Antibiotic profile of enterobacter isolates from various clinical samples – A study from a tertiary care hospital

Vinay Kumar¹, Smitha Rockey²,*

¹M.Sc, ²Assistant Professor, Dept. of Microbiology, St. Johns Medical College & Hospital, Bangalore, Karnataka, India

*Corresponding Author:
Email: smitharockey@rediffmail.com

Abstract
Introduction: Enterobacter species, especially Enterobacter cloacae are important nosocomial pathogens. They cause infections of the lower respiratory tract, skin, soft tissue, urinary tract and occasionally sepsis. Enterobacter species are often seen in natural habitats like water and soil. Enterobacter species are often resistant to various antibiotics in a hospital setting. Resistance to antimicrobials can also develop during therapy. The emergence of antimicrobial resistance, especially to third generation cephalosporins is a concern in the management of Enterobacter infections.

Objectives: To evaluate the antimicrobial resistance pattern of Enterobacter species isolated from patients attending a tertiary care hospital.

Materials and Methods: The present study was conducted in the Microbiology department of a tertiary care hospital. The study was conducted from January 2012 to December 2012. Enterobacter species were isolated from the clinical samples of patients using standard microbiological methods of bacterial culture and identification. Antibiotic susceptibility testing was done by Kirby-Bauer disc diffusion method, according to Clinical laboratory standard institute (CLSI) guidelines. Minimum inhibitory concentration (MIC) of cefotaxime was determined by the micro-broth dilution method.

Results: During the study period, 100 isolates of Enterobacter species were isolated from clinical samples. The predominant species of Enterobacter isolated was Enterobacter cloacae (69%), followed by Enterobacter aerogenes (25%). Antibiotic susceptibility testing showed that most of the isolates were resistant to ampicillin (86%). The resistance to cefotaxime was 33%.

Conclusion: Enterobacter species in a tertiary care setting could be resistant to common antibiotics like ampicillin and cephalosporins. Clinicians should be aware of these resistance patterns while choosing an empiric antibiotic regimen.

Keywords: Enterobacter species, Enterobacter cloacae, Enterobacter aerogenes, Antimicrobial resistance pattern, cephalosporin resistance.

Introduction
Enterobacter species are one of the frequently isolated Gram-negative bacilli in a Microbiology laboratory. These bacteria are emerging as important causes of nosocomial infections like urinary tract infections, wound infections, pneumonia, bacteremia and meningitis. Enterobacter species are often found in natural habitats like water and soil.¹

Enterobacter species are important as causes of nosocomial infections. They are also implicated in various community-acquired infections. Enterobacter species are becoming resistant to many antibiotics.² The information on the characterization and antibiogram of Enterobacter species causing human infections is limited. Hence the present study is designed to characterize the Enterobacter species isolated from the clinical samples of patients and to analyze their antimicrobial susceptibility pattern.

Materials and Methods
This was a prospective study conducted at the Department of Microbiology, St John’s Medical College and Hospital, Bengaluru, south India, from January 2012 to December 2012. Enterobacter species isolated in the laboratory from various clinical samples of patients treated in the hospital were included in the study. These clinical samples included urine, pus, blood, sputum and sterile body fluids. Samples included those from patients admitted in the hospital as well as those from patients treated as outpatients. The isolates that were presumptively identified as Enterobacter species were subjected to further biochemical tests for speciation. Antibiotic susceptibility testing was done by Kirby- Bauer Disc diffusion method, according to Clinical Laboratory Standard Institute (CLSI) guidelines. Minimum inhibitory concentration (MIC) of cefotaxime was determined for 50 isolates by the micro-broth dilution method according to Clinical Laboratory Standard Institute (CLSI) guidelines.

Results
100 isolates of Enterobacter species were collected from various clinical samples during the study period. These isolates were from pus (36%), urine (23%), sputum (17%), blood (6%), sterile body fluids (2%) and miscellaneous samples (16%) including ear swab and cervical swab(Table 1).
Table 1: Clinical specimens from which Enterobacter were isolated

<table>
<thead>
<tr>
<th>Clinical specimen</th>
<th>Number</th>
</tr>
</thead>
<tbody>
<tr>
<td>Pus</td>
<td>36/100</td>
</tr>
<tr>
<td>Urine</td>
<td>23/100</td>
</tr>
<tr>
<td>Sputum</td>
<td>17/100</td>
</tr>
<tr>
<td>Blood</td>
<td>6/100</td>
</tr>
<tr>
<td>Sterile body fluids</td>
<td>2/100</td>
</tr>
<tr>
<td>Miscellaneous (Ear swab/ Eye swab/ Cervical swab)</td>
<td>16/100</td>
</tr>
</tbody>
</table>

Most of the isolates were from patients admitted to various wards (72%), while 6% of the isolates were from patients admitted to the intensive care units (ICU). Remaining isolates (22%) were from patients treated in the out-patient department (OPD). 76% of the patients were males. The mean age of the patients was 38 years (18-75 years).

These isolates were further analyzed by biochemical tests to confirm their identification and for speciation. Out of 100 isolates, 69 isolates were identified as Enterobacter cloacae, 25 isolates were identified as Enterobacter aerogenes, three isolates were identified as Enterobacter intermedium, two isolates were identified as Enterobacter kobei and one isolate was identified as Enterobacter cancerogenus.

Antibiotic susceptibility testing was done for all 100 isolates using Kirby Bauer disc diffusion method. Sensitivity to the antibiotics are given in the Table 2.

