FEVER MICRO TEST
BRUCELLA MELITENSI S

Determination of antibodies associated with Brucella Melitensis infections by coloured bacterial suspension in microplate

TEST SUMMARY
The antibodies associated with Brucella Melitensis infections cause agglutination of inactive bacteria present in suspension. The intravalit colourings permits an easier reading of agglutination formation.

SAMPLES
Fresh clear serum. Stability 7 days at 2-8°C. Freeze for longer period at -20°C, and keep at room temperature before the analysis. Do not freeze repeatedly. Turbid samples have to be centrifuged.

REAGENTS
Suspension: Coloured intravalit inactive bacterial suspension; conservative and stabilizer.

Brucella positive control: Solution of rabbit antisera that gives a clear agglutination with Brucella Suspension; conservative and stabilizer.

Negative control: Proteic bovine solution that doesn’t react with suspension; conservative and stabilizer.

RESULTS INTERPRETATION
A coloured bottom with a clear point shape, on the well bottom, indicates negativity. An agglutinate that cover all the well bottom indicates a clear positivity, while, a no uniform agglutinate with a bottom in the centre, on the well bottom, indicates a feeble positivity.

The serum titre is given by a high dilution in which there is a feeble positivity.

DIAGNOSTIC VALUES
Titers greater 1/80 indicates recent infection.

It is a distinctive sign for the infection diagnosis the significant increase of titre between examined samples after some days.

NOTE
• If the results are incompatible with clinical presentation, they have to be evaluated within a total clinical study.

CALIBRATION/QUALITY CONTROL
Positive and Negative control sera should be always used to distinguish an eventual background’s agglutination of reactive.

PROCEDURE
In a microplate with "U" wells dilute the serum with physiologic solution (100 µl of serum with 900 µl of physiologic). Discharge 100 µl from last well (well n° 9).

<table>
<thead>
<tr>
<th>Well</th>
<th>1</th>
<th>2</th>
<th>3</th>
<th>4</th>
<th>5</th>
<th>6</th>
<th>7</th>
<th>8</th>
<th>9</th>
<th>Suspension Brucella Melitensis 3 x 10 ml</th>
</tr>
</thead>
<tbody>
<tr>
<td>Physiological serum</td>
<td>100 µl</td>
<td>100 µl</td>
<td>100 µl</td>
<td>100 µl</td>
<td>100 µl</td>
<td>100 µl</td>
<td>100 µl</td>
<td>100 µl</td>
<td>100 µl</td>
<td>Brucella Positive control 1 x 0.5 ml</td>
</tr>
<tr>
<td>Diluted serum</td>
<td>100 µl</td>
<td>100 µl</td>
<td>100 µl</td>
<td>100 µl</td>
<td>100 µl</td>
<td>100 µl</td>
<td>100 µl</td>
<td>100 µl</td>
<td>100 µl</td>
<td>Negative control 1 x 0.5 ml</td>
</tr>
</tbody>
</table>

Drain the plate by slow rotations for 20-30 sec. Incubate at 37°C for 16-18 h or at 22°C for 2 days, to improve bottoms formation. It is advisable put the plate in the fridge after the incubation for 2 hours.

<table>
<thead>
<tr>
<th>Comptitors</th>
<th>MASCIA BRUNELLI</th>
<th>TOT.</th>
</tr>
</thead>
<tbody>
<tr>
<td>+</td>
<td>17</td>
<td>0</td>
</tr>
<tr>
<td>+</td>
<td>0</td>
<td>33</td>
</tr>
<tr>
<td><strong>TOT.</strong></td>
<td>17</td>
<td>33</td>
</tr>
</tbody>
</table>

Specificity
A comparison with an available commercial method gave following results on 50 samples compared, giving a specificity = 100%.

REFERENCES
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