CHROMagar™ VRE

**MEDIUM PURPOSE**

Chromogenic medium for detection of Van A/Van B VR E. faecalis & VR E. faecium.

There are two types of vancomycin resistance in enterococci. The first type is intrinsic resistance (mostly Van C type but also Van D, Van E, Van F, etc) found in *E. gallinarum* and *E. casseliflavus/E. flavescens* and demonstrates a low-level resistance to vancomycin. The second type of vancomycin resistance in enterococci is acquired resistance (Van A & Van B types), mostly seen in *E. faecium* and *E. faecalis*. Therefore, to avoid the spread of this resistance to more virulent pathogens (*S. aureus*, for instance) it is crucial to promptly detect the presence of any of these two species in the patient, and accurately differentiate them from other Enterococci.

**COMPOSITION**

The product is composed of a powder base (B) and 1 supplement (S).

<table>
<thead>
<tr>
<th>Product</th>
<th>Base (B)</th>
<th>Supplement (S)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Total g/L</td>
<td>67.3 g/L</td>
<td>0.06 g/L</td>
</tr>
<tr>
<td>Composition g/L</td>
<td>Agar 15.0</td>
<td>Selective mix 0.06</td>
</tr>
<tr>
<td>Peptones and yeast extract 20.0</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Salts 5.0</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Chromogenic mix 27.3</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Aspect</td>
<td>Powder Form</td>
<td>Powder Form</td>
</tr>
<tr>
<td>STORAGE</td>
<td>15-30 °C</td>
<td>2-8 °C</td>
</tr>
</tbody>
</table>

**FINAL MEDIA pH**

6.9 +/- 0.5

**PREPARATION (Calculation for 1 L)**

**Step 1**

Preparation of the base CHROMagar™ VRE Base (B)

- Disperse slowly 67.3 g of powder base in 1 L of purified water.
- Stir until agar is well thickened.
- Autoclave at 110 °C during 5 min.
  
**DO NOT AUTOCLAVE AT 121 °C. DO NOT HEAT LONGER THAN 5 MIN.**
  
- Cool in a water bath to 45-50 °C. Swirl or stir gently to homogenize.

**Step 2**

Preparation of the Supplement (S)

- Prepare a stock solution of the CHROMagar™ VRE supplement:
  
  Add 60 mg to 1 mL (0.5 mL ethanol/0.5 mL sterile purified water).
  
  Swirl well until complete dissolution.

**Advice 2:** This CHROMagar™ VRE supplement stock solution should be used immediately after preparation, or can be stored at -20 °C and used within one month.

**Step 3**

Mixing of the prepared base (B) and the prepared supplement (S)

- Add 1 mL of CHROMagar™ VRE supplement solution to CHROMagar™ VRE base cooled at 45-50 °C (final 60 mg/L).
  
  Swirl gently to homogenize.
  
  Pour into sterile Petri dishes.
  
  Let it solidify and dry.

**Storage**

- Store in the dark before use.
  
- Prepared media plates can be kept for one day at room temperature.
  
- Plates can be stored for up to one month under refrigeration (2/8 °C) if properly prepared and protected from light and dehydration.

**REFERENCES**

<table>
<thead>
<tr>
<th>Pack Size</th>
<th>Ordering References</th>
<th>Base</th>
<th>Supplement</th>
</tr>
</thead>
<tbody>
<tr>
<td>5000 mL</td>
<td>VR952</td>
<td>VR952(B)</td>
<td>Weight: 336.5 g</td>
</tr>
<tr>
<td>10 kg</td>
<td>VR953-10kg</td>
<td>VR953-10 kg</td>
<td>Weight: 10 kg</td>
</tr>
</tbody>
</table>

**COMPUTED AND PREPARED MEDIA**

<table>
<thead>
<tr>
<th>1 L</th>
<th>5 L</th>
</tr>
</thead>
<tbody>
<tr>
<td>60 mg into 0.5 mL of ethanol + 0.5 mL sterile purified water</td>
<td>300 mg into 2.5 mL of ethanol + 2.5 mL sterile purified water</td>
</tr>
</tbody>
</table>

**HELPING CALCULATION**

- Final Media
- Add 1 mL of supplement to the prepared base
- Add 5 mL of supplement to the prepared base
**SPECIMEN COLLECTION AND HANDLING**

CHROMagar™ VRE can be used with the following specimens: stools and rectal swabs.

This medium can be also used in environmental field with the following specimens: clinical material.

Use of transport devices approved for collection of such specimens is recommended.

**MATERIAL REQUIRED BUT NOT PROVIDED**

Standard microbiological laboratory material for culture media preparation, control, streaking, incubation and waste disposal.

**INOCULATION**

Related samples can be processed by direct streaking on the plate, as well as prior appropriate enrichment step.

- If the agar plate has been refrigerated, allow to warm to room temperature before inoculation.
- Streak sample onto plate
- Incubate in aerobic conditions at 35-37 °C for 24 hours.

