td>71.43</td>
<td>71.43</td>
<td>57.14</td>
<td>50</td>
<td>78.57</td>
<td>21.43</td>
<td>28.57</td>
<td>14.28</td>
</tr>
</tbody>
</table>

The most common Gram negative isolate *E. coli* showed high resistant to ceftriaxone (62.89%) and ceftazidime (54.12%) with good susceptibility to ticarcyl (97.32%), imipenem (95.68%), meropenem (95.88%), Colistin (97.43%) and amoxyclav (90.52%) respectively. The second most common isolates among Gram negative isolates, *Klebsiella spp.* showed high sensitivity to piperacillin-tazobactam (95%), imipenem (92%), meropenem (92%), ticarcyl (92%) and colistin (86%). It showed high resistance to ceftriaxone (58%), nitrofurantoin (58%), amikacin (54%), cefixime (53%) and ciprofloxacin (53%). *Pseudomonas spp.* which is the 3rd most predominant Gram negative isolate gave high sensitivity to Ticarcyl (85%), colistin (81%) and netilmicin 78%. Among the tested antibiotics it showed high resistance to ceftriaxone (81.48%), ciprofloxacin and ceftazidime (77.78%) and nitrofurantoin (74.07%) (Table 2).

Discussion

Urinary tract infections are common clinical conditions worldwide and the pattern of antimicrobial resistance varies in different regions. This present study describes the relationships between isolated bacterial agents and antibiotic resistance of UTIs. The sex distribution of patients in our study is almost consistent with those of other reported studies, showing a statistically predominance of females with UTI (68.39% of the positive cultures). This result is almost similar to those reported from many other centers. Approximately 1 in 3 women will require antimicrobial treatment for a UTI before age 24, and 40% to 50% of women will suffer from UTI during their lifetime. It is assumed that the short urethra in girls predisposes them to ascending infection. Furthermore, management of mic-turition in women is important. Management mistakes made by women include cleaning perineum forward from the anus to the vulva that can cause urinary tract infection.
Sexual activity has been reported to influence higher prevalence of UTI in females\(^1\). Considering the fact that most of infecting organisms are commensals of perianal and vaginal regions, emphasis on personal hygiene especially in females may be important in reducing the incidence of UTI\(^2\). Males are less prone to UTIs possibly because of their longer urethra and the presence of antimicrobial substances in prostatic fluid\(^3\).

The results of our study show that among the heterogeneous causative organisms of UTI, Enterobacteriaceae are the predominant pathogens and E. coli is still the single most common uropathogen. This corresponds with the data obtained by other investigators\(^\text{10-18}\). In addition, coagulase negative Staphylococcus spp. was the most common cause of UTI among Gram positive bacteria. Recent studies have revealed the importance of coagulase negative Staphylococcus species in urinary tract infections\(^1\). Escherichia coli (67.6\%) is the most common organism causing urinary tract infections in this study followed by Klebsiella species (8.23\%) among gram negative isolates. This is in accordance with earlier study Ranjbar et al, and Amin et al\(^\text{20,21}\). The highest percentages of resistance of Escherichia coli causing urinary tract infections were found for ceftriaxone (62.89\%), ceftazidime (54.12\%), gentamicin (44.85\%), amikacin (42.78\%), cefotaxime (42.47\%) whereas the highest percentages of sensitivity were seen for colistin (97.43\%), Tigecycline (97.32\%), meropenem (95.88\%), imipenem (95.68\%), and amoxyclav (90.52\%).

Over the last decade, the treatment of choice for UTIs has changed owing to the rate of resistance and high level of therapeutic failure\(^2\). This study revealed a higher prevalence rate of resistance among E. coli to the commonly prescribed antibiotic agents such as ceftriaxone, ceftazidime, gentamicin, amikacin and ciprofloxacin. The low susceptibility like higher resistance of the isolates to the common and comparatively cheap administered antibiotics is not surprising because these drugs are more commonly misused, thereby leading to the development of resistance, as previously reported\(^2\). Increased resistance in quinolones against E. coli may reflect the overuse of these drugs for the treatment of UTI\(^1\). Thus reducing the number of prescription for a particular antibiotic can lead to a decrease in resistance rates. Another factor could be the generalized use of fluoroquinolone in animals feed especially in poultry and the subsequent transmission of resistant to strains from animals to humans\(^2\). These findings are of great importance and imply that these antibiotics cannot be used as empirical therapy for UTI, particularly in the study area. On the other hand, while lower resistance was detected to colistin, tigecycline, imipenem, and meropenem. The low resistance could be because they are not easily accessible and relatively expensive in price compared to others. Thus, these drugs could be considered as alternative options in the empirical treatment of serious UTI patients.

In the last decades, the number of reports about appearance of bacteria with antibiotic resistance has increased all over the world\(^2\). This study, like others, shows clearly that there are significant geographic differences in the susceptibility of commonly used antimicrobials against UTIs\(^2\). As a result, accurate and appropriate knowledge on local epidemiology and patterns of antimicrobial resistance in uropathogens are essential to design a clinically effective therapy for UTI patients. However in this study, we observed a highly resistance to commonly used antibiotics particularly among Gram negative isolates. Studies like the present study are useful in determining local trends and risk factors for antimicrobial resistance and furthermore it is important to notify physicians and other health workers on performing antibiotic susceptibility test before giving blind antibiotic therapy.

**Conclusion**

Urinary tract infections are one of the most common causes for seeking medical attention in the community and effective management of patients depends on the identification of the type of organisms that cause the disease and the selection of an appropriate antibiotic agent to the organism in question. This study provides valuable data to compare and monitor the status of antimicrobial resistance. Thus, amoxyclav was found to be the most appropriate oral antibiotic and tigecycline, colistin imipenem and meropenem were the most appropriate parenteral antibiotics, for the empirical therapy of UTIs.

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None

**Conflict Of Interest**

The authors have no conflicts of interest to disclose.

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**Authors’ contributions**

Uddin BMM, Roy S, Hossen MM conceived and designed the study, analyzed the data, interpreted the results, and wrote up the draft manuscript. Akhand F, Bari S, Chowdhury SJ, Mahjabin M, Salman MN...
contributed to the analysis of the data, interpretation of the results and critically reviewing the manuscript. Uddin BMM involved in the manuscript review and editing. All authors read and approved the final manuscript.

Data Availability
Any inquiries regarding supporting data availability of this study should be directed to the corresponding author and are available from the corresponding author on reasonable request.

Ethics Approval and Consent to Participate
Ethical approval for the study was obtained from the Institutional Review Board. As this was a prospective study the written informed consent was obtained from all study participants. All methods were performed in accordance with the relevant guidelines and regulations.

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