Aerococcus urinae and Aerococcus sanguinicola: Susceptibility testing of 120 isolates to eight antimicrobial agents using disc diffusion, Etest and broth microdilution techniques

Derya Carkaci1,2, Xiaohui Chen Nielsen1, Kurt Fuurstved2, Robert Skov2, Ole Skovgaard3, Anne Bonde Jensen1, Janne Føns Møller1, Hanne Junker1, Emilio Pérez Trallero4, Reto Lienhard5, Jenny Åhlman6, Erika Matuschek6, Gunnar Kahlmeter6 & Jens Jørgen Christensen1

1 Department of Clinical Microbiology, Slagelse Hospital, Slagelse, Denmark
2 Department of Microbiology and Infection Control, Reference Laboratory Statens Serum Institut, Copenhagen, Denmark
3 Department of Science, Systems and Models, Roskilde University, Roskilde, Denmark
4 Department of Microbiology, Hospital Universitario Domusia, San Sebastián, Spain
5 ADMED Microbiology, La Chaux de Fonds, Switzerland
6 EUCAST Development Laboratory, Clinical Microbiology, Central Hospital, Växjö, Sweden

The authors have nothing to disclose.

Introduction & Objective

Antimicrobial susceptibility testing may be challenging when examining fastidious microorganisms, to which Aerococcus urinae (A. urinae) and Aerococcus sanguinicola (A. sanguinicola) belong.

A. urinae and A. sanguinicola isolates have most often been found in urine from patients with urinary tract infections, and more seldom, in blood from patients suffering from sepsis and/or infective endocarditis.

The outcome of patients with A. urinae and A. sanguinicola associated infections is dependent on rapid administration of appropriate antimicrobial therapy.

The objective of this study was to create basis for establishing epidemiological cut-off (ECOFF) values and clinical breakpoints for clinical isolates of A. urinae and A. sanguinicola.

Methods

Clinical isolates of A. urinae (N=83) and A. sanguinicola (N=37) were isolated from urine and blood samples and originated from clinical microbiology departments in Denmark and kindly provided from two collaborators in Switzerland and Spain. All isolates were species identified using MALDI-TOF mass spectrometry.

Three susceptibility testing methods were used: disc diffusion and Etest, according to EUCAST methodology and the Sensititre susceptibility broth microdilution (BMD) methodology from TRISK Diagnostics.

The following antimicrobial agents were tested: Penicillin (benzylpenicillin for disc diffusion and BMD), cefotaxime, meropenem, erythromycin, clindamycin, vancomycin, lincomycin, and rifampicin (only disc diffusion and Etest). The discs and Etest strips were purchased from Oxoid (UK) and bioMérieux (France), respectively. See table 1 for disc and Etest strip information.

Preparation of inocula, inoculation and incubation on plates were performed according to EUCAST methodology using Oxford plates. Incubation was done at 35°C in aerobic atmosphere, 5% CO2 for 16-20 hours.

The susceptibility testing methods were performed in parallel.

Results

<table>
<thead>
<tr>
<th>Antimicrobial agent</th>
<th>A. urinae</th>
<th>A. sanguinicola</th>
</tr>
</thead>
<tbody>
<tr>
<td>Method</td>
<td>Etest</td>
<td>BMD</td>
</tr>
<tr>
<td>Penicillin</td>
<td>1.1</td>
<td>0.08-0.125</td>
</tr>
<tr>
<td>Cefotaxime</td>
<td>2.0</td>
<td>0.06-0.125</td>
</tr>
<tr>
<td>Meropenem</td>
<td>1.5</td>
<td>0.06-0.125</td>
</tr>
<tr>
<td>Erythromycin</td>
<td>1.5</td>
<td>0.06-0.125</td>
</tr>
<tr>
<td>Clindamycin</td>
<td>1.0</td>
<td>0.06-0.125</td>
</tr>
<tr>
<td>Vancomycin</td>
<td>0.01</td>
<td>0.06-0.125</td>
</tr>
<tr>
<td>Lincomycin</td>
<td>1.0</td>
<td>0.06-0.125</td>
</tr>
<tr>
<td>Rifampicin</td>
<td>0.5</td>
<td>0.06-0.125</td>
</tr>
</tbody>
</table>

Table 1: Disc diffusion inhibition zone distribution and MICs for Etest and BMD for the eight tested antimicrobial agents.

Growth was confluent on MH-F plates for all isolates.

Disc diffusion and MICs (Etest and BMD) results could be read after 16-20 h incubation. N=15 A. urinae MICs were read after 48 h incubation due to insufficient growth.

Differences between MICs (Etest and BMD) were within one dilution. No obvious difference in susceptibility pattern was observed between the two species.

Range of disc diffusion inhibition zones and MICs obtained by Etest and BMD can be found in Table 1 for all A. urinae and A. sanguinicola isolates.

Figure 1a show distribution of disc diffusion inhibition zones for benzylpenicillin, figure 1b MIC distributions for Etest penicillin and figure 1c MIC distributions for BMD benzylpenicillin.

Conclusions

- A. urinae and A. sanguinicola isolates had low MICs to all the eight tested antimicrobial agents.
- A good correlation between disc diffusion inhibition zones and MICs were observed for the three susceptibility testing methods of disc diffusion, Etest and BMD.
- This study demonstrated that a relevant basis is present for establishing ECOFF values and clinical breakpoints for A. urinae and A. sanguinicola isolates to penicillin, cefotaxime, meropenem, erythromycin, clindamycin, vancomycin, lincomycin, and rifampicin.

For more information, please contact: deca@regionsjaelland.dk