Haemophilus influenzae

Benzylpenicillin 1 unit as screen for beta-lactam resistance
Haemophilus influenzae
MIC and zone diameter correlates

- The following histograms present inhibition zone diameter distributions from EUCAST antimicrobial susceptibility testing. In most, the different colours of the bars indicate different MIC values. In some, the colours of the bars indicate a resistance gene or a resistance mechanism.

- The distributions include data for wild-type isolates and for isolates with acquired resistance mechanisms. A large number of isolates with MIC values close to the edge of the wild-type distribution and/or close to EUCAST clinical breakpoints were intentionally included. These distributions can not be used to infer resistance rates or the performance of the tests with routine isolates.

- For some agents, isolates were tested on more than one occasion, including parallel tests with disks and media from several manufacturers. When this is the case, data are presented as both the “number of isolates tested” and the “total number of MIC-zone diameter correlates”, including replicate tests and parallel tests with disks and media from different sources.
Haemophilus influenzae
Materials and methods

• Antimicrobial susceptibility testing was performed on a collection of *Haemophilus influenzae*, including many isolates with beta-lactam resistance. Disk diffusion was performed on MH-F media according to EUCAST methodology and MIC determination was performed with the ISO broth microdilution method using MH-F broth.

• The following graphs present inhibition zone distributions for benzylpenicillin 1 unit with MIC values for clinically important beta-lactam agents or resistance mechanisms as coloured bars. A nitrocefin-based disk test was used to analyse β-lactamase production and PCR was used to detect PBP mutations.

• The distributions in this presentation are the result of a collaboration between EUCAST, JMI Laboratories (USA), Laboratory Specialists Inc. (USA) and Sykehuset i Vestfold, Tønsberg (Norway).

• This presentation is based on EUCAST Clinical Breakpoint Tables v. 10.0.
Changes from previous version (5.0)

<table>
<thead>
<tr>
<th>Changes</th>
</tr>
</thead>
<tbody>
<tr>
<td>• Information added on the use of benzylpenicillin 1 unit as screen for beta-lactam resistance.</td>
</tr>
<tr>
<td>• Flow chart updated.</td>
</tr>
<tr>
<td>• MIC breakpoints changed for amoxicillin oral, amoxicillin-clavulanic acid oral and cefuroxime oral.</td>
</tr>
<tr>
<td>• Data added for piperacillin-tazobactam.</td>
</tr>
<tr>
<td>• New distribution for benzylpenicillin vs. meropenem meningitis.</td>
</tr>
</tbody>
</table>
**Haemophilus influenzae**

Screen for beta-lactam resistance

- The benzylpenicillin 1 unit disk screen test shall be used to exclude beta-lactam resistance mechanisms.

- The screening breakpoint for benzylpenicillin 1 unit is $S \geq 12$ mm.

- When the screen is negative, all beta-lactam agents for which clinical breakpoints are available, including those with a “Note”, can be reported susceptible without further testing, except for oral preparations of amoxicillin, amoxicillin-clavulanic acid and cefuroxime, which if reported, should be reported “susceptible, increased exposure” (I).

- When the screen is positive, see flow chart for interpretation.
**Haemophilus influenzae**

Screen for beta-lactam resistance

**Benzylpenicillin (PCG) 1 unit disk test**

Always perform in parallel with testing of other beta-lactam agents

- **PCG 1 unit zone diameter ≥ 12 mm**
  - Excludes all beta-lactam resistance mechanisms
  - Report susceptible (S) to any beta-lactam agents for which clinical breakpoints are available, including those with “Note”, except for the oral preparations of amoxicillin, amoxicillin-clavulanic and cefuroxime, which if reported, should be reported “susceptible, increased exposure” (I).