**INTERPRETATION**

<table>
<thead>
<tr>
<th>Microorganism</th>
<th>Typical colony appearance</th>
</tr>
</thead>
<tbody>
<tr>
<td><em>VRE. faecium</em></td>
<td>pink to mauve</td>
</tr>
<tr>
<td><em>VRE. faecalis</em></td>
<td></td>
</tr>
<tr>
<td><em>VRE. gallinarum</em></td>
<td>blue or inhibited</td>
</tr>
<tr>
<td><em>VRE. casseliflavus</em></td>
<td></td>
</tr>
<tr>
<td>Other Gram (+) bacteria</td>
<td>inhibited</td>
</tr>
<tr>
<td>Gram (-) bacteria</td>
<td>inhibited</td>
</tr>
<tr>
<td>Yeasts and mould</td>
<td>mostly inhibited</td>
</tr>
</tbody>
</table>

**PERFORMANCE**

In the following study, 95 rectal swabs were tested and read after 24 h incubation at 35-37 °C in aerobiosis.

<table>
<thead>
<tr>
<th>CHROMagar™ VRE</th>
<th>Reference Method (VRE Select)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Sensitivity</td>
<td>95.5 % *</td>
</tr>
<tr>
<td>Specificity</td>
<td>90.4 % *</td>
</tr>
</tbody>
</table>

* Data obtained from the study «Evaluation of Three Chromogenic Media for Detection of Vancomycin-Resistant Enterococci in a tertiary-care Hospital» M.L. Miller et Al. CACMID 2011

**LIMITATIONS AND COMPLEMENTARY TESTS**

- Vancomycin R-type should be determined and confirmed by further testing.
- Some rare strains of *Lactobacilli, Pediococcus* can sometimes appear as pinpoint mauve colonies. However, they can be differentiated with PYR test: PYR(+) --> VRE; PYR(-) --> *Lactobacilli, Pediococcus*.
- Further to 24 h incubation, some rare strains of *E. gallinarum* can sometimes appear as mauve colonies.

**QUALITY CONTROL**

Please perform Quality Control according to the use of the medium and the local QC regulations and norms.

Good preparation of the medium can be tested, isolating the following ATCC strains:

<table>
<thead>
<tr>
<th>Microorganism</th>
<th>Typical colony appearance</th>
</tr>
</thead>
<tbody>
<tr>
<td><em>VRE. faecalis</em> ATCC® 51575</td>
<td>mauve, small</td>
</tr>
<tr>
<td><em>E. faecalis</em> ATCC® 29212</td>
<td>inhibited</td>
</tr>
<tr>
<td><em>E. casseliflavus</em> ATCC® 700327</td>
<td>inhibited</td>
</tr>
<tr>
<td><em>E. gallinarum</em> ATCC® 49573</td>
<td>inhibited</td>
</tr>
<tr>
<td><em>E. coli</em> ATCC® 25922</td>
<td>inhibited</td>
</tr>
<tr>
<td><em>S. aureus</em> ATCC® 25923</td>
<td>inhibited</td>
</tr>
</tbody>
</table>

**WARNINGS AND PRECAUTIONS**

- For Research Use Only (RUO). Not for use in diagnostic procedures.
- This laboratory product should be used only by trained personnel (healthcare professional, etc). Wear appropriate protective clothing, gloves and eye/face protection and handle appropriately with procedures and good laboratory practices.
- Use of the medium may be difficult for people who have problems recognising colours.
- For a good microbial detection, collection and transport of specimen should be well handled and adapted to the particular specimen according to good laboratory practices.
- Culture media should not be used as manufacturing material or components.
- Do not ingest or inhale the product.
- Do not use the product after the expiry date.
- Do not use the product if it show any evidence of contamination or any sign of deterioration.
- Do not use the product if the packaging is damaged.
- Any change or modification in the procedure may affect the results.
- Any change or modification of the required storage temperature may affect the performance of the product.
- Unappropriate storage may affect the shelf life of the product.
- Recap the bottles/vials tightly after each preparation and keep them in a low humidity environment, protected from moisture and light.
- Reading and interpretation should be performed using isolated colonies.
• Interpretation of the test results should be made taking into consideration colonial and microscopic morphology and if necessary, the results of any other tests performed.
• Laboratory, chemical or biohazardous wastes must be handled and discarded in accordance with all local and national regulations.
• For hazard and precaution recommendations related to some chemical components in this medium, please refer to the pictogram(s) mentioned on the labels. The Safety Data Sheet (SDS) is available on www.chromagar.com

DISPOSAL OF WASTE
After use, all plates and any other contaminated materials must be sterilized or disposed of by appropriate internal procedures and in accordance with local legislations. Plates can be destroyed by autoclaving at 121 °C for at least 20 minutes.

LITERATURE REFERENCES
Please refer to our website page «Publications» for scientific publications about this particular product.
Web link: http://www.chromagar.com/publication.php

IFU/LABEL INDEX
REF Catalogue reference
 Consult instructions for use
 Quantity of powder sufficient for X liters of media
 Expiry date
 Required storage temperature
 Store away from humidity
 Protect from light
 Manufacturer

REVISION HISTORY
This is version V6.0 of this document
Changing version is related to the new 3 pages format of the IFU.