MIC of cefotaxime was determined for 50 isolates of Enterobacter species using micro-broth dilution method according to CLSI guidelines. Out of these 50 isolates, 25 isolates were sensitive, and 25 isolates were resistant to cefotaxime by disc diffusion method. The results of both methods were correlating. Out of the 25 resistant isolates, eight isolates showed an MIC value of 16µg/ml, 12 isolates showed an MIC of 32µg/ml and five isolates showed an MIC of > or = 64µg/ml.

Table 2: Antibiotic susceptibility of Enterobacter isolates

<table>
<thead>
<tr>
<th>Antimicrobial agent</th>
<th>Sensitivity</th>
</tr>
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<tbody>
<tr>
<td>Ampicillin</td>
<td>14%</td>
</tr>
<tr>
<td>Cotrimoxazole</td>
<td>73%</td>
</tr>
<tr>
<td>Ciprofloxacin</td>
<td>87%</td>
</tr>
<tr>
<td>Gentamicin</td>
<td>64%</td>
</tr>
<tr>
<td>Amikacin</td>
<td>73%</td>
</tr>
<tr>
<td>Netilmicin</td>
<td>84%</td>
</tr>
<tr>
<td>Cephalexin</td>
<td>43%</td>
</tr>
<tr>
<td>Cefotaxime</td>
<td>33%</td>
</tr>
<tr>
<td>Ceftazidime</td>
<td>52%</td>
</tr>
<tr>
<td>Piperacillin-Tazobactam</td>
<td>74%</td>
</tr>
<tr>
<td>Meropenem</td>
<td>78%</td>
</tr>
</tbody>
</table>
Discussion

Enterobacter species are often isolated in the Microbiology laboratory. Enterobacter species are present in various natural habitats. They are also able to survive on skin and dry surfaces. These factors help them in causing nosocomial infections. The incidence of nosocomial infections due to Enterobacter is increasing and is an important cause of concern due to the emergence of resistance to commonly used antibiotics like penicillins, third generation cephalosporins and quinolones.

In the present study, 100 isolates of Enterobacter species were obtained from different clinical samples. These clinical samples were pus (36%), urine (23%), sputum (17%), blood (6%), sterile body fluids (2%) and Miscellaneous samples (16%) including ear swab and cervical swabs. In a previous study on Enterobacter isolates, 42% were from pus, 41% from urine, 15% from blood and 0.9% from CSF.

Enterobacter cloacae (69%) was the predominant species of Enterobacter in our study and this finding correlated with previous studies. Enterobacter cloacae is widely distributed in the environment and may be a part of normal gut flora in most of the individuals. Other Enterobacter species isolated in this study are Enterobacter aerogenes (25%), Enterobacter intermedius (3%), Enterobacter kobei (2%) and Enterobacter cancerogenus (1%).

In the present study, 6% Enterobacter species were isolated from cases of bacteremia. A previous study showed that the rate of Enterobacter bacteremia was 3.9 per 1000 admitted patients. In a study by Chen et al, Enterobacter ranked fourth as cause of bacteremia, accounting for 11.3% of bacteremia.

Antibiotic susceptibility testing using Kirby-Bauer disc diffusion method was done for all the isolates. 14% of the isolates in this study were sensitive to ampicillin. This finding shows a higher sensitivity to the antibiotic compared to a previous study where only 5.4% of the Enterobacter isolates were sensitive to ampicillin. But sensitivity to ciprofloxacin (87%) in this study correlates with the findings of previous studies. In the present study, 78% Enterobacter isolates were sensitive to meropenem where as in a previous study 100% of the isolates were sensitive to the antibiotic.

Resistance to cefotaxime was found to be 33% in this study. In a previous study, 46.5% Enterobacter isolates were resistant to cefotaxime. In a study of Enterobacter blood stream infections by Kang et al, cephalosporin resistance was seen in 47% of isolates. Enterobacter species has emerged as one of the commonest Gram-negative bacilli resistant to third generation cephalosporins.

MIC of cefotaxime was determined for 50 isolates by micro broth dilution method. Out of these 50 isolates, 25 isolates were sensitive to cefotaxime and 25 isolates were resistant to the antibiotic by disc diffusion method. The results of disc diffusion and micro broth dilution methods were compared and were found to be correlating. Out of the 25 isolates, which were resistant to cefotaxime, 5 isolates (25%) showed an MIC > or = 64 mg/L. In a previous study by Mordi et al, where results of MIC of cefotaxime were available for 25 Enterobacter isolates, 72% isolates showed an MIC > or = 64mg/L. While multi-drug resistance in our study was 56%, it was 72.9% in a previous study. In our study, five out of six strains isolated from ICU were multi-drug resistant. This correlates with previous findings that drug-resistant Enterobacter species are more likely to be isolated from patients in ICUs compared to other areas in the hospital.

Enterobacter infections caused by strains which are resistant to third generation cephalosporins are associated with higher mortality. Development of resistance is seen in Enterobacter species during antimicrobial therapy with expanded spectrum cephalosporins. In a study on Enterobacter bacteremia by Chow et al, it was shown that an overall rate of resistance emerged during therapy with third generation cephalosporin was 19%. Previous use of extended-spectrum cephalosporins was associated with resistance to these drugs in patients with Enterobacter bacteremia.

One of the limitations of our study was its small sample size. We did not stratify the specimens according to the wards. Information on antibiotic treatment and outcome of the patients was not available. Species wise comparison of the susceptibility pattern was also not available. Further studies with larger sample size could be helpful.

Conclusion

Enterobacter species especially Enterobacter cloacae is an important cause of nosocomial infection recent years. Most of these isolates are resistant to many antibiotics. They can also develop resistance during antimicrobial therapy. The resistance of Enterobacter species to cephalosporins and newer
broad-spectrum agents can be a major problem in the treatment of Enterobacter infections.

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**References**


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