- **PCG 1 unit zone diameter < 12 mm**
  - Beta-lactam resistance mechanism detected (Beta-lactamase and/or PBP3 mutations)
  - Test for beta-lactamase

  - **Beta-lactamase positive**
    - With or without PBP3 mutations
    - Report ampicillin, amoxicillin and piperacillin (without beta-lactamase inhibitor) resistant (R)

  - **Beta-lactamase negative**
    - PBP3 mutations only

  - **Other beta-lactam agents**

- **Amoxicillin-clavulanic acid 2-1 µg ≥ 15 mm**
  - Beta-lactamase only
  - Report susceptible (S) to any beta-lactam agents for which clinical breakpoints are available, including those with “Note”, except for the oral preparations of amoxicillin-clavulanic and cefuroxime, which if reported, should be reported as “susceptible, increased exposure” (I).

- **Amoxicillin-clavulanic acid 2-1 µg < 15 mm**
  - Beta-lactamase and PBP3 mutations
  - Report according to the clinical breakpoints for the agent in question. For cefepime, cefpodoxime and imipenem, see below*

*For cefepime, cefpodoxime and imipenem, if resistant by both screen and agent disk diffusion test, report resistant. If resistant by screen test and susceptible by agent disk diffusion test, determine the MIC of the agent and interpret according to the clinical breakpoints.
Explanation of graphs:

- These graphs show zone diameter distributions with MIC values or resistance mechanisms as coloured bars. Colours are related to current EUCAST MIC breakpoints.

Agent X

- MIC (mg/L)
  - ≥8
  - 4
  - 2
  - 1
  - 0.5
  - 0.25
- Resistant
- Susceptible, increased exposure
- Susceptible, standard dosing regimen

Agent Y

- MIC (mg/L)
  - ≥64
  - 32
  - 16
  - 8
  - 4
  - 2
  - 1
  - 0.5
  - ≤0.25
- Resistant
- Susceptible, increased exposure
Benzylpenicillin 1 unit vs. β-lactam resistance mechanism

*H. influenzae*, 137 clinical isolates

(1 data source)

**Breakpoints**

Benzylpenicillin zone diameter (screen) \( S \geq 12 \text{ mm} \)
**Benzylpenicillin 1 unit vs. Ampicillin MIC**

*H. influenzae*, 167 isolates (212 correlates)

(3 data sources)

**Breakpoints**

Ampicillin MIC

S≤1, R>1 mg/L

Benzylpenicillin zone diameter (screen)

S≥12 mm

**ECOFF**

1 mg/L

**MIC (mg/L)**

- ≥64
- 32
- 16
- 8
- 4
- 2
- 1
- 0.5
- 0.25
- 0.125
- ≤0.06
Benzylpenicillin 1 unit vs. Ampicillin-sulbactam MIC

*H. influenzae*, 145 isolates

(1 data source)

**Breakpoints**

<table>
<thead>
<tr>
<th>Ampicillin-sulbactam MIC</th>
<th>S≤1, R&gt;1 mg/L</th>
</tr>
</thead>
<tbody>
<tr>
<td>Benzylpenicillin zone diameter (screen)</td>
<td>S≥12 mm</td>
</tr>
</tbody>
</table>

**MIC (mg/L)**
- 8
- 4
- 2
- 1
- 0.5
- 0.25
- 0.125
- ≤0.06

**ECOFF**
1 mg/L

MICs with fixed concentration of sulbactam at 4 mg/L.
Benzylpenicillin 1 unit vs. Amoxicillin MIC
H. influenzae, 173 isolates (226 correlates)

(3 data sources)

Breakpoints (iv)
Amoxicillin MIC
S≤2, R>2 mg/L

Benzylpenicillin zone diameter (screen)
S≥12 mm

ECOFF
2 mg/L
Benzylpenicillin 1 unit vs. Amoxicillin MIC
*H. influenzae*, 173 isolates (226 correlates)

(3 data sources)

