CHROMagar™ STEC

MEDIUM PURPOSE
Chromogenic medium for detection of Shