European Subcommitte on Anti-fungal Susceptibility Testing (EUCAST AFST)

Routine and extended internal quality control for antifungal susceptibility as recommended by EUCAST

Version 2.0, valid from 22nd of June, 2018

This document should be cited as

<table>
<thead>
<tr>
<th>General</th>
<th>Page</th>
</tr>
</thead>
<tbody>
<tr>
<td>Notes</td>
<td>2</td>
</tr>
</tbody>
</table>

<table>
<thead>
<tr>
<th>Routine quality control</th>
<th>Page</th>
</tr>
</thead>
<tbody>
<tr>
<td>Candida krusei ATCC 6258</td>
<td>3</td>
</tr>
<tr>
<td>Candida parapsilosis ATCC 22019</td>
<td>4</td>
</tr>
<tr>
<td>Candida albicans CNM-CL F8555</td>
<td>5</td>
</tr>
<tr>
<td>Candida krusei CNM-CL-3403</td>
<td>6</td>
</tr>
<tr>
<td>Aspergillus fumigatus ATCC 204305</td>
<td>7</td>
</tr>
<tr>
<td>Aspergillus flavus ATCC 204304</td>
<td>8</td>
</tr>
<tr>
<td>Aspergillus flavus CNM-CM-1813</td>
<td>9</td>
</tr>
<tr>
<td>A. fumigatus SSI-4524</td>
<td>10</td>
</tr>
<tr>
<td>A. fumigatus SSI-5586</td>
<td>11</td>
</tr>
</tbody>
</table>
Notes

1. In EUCAST quality control (QC) tables, both ranges and targets are listed. Repeat testing of EUCAST quality control strains should yield individual MIC values randomly distributed within the recommended ranges. If the number of tests is ≥10, the mode MIC should be the target value.

2. EUCAST quality control strains for routine QC are used to monitor test performance. Control tests should be set up and checked daily, at least for antifungal agents which are part of routine panels.

3. For information about the reference methods for susceptibility testing of yeast and moulds see Methods in antifungal susceptibility testing of yeasts and Methods in antifungal susceptibility testing of moulds, respectively.
**Candida krusei ATCC 6258**

<table>
<thead>
<tr>
<th>Antifungal Agent</th>
<th>MIC (mg/L) (^b)</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>Target</td>
</tr>
<tr>
<td>Amphotericin B</td>
<td>0.25-0.5</td>
</tr>
<tr>
<td>Anidulafungin</td>
<td>0.03</td>
</tr>
<tr>
<td>Caspofungin</td>
<td>NA(^c)</td>
</tr>
<tr>
<td>Fluconazole</td>
<td>32.0</td>
</tr>
<tr>
<td>Flucytosine</td>
<td>2.0</td>
</tr>
<tr>
<td>Isavuconazole</td>
<td>0.03</td>
</tr>
<tr>
<td>Itraconazole</td>
<td>0.06</td>
</tr>
<tr>
<td>Micafungin</td>
<td>0.06</td>
</tr>
<tr>
<td>Voriconazole</td>
<td>0.06-0.125</td>
</tr>
<tr>
<td>Posaconazole</td>
<td>0.03</td>
</tr>
</tbody>
</table>

\(^a\) ATCC: American Type Culture Collection.  
\(^b\) *Candida* QC strains requires reading in a spectrophotometer after one day of incubation.  
\(^c\) Not available
**Candida parapsilosis ATCC 22019**

<table>
<thead>
<tr>
<th>Antifungal Agent</th>
<th>MIC (mg/L)</th>
<th>Target</th>
<th>Range</th>
</tr>
</thead>
<tbody>
<tr>
<td>Amphotericin B</td>
<td>0.25-0.5</td>
<td>0.125-1.0</td>
<td></td>
</tr>
<tr>
<td>Anidulafungin</td>
<td>0.5</td>
<td>0.25-1.0</td>
<td></td>
</tr>
<tr>
<td>Caspofungin</td>
<td>NA c</td>
<td>NA c</td>
<td></td>
</tr>
<tr>
<td>Fluconazole</td>
<td>1.0</td>
<td>0.5-2.0</td>
<td></td>
</tr>
<tr>
<td>Flucytosine</td>
<td>0.25</td>
<td>0.125-0.5</td>
<td></td>
</tr>
<tr>
<td>Isavuconazole</td>
<td>0.015</td>
<td>0.008-0.03</td>
<td></td>
</tr>
<tr>
<td>Itraconazole</td>
<td>0.06</td>
<td>0.03-0.125</td>
<td></td>
</tr>
<tr>
<td>Micafungin</td>
<td>1.0</td>
<td>0.5-2.0</td>
<td></td>
</tr>
<tr>
<td>Voriconazole</td>
<td>0.03</td>
<td>0.015-0.06</td>
<td></td>
</tr>
<tr>
<td>Posaconazole</td>
<td>0.03</td>
<td>0.015-0.06</td>
<td></td>
</tr>
</tbody>
</table>

*a* ATCC: American Type Culture Collection.

*b* *Candida* QC strains requires reading in a spectrophotometer after one day of incubation.

*c* Not available
**Candida albicans CNM-CL F8555**

<table>
<thead>
<tr>
<th>Antifungal Agent</th>
<th>MIC (mg/L)</th>
<th>Target</th>
<th>Range</th>
</tr>
</thead>
<tbody>
<tr>
<td>Amphotericin B</td>
<td>0.125-0.25</td>
<td>0.06-0.5</td>
<td></td>
</tr>
<tr>
<td>Anidulafungin</td>
<td>NA</td>
<td>NA</td>
<td></td>
</tr>
<tr>
<td>Caspofungin</td>
<td>NA</td>
<td>NA</td>
<td></td>
</tr>
<tr>
<td>Fluconazole</td>
<td>64.0</td>
<td>32.0-128.0</td>
<td></td>
</tr>
<tr>
<td>Flucytosine</td>
<td>0.125</td>
<td>0.06-0.25</td>
<td></td>
</tr>
<tr>
<td>Isavuconazole</td>
<td>NA</td>
<td>NA</td>
<td></td>
</tr>
<tr>
<td>Itraconazole</td>
<td>0.5</td>
<td>0.25-1.0</td>
<td></td>
</tr>
<tr>
<td>Micafungin</td>
<td>NA</td>
<td>NA</td>
<td></td>
</tr>
<tr>
<td>Voriconazole</td>
<td>1.0</td>
<td>0.5-2.0</td>
<td></td>
</tr>
<tr>
<td>Posaconazole</td>
<td>0.25</td>
<td>0.125-0.5</td>
<td></td>
</tr>
</tbody>
</table>

*a* CNM-CL: Yeast collection of the Spanish National Centre of Microbiology. Available from the EUCAST Development Laboratory (EDL) for fungi: [http://www.eucast.org/organization/developmentlaboratories/](http://www.eucast.org/organization/developmentlaboratories/)

*b* *Candida* QC strains requires reading in a spectrophotometer after one day of incubation.

*c* Not available
**Candida krusei CNM-CL-3403**

<table>
<thead>
<tr>
<th>Antifungal Agent</th>
<th>MIC (mg/L)</th>
<th>Target</th>
<th>Range</th>
</tr>
</thead>
<tbody>
<tr>
<td>Amphotericin B</td>
<td>0.5</td>
<td>0.25-1.0</td>
<td></td>
</tr>
<tr>
<td>Anidulafungin</td>
<td>NA&lt;sup&gt;c&lt;/sup&gt;</td>
<td>NA&lt;sup&gt;c&lt;/sup&gt;</td>
<td></td>
</tr>
<tr>
<td>Caspofungin</td>
<td>NA&lt;sup&gt;c&lt;/sup&gt;</td>
<td>NA&lt;sup&gt;c&lt;/sup&gt;</td>
<td></td>
</tr>
<tr>
<td>Fluconazole</td>
<td>32.0</td>
<td>16.0-64.0</td>
<td></td>
</tr>
<tr>
<td>Flucytosine</td>
<td>4.0</td>
<td>2.0-8.0</td>
<td></td>
</tr>
<tr>
<td>Isavuconazole</td>
<td>NA&lt;sup&gt;b&lt;/sup&gt;</td>
<td>NA&lt;sup&gt;c&lt;/sup&gt;</td>
<td></td>
</tr>
<tr>
<td>Itraconazole</td>
<td>0.25</td>
<td>0.125-0.5</td>
<td></td>
</tr>
<tr>
<td>Micafungin</td>
<td>NA&lt;sup&gt;c&lt;/sup&gt;</td>
<td>NA&lt;sup&gt;c&lt;/sup&gt;</td>
<td></td>
</tr>
<tr>
<td>Voriconazole</td>
<td>0.25</td>
<td>0.125-0.5</td>
<td></td>
</tr>
<tr>
<td>Posaconazole</td>
<td>0.125</td>
<td>0.06-0.25</td>
<td></td>
</tr>
</tbody>
</table>

