Reliability of Kirby-Bauer Disk Diffusion Method for Detecting Carbapenem Resistance in \textit{Acinetobacter baumannii-calcoaceticus} Complex Isolates

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We recently read an article on carbapenem susceptibility testing errors in studies that used three automated systems, disk diffusion (DD), Ettest (ET), and broth microdilution (BMD), to investigate \textit{Acinetobacter baumannii-calcoaceticus} complex (ABC) isolates (1). The authors concluded from the study that the manual methods are more accurate than automated methods. The error rates for testing susceptibility to imipenem and meropenem by DD and the Ettest were found to be within an acceptable range compared with MIC detected by BMD. Carbapenems are widely used against multidrug-resistant (MDR) ABC isolates (2). However, the pathogen is associated with increasing carbapenem resistance, which limits therapeutic options and challenges effective hospital infection control. Thus, as antimicrobial resistance increases, accurate susceptibility testing to guide therapeutic options is essential. Agar microdilution and broth microdilution for detecting carbapenem resistance are the recommended methods, but they are impractical to implement as routine tests in many clinical laboratories with a high isolate load that cannot afford automated susceptibility testing or the Ettest. The Kirby-Bauer disk diffusion (KBDD) method is more economical and the standardized one used for testing susceptibility to most of the antibiotics. However, only a few studies have provided data on its reliability for testing carbapenems against ABC isolates (1, 3).

In the present study, the KBDD method using discs (10 μg each) of imipenem (BD), meropenem (Hi-Media), and doripenem (BD) was compared with the bioMérieux Ettest (0.002 to 32 μg/ml) on 124 nonrepeat MDR ABC strains isolated from blood, respiratory, and pus specimens. The isolates were identified using standard bacteriological procedures (4), and interpretation of susceptibility testing was done according to CLSI guidelines (5). \textit{Escherichia coli} ATCC 25922 and \textit{Pseudomonas aeruginosa} ATCC 27853 were used as the quality control strains. Taking the Ettest as the reference, discordance between it and the KBDD method was categorized as a very major error (reported susceptible when resistant), a major error (reported resistant when susceptible), or a minor error (reported intermediate when resistant or susceptible or vice versa). By the use of CLSI and FDA breakpoints for imipenem and meropenem, no discrepancy was found between the KBDD method and the Ettest in the case of imipenem. However, in the case of meropenem, nine strains that were resistant by the KBDD method came out to be intermediate sensitive by the Ettest. Thus, the KBDD method showed a minor error rate of 7.3% and no very major or major error for meropenem was recorded, which is acceptable. CLSI does not provide breakpoints for doripenem, but when the same breakpoints used for imipenem and meropenem were applied to doripenem, a major error of 2.5% and a minor error of 21.8% were observed, proving the lower accuracy rate of the KBDD method. According to EUCAST breakpoints (Table 1), no discrepancy was found between the KBDD method and the Ettest for all the three carbapenems, though minor error rates could not be calculated for doripenem, since intermediate breakpoints are not defined. Joseph et al. (3) showed that the KBDD method can be reliably used for routine testing of meropenem in ABC isolates.

Despite of the limitation of the study that the Ettest was used as the reference method for MIC determination, our results support the finding that the KBDD method can be reliably used for routine testing of carbapenem susceptibility in ABC isolates. However, an international standard is required for doripenem breakpoints to further support this finding.

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REFERENCES


TABLE 1 Susceptibility breakpoints

<table>
<thead>
<tr>
<th>Antibiotic</th>
<th>CLSI MIC (μg/ml)</th>
<th>Disk (mm)</th>
<th>FDA MIC (μg/ml)</th>
<th>Disk (mm)</th>
<th>EUCAST MIC (μg/ml)</th>
<th>Disk (mm)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Imipenem</td>
<td>≤4</td>
<td>≥16</td>
<td>≤4</td>
<td>≥16</td>
<td>≧2</td>
<td>≥23</td>
</tr>
<tr>
<td>Meropenem</td>
<td>≤4</td>
<td>≥16</td>
<td>≤4</td>
<td>≥16</td>
<td>≧2</td>
<td>≥21</td>
</tr>
<tr>
<td>Doripenem</td>
<td>≤1</td>
<td>≥17</td>
<td>≤1</td>
<td>≥17</td>
<td>≧1</td>
<td>≥21</td>
</tr>
</tbody>
</table>

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