<table>
<thead>
<tr>
<th>MIC (mg/L)</th>
<th>Breakpoints (oral)</th>
</tr>
</thead>
<tbody>
<tr>
<td>≥128</td>
<td>Amoxicillin MIC S≤0.001, R&gt;2 mg/L</td>
</tr>
<tr>
<td>64</td>
<td>ECOFF 2 mg/L</td>
</tr>
<tr>
<td>32</td>
<td>Benzylpenicillin zone diameter (screen) S≥12 mm</td>
</tr>
</tbody>
</table>
Benzylpenicillin 1 unit vs. Amoxicillin-clavulanic acid MIC

*H. influenzae*, 114 isolates

(2 data sources)

<table>
<thead>
<tr>
<th>MIC (mg/L)</th>
<th>No of observations</th>
</tr>
</thead>
<tbody>
<tr>
<td>≥16</td>
<td>60</td>
</tr>
<tr>
<td>8</td>
<td>50</td>
</tr>
<tr>
<td>4</td>
<td>40</td>
</tr>
<tr>
<td>2</td>
<td>30</td>
</tr>
<tr>
<td>1</td>
<td>20</td>
</tr>
<tr>
<td>0.5</td>
<td>10</td>
</tr>
<tr>
<td>0.25</td>
<td>5</td>
</tr>
<tr>
<td>≤0.125</td>
<td>0</td>
</tr>
</tbody>
</table>

**Breakpoints (iv)**

- **Amoxicillin-clavulanic acid MIC**: S≤2, R>2 mg/L
- **Benzylpenicillin zone diameter (screen)**: S≥12 mm
- **ECOFF**: 2 mg/L
Benzylpenicillin 1 unit vs. Amoxicillin-clavulanic acid MIC

*H. influenzae*, 114 isolates

(2 data sources)

MICs with fixed concentration of clavulanic acid at 2 mg/L.

**Breakpoints (oral)**

- Amoxicillin-clavulanic acid MIC: S≤0.001, R>2 mg/L
- Benzylpenicillin zone diameter (screen): S≥12 mm

**ECOFF**

2 mg/L
Benzylpenicillin 1 unit vs. Piperacillin-tazobactam MIC

*H. influenzae*, 158 isolates

(2 data sources)

**Breakpoints**

<table>
<thead>
<tr>
<th>Breakpoint Description</th>
<th>S ≤ 0.25, R &gt; 0.25 mg/L</th>
<th>ECOFF</th>
</tr>
</thead>
<tbody>
<tr>
<td>Piperacillin-tazobactam MIC</td>
<td></td>
<td>0.06 mg/L</td>
</tr>
<tr>
<td>Benzylpenicillin zone diameter (screen)</td>
<td>S ≥ 12 mm</td>
<td></td>
</tr>
</tbody>
</table>
Benzylpenicillin 1 unit vs. Cefepime MIC

*H. influenzae*, 189 isolates

(2 data sources)

<table>
<thead>
<tr>
<th>MIC (mg/L)</th>
<th>No. of observations</th>
</tr>
</thead>
<tbody>
<tr>
<td>2</td>
<td>1</td>
</tr>
<tr>
<td>1</td>
<td>0</td>
</tr>
<tr>
<td>0.5</td>
<td>2</td>
</tr>
<tr>
<td>0.25</td>
<td>3</td>
</tr>
<tr>
<td>0.125</td>
<td>2</td>
</tr>
<tr>
<td>0.06</td>
<td>1</td>
</tr>
<tr>
<td>0.03</td>
<td>0</td>
</tr>
</tbody>
</table>

**Breakpoints**

- **Cefepime MIC**: $S \leq 0.25$, $R > 0.25$ mg/L
- **Benzylpenicillin zone diameter (screen)**: $S \geq 12$ mm
- **ECOFF**: 0.25 mg/L

**Legend**

- 2
- 1
- 0.5
- 0.25
- 0.125
- 0.06
- 0.03

**Inhibition zone diameter (mm)**

- **Breakpoints**
  - Cefepime MIC: $S \leq 0.25$, $R > 0.25$ mg/L
  - Benzylpenicillin zone diameter (screen): $S \geq 12$ mm
  - ECOFF: 0.25 mg/L
Benzylpenicillin 1 unit vs. Cefixime MIC
*H. influenzae*, 147 isolates