<sup>a</sup> CNM-CL: Yeast collection of the Spanish National Centre of Microbiology. Available from the EUCAST Development Laboratory (EDL) for fungi: [http://www.eucast.org/organization/developmentlaboratories/](http://www.eucast.org/organization/developmentlaboratories/)

<sup>b</sup> Candida QC strains requires reading in a spectrophotometer after one day of incubation.

<sup>c</sup> Not available
Aspergillus fumigatus ATCC 204305

<table>
<thead>
<tr>
<th>Antifungal Agent</th>
<th>MIC (mg/L)</th>
<th>Target</th>
<th>Range</th>
</tr>
</thead>
<tbody>
<tr>
<td>Amphotericin B</td>
<td>0.5</td>
<td>0.25-1.0</td>
<td></td>
</tr>
<tr>
<td>Caspofungin</td>
<td>NA c</td>
<td>NA c</td>
<td></td>
</tr>
<tr>
<td>Isavuconazole</td>
<td>NA c</td>
<td>NA c</td>
<td></td>
</tr>
<tr>
<td>Itraconazole</td>
<td>0.25</td>
<td>0.125-0.5</td>
<td></td>
</tr>
<tr>
<td>Micafungin</td>
<td>NA c</td>
<td>NA c</td>
<td></td>
</tr>
<tr>
<td>Voriconazole</td>
<td>0.5</td>
<td>0.25-1.0</td>
<td></td>
</tr>
<tr>
<td>Posaconazole</td>
<td>0.06-0.125</td>
<td>0.03-0.25</td>
<td></td>
</tr>
</tbody>
</table>

a ATCC: American Type Culture Collection.
b Aspergillus QC strains should be read visually at a no growth inhibition endpoint after 2 days of incubation.
c Not available
**Aspergillus flavus ATCC 204304**

<table>
<thead>
<tr>
<th>Antifungal Agent</th>
<th>MIC (mg/L)</th>
<th>Target</th>
<th>Range</th>
</tr>
</thead>
<tbody>
<tr>
<td>Amphotericin B</td>
<td>1.0</td>
<td>0.5-2.0</td>
<td></td>
</tr>
<tr>
<td>Caspofungin</td>
<td>NA&lt;sup&gt;c&lt;/sup&gt;</td>
<td>NA&lt;sup&gt;c&lt;/sup&gt;</td>
<td></td>
</tr>
<tr>
<td>Isavuconazole</td>
<td>NA&lt;sup&gt;c&lt;/sup&gt;</td>
<td>NA&lt;sup&gt;c&lt;/sup&gt;</td>
<td></td>
</tr>
<tr>
<td>Itraconazole</td>
<td>0.25</td>
<td>0.125-0.5</td>
<td></td>
</tr>
<tr>
<td>Micafungin</td>
<td>NA&lt;sup&gt;c&lt;/sup&gt;</td>
<td>NA&lt;sup&gt;c&lt;/sup&gt;</td>
<td></td>
</tr>
<tr>
<td>Voriconazole</td>
<td>1.0</td>
<td>0.5-2.0</td>
<td></td>
</tr>
<tr>
<td>Posaconazole</td>
<td>0.25</td>
<td>0.125-0.5</td>
<td></td>
</tr>
</tbody>
</table>

<sup>a</sup> ATCC: American Type Culture Collection.

<sup>b</sup> *Aspergillus* QC strains should be read visually at a no growth inhibition endpoint after 2 days of incubation.

