Benomyl Assay

Benomyl Haploid/Diploid Assay

Pin the cells you wish to test onto YPD + Benomyl Media and YPD + DMSO Media. Remember to include FY4 and FY4/5 as controls.

Incubate at 30 C.

YPD + Benomyl Media – 1 L

20 g Peptone
10 g Yeast Extract
20 g Agar

Bring to 948 mL with H2O.
Add magnetic stir bar.
Autoclave
Add 50 mL 40% Glucose. Mix
Cool media to 65 C

With sterile glass pipette and bulb slowly add 2 mL 10 mg/mL Benomyl preheated to 65 C drop by drop to the rest of the media while it is mixing on stirplate. Be careful; if the Benomyl is added too fast or at too cool of a temperature it with crash out of the media.
Pour into plates.

YPD + DMSO Media – 1L

20 g Peptone
10 g Yeast Extract
20 g Agar

Bring to 948 mL with H2O.
Autoclave
Add 50 mL 40% Glucose. Mix
Cool media to 65 C
Add 2 mL DMSO. Mix
Pour into plates.

Examples

Plate Layout

<p>| | | | | | | | | | | | |</p>
<table>
<thead>
<tr>
<th></th>
<th></th>
<th></th>
<th></th>
<th></th>
<th></th>
<th></th>
<th></th>
<th></th>
<th></th>
<th></th>
<th></th>
</tr>
</thead>
<tbody>
<tr>
<td>A</td>
<td>FY4</td>
<td>FY4/5</td>
<td>FY4</td>
<td>FY4/5</td>
<td>FY4</td>
<td>FY4</td>
<td>FY4</td>
<td>FY4</td>
<td>FY4</td>
<td>FY4</td>
<td>FY4/5</td>
</tr>
<tr>
<td>B</td>
<td>FY4/5</td>
<td>FY4</td>
<td>FY4/5</td>
<td>FY4</td>
<td>FY4</td>
<td>FY4</td>
<td>FY4</td>
<td>FY4</td>
<td>FY4</td>
<td>FY4</td>
<td>FY4/5</td>
</tr>
<tr>
<td>C</td>
<td>FY4</td>
<td>FY4/5</td>
<td>FY4</td>
<td>FY4/5</td>
<td>FY4</td>
<td>FY4</td>
<td>FY4</td>
<td>FY4</td>
<td>FY4</td>
<td>FY4</td>
<td>FY4/5</td>
</tr>
<tr>
<td>D</td>
<td>FY4/5</td>
<td>FY4</td>
<td>FY4/5</td>
<td>FY4</td>
<td>FY4</td>
<td>FY4</td>
<td>FY4</td>
<td>FY4</td>
<td>FY4</td>
<td>FY4</td>
<td>FY4/5</td>
</tr>
<tr>
<td>E</td>
<td>FY4</td>
<td>FY4/5</td>
<td>FY4</td>
<td>FY4/5</td>
<td>FY4</td>
<td>FY4</td>
<td>FY4</td>
<td>FY4</td>
<td>FY4</td>
<td>FY4</td>
<td>FY4/5</td>
</tr>
<tr>
<td>F</td>
<td>FY4/5</td>
<td>FY4</td>
<td>FY4/5</td>
<td>FY4</td>
<td>FY4</td>
<td>FY4</td>
<td>FY4</td>
<td>FY4</td>
<td>FY4</td>
<td>FY4</td>
<td>FY4/5</td>
</tr>
</tbody>
</table>
FY4 and FY4/5 grown in YPD on YPD + DMSO

FY4 and FY4/5 grown in YPD on YPD + Benomyl

FY4 and FY4/5 grown in Gln-Lim on YPD + DMSO

FY4 and FY4/5 grown in YPD + Benomyl