(1 data source)

### Breakpoints

<table>
<thead>
<tr>
<th>Cefixime MIC</th>
<th>S≤0.125, R&gt;0.125 mg/L</th>
</tr>
</thead>
<tbody>
<tr>
<td>Benzylpenicillin zone diameter (screen)</td>
<td>S≥12 mm</td>
</tr>
</tbody>
</table>

### ECOFF

0.125 mg/L
Benzylpenicillin 1 unit vs. Cefotaxime MIC
*H. influenzae*, 161 isolates (193 correlates)

(3 data sources)

No of observations

Inhibition zone diameter (mm)

<table>
<thead>
<tr>
<th>MIC (mg/L)</th>
<th>Breakpoints</th>
</tr>
</thead>
</table>
| 1          | Cefotaxime MIC  
S≤0.125, R>0.125 mg/L  
ECOFF 0.06 mg/L |
| 0.5        | Benzylpenicillin zone diameter (screen)  
S≥12 mm |
| 0.25       |
| 0.125      |
| 0.06       |
| 0.03       |
| 0.016      |
| 0.008      |
| ≤0.004     |
Benzylpenicillin 1 unit vs. Cefpodoxime MIC

*H. influenzae*, 146 isolates

(1 data source)

**Breakpoints**

<table>
<thead>
<tr>
<th>Breakpoints</th>
<th>ECOFF</th>
</tr>
</thead>
<tbody>
<tr>
<td>Cefpodoxime MIC</td>
<td>S≤0.25, R&gt;0.25 mg/L</td>
</tr>
<tr>
<td>Benzylpenicillin zone diameter (screen)</td>
<td>S≥12 mm</td>
</tr>
</tbody>
</table>
Benzylpenicillin 1 unit vs. Ceftaroline MIC

*H. influenzae*, 105 isolates

(1 data source)

**Breakpoints**

- **Ceftaroline MIC**
  - S ≤ 0.03, R > 0.03 mg/L
  - ECOFF: 0.03 mg/L

- **Benzylpenicillin zone diameter (screen)**
  - S ≥ 12 mm
Benzylpenicillin 1 unit vs. Ceftibuten MIC

*H. influenzae*, 147 isolates

(1 data source)

**Breakpoints**

<table>
<thead>
<tr>
<th>Ceftibuten MIC</th>
<th>Benzylpenicillin zone diameter (screen)</th>
<th>ECOFF</th>
</tr>
</thead>
<tbody>
<tr>
<td>S≤1, R&gt;1 mg/L</td>
<td>S≥12 mm</td>
<td>0.5 mg/L</td>
</tr>
</tbody>
</table>

**MIC (mg/L)**

- ≥16
- 8
- 4
- 2
- 1
- 0.5
- 0.25
- 0.125
- 0.06
- 0.03
- 0.016
**Benzylpenicillin 1 unit vs. Ceftriaxone MIC**

*H. influenzae*, 174 isolates

(2 data sources)

### Breakpoints

<table>
<thead>
<tr>
<th>Breakpoints</th>
<th>MIC (mg/L)</th>
<th>ECOFF (mg/L)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Ceftriaxone MIC</td>
<td>S ≤ 0.125, R &gt; 0.125</td>
<td>0.06</td>
</tr>
<tr>
<td>Benzylpenicillin zone diameter (screen)</td>
<td>S ≥ 12 mm</td>
<td></td>
</tr>
</tbody>
</table>
Benzylpenicillin 1 unit vs. Cefuroxime MIC

H. influenzae, 188 isolates

(2 data sources)

Breakpoints (iv)

<table>
<thead>
<tr>
<th>Cefuroxime MIC</th>
<th>Benzylpenicillin zone diameter (screen)</th>
<th>ECOOFF</th>
</tr>
</thead>
<tbody>
<tr>
<td>S≤1, R&gt;2 mg/L</td>
<td>S≥12 mm</td>
<td>2 mg/L</td>
</tr>
</tbody>
</table>
Benzylpenicillin 1 unit vs. Cefuroxime MIC