<sup>c</sup> Not available
**Aspergillus flavus CNM-CM-1813**

<table>
<thead>
<tr>
<th>Antifungal Agent</th>
<th>MIC (mg/L)</th>
<th>Target</th>
<th>Range</th>
</tr>
</thead>
<tbody>
<tr>
<td>Amphotericin B</td>
<td>2.0</td>
<td>1.0-4.0</td>
<td></td>
</tr>
<tr>
<td>Caspofungin</td>
<td>NA c</td>
<td>NA c</td>
<td></td>
</tr>
<tr>
<td>Isavuconazole</td>
<td>NA c</td>
<td>NA c</td>
<td></td>
</tr>
<tr>
<td>Itraconazole</td>
<td>0.25</td>
<td>0.125-0.5</td>
<td></td>
</tr>
<tr>
<td>Micafungin</td>
<td>NA c</td>
<td>NA c</td>
<td></td>
</tr>
<tr>
<td>Voriconazole</td>
<td>1.0</td>
<td>0.5-2.0</td>
<td></td>
</tr>
<tr>
<td>Posaconazole</td>
<td>0.25</td>
<td>0.125-0.5</td>
<td></td>
</tr>
</tbody>
</table>

*a* CNM-CM: Spanish National Centre for Microbiology, Filamentous Fungi Culture Collection. Available from the EUCAST Development Laboratory (EDL) for fungi: [http://www.eucast.org/organization/developmentlaboratories/](http://www.eucast.org/organization/developmentlaboratories/)

*b* Aspergillus QC strains should be read visually at a no growth inhibition endpoint after 2 days of incubation.

*c* Not available
**A. fumigatus SSI-4524**

QC strain harbouring TR34/L98H substitution for quality control of azole-containing agar plates.

Each well of the plate contains the following azoles: well 1 (itraconazole 4 mg/L), well 2 (voriconazole 2 mg/L), well 3 (posaconazole 0.5 mg/L), and well 4 (drug free control well).

Expected growth scores after 48 h.

<table>
<thead>
<tr>
<th>Score</th>
<th>Description</th>
</tr>
</thead>
<tbody>
<tr>
<td>0</td>
<td>no visible growth</td>
</tr>
<tr>
<td>0.5</td>
<td>1-5 tiny colonies</td>
</tr>
<tr>
<td>1</td>
<td>weak/minimal growth</td>
</tr>
<tr>
<td>2</td>
<td>clearly visible growth</td>
</tr>
<tr>
<td>3</td>
<td>un-inhibited prominent growth</td>
</tr>
</tbody>
</table>

Score definition:
0: no visible growth; 0.5: 1-5 tiny colonies; 1: weak/minimal growth (either as >5 tiny colonies or confluent weak growth where the agar has been inoculated (covering ≤ half the well)); 2: clearly visible growth with hyphal extension but not covering the entire well (and less than the control well); 3: un-inhibited prominent growth covering the majority of the well and similar to that in the control well.

SSI: Statens Serum Institut, Copenhagen, Denmark
Available from the EUCAST Development Laboratory (EDL) for fungi:
http://www.eucast.org/organization/developmentlaboratories/
A. fumigatus SSI-5586

QC strain harbouring G54W substitution for quality control of azole-containing agar plates.

Each well of the plate contains the following azoles: well 1 (itraconazole 4 mg/L), well 2 (voriconazole 2 mg/L), well 3 (posaconazole 0.5 mg/L), and well 4 (drug free control well).

Expected growth pattern after 48 h:

<table>
<thead>
<tr>
<th>Score</th>
<th>Description</th>
</tr>
</thead>
<tbody>
<tr>
<td>2 - 3</td>
<td>0: no visible growth; 0.5: 1-5 tiny colonies; 1: weak/minimal growth (either as &gt;5 tiny colonies or confluent weak growth where the agar has been inoculated (covering ≤ half the well)); 2: clearly visible growth with hyphal extension but not covering the entire well (and less than the control well); 3: uninhibited prominent growth covering the majority of the well and similar to that in the control well.</td>
</tr>
</tbody>
</table>

SSI: Statens Serum Institut, Copenhagen, Denmark
Available from the EUCAST Development Laboratory (EDL) for fungi: [http://www.eucast.org/organization/developmentlaboratories/](http://www.eucast.org/organization/developmentlaboratories/)