_H. influenzae_, 188 isolates

(2 data sources)

**Breakpoints (oral)**

<table>
<thead>
<tr>
<th>Cefuroxime MIC</th>
<th>Benzylpenicillin zone diameter (screen)</th>
<th>ECOFF</th>
</tr>
</thead>
<tbody>
<tr>
<td>S≤0.001, R&gt;1 mg/L</td>
<td>S≥12 mm</td>
<td>2 mg/L</td>
</tr>
</tbody>
</table>
Benzylpenicillin 1 unit vs. Ertapenem MIC

*H. influenzae*, 124 isolates

(1 data source)

**Breakpoints**

<table>
<thead>
<tr>
<th>Breakpoint Description</th>
<th>MIC (mg/L)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Ertapenem MIC</td>
<td>≤0.5, R&gt;0.5 mg/L</td>
</tr>
<tr>
<td>Benzylpenicillin zone diameter (screen)</td>
<td>≥12 mm</td>
</tr>
</tbody>
</table>
Benzylpenicillin 1 unit vs. Imipenem MIC

*H. influenzae*, 116 isolates

(2 data sources)

<table>
<thead>
<tr>
<th>MIC (mg/L)</th>
<th>No of observations</th>
</tr>
</thead>
<tbody>
<tr>
<td>8</td>
<td>6</td>
</tr>
<tr>
<td>4</td>
<td>6</td>
</tr>
<tr>
<td>2</td>
<td>2</td>
</tr>
<tr>
<td>1</td>
<td>2</td>
</tr>
<tr>
<td>0.5</td>
<td>2</td>
</tr>
<tr>
<td>0.25</td>
<td>2</td>
</tr>
<tr>
<td>0.125</td>
<td>2</td>
</tr>
<tr>
<td>≤0.06</td>
<td>2</td>
</tr>
</tbody>
</table>

**Breakpoints**

<table>
<thead>
<tr>
<th></th>
<th>ECOFF</th>
</tr>
</thead>
<tbody>
<tr>
<td>Imipenem MIC</td>
<td>S≤2, R&gt;2 mg/L</td>
</tr>
<tr>
<td>Benzylpenicillin zone diameter (screen)</td>
<td>S≥12 mm</td>
</tr>
<tr>
<td></td>
<td>2 mg/L</td>
</tr>
</tbody>
</table>

**Inhibition zone diameter (mm)**

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**ECOFF**

- Benzylpenicillin: S≥12 mm
- Imipenem: S≤2, R>2 mg/L

**Breakpoints**

- Imipenem MIC
- Benzylpenicillin zone diameter (screen)
Benzylpenicillin 1 unit vs. Meropenem MIC
*H. influenzae*, 200 isolates (298 correlates)

(4 data sources)

**Breakpoints (non-meningitis)**

<table>
<thead>
<tr>
<th>Meropenem MIC</th>
<th>Benzylpenicillin zone diameter (screen)</th>
</tr>
</thead>
<tbody>
<tr>
<td>S≤2, R&gt;2 mg/L</td>
<td>S≥12 mm</td>
</tr>
</tbody>
</table>

**ECOFF**

0.25 mg/L

**MIC (mg/L)**

- 0.5
- 0.25
- 0.125
- 0.06
- 0.03
- ≤0.016
Benzylpenicillin 1 unit vs. Meropenem MIC
*H. influenzae*, 200 isolates (298 correlates)

(4 data sources)

For benzylpenicillin screen positive isolates (zone <12 mm), determine the MIC for meropenem in meningitis.

### Breakpoints (meningitis)
- **Meropenem MIC**
  - S\(\leq0.25\), R>0.25 mg/L
  - ECOFF 0.25 mg/L
- **Benzylpenicillin zone diameter (screen)**
  - S\(\geq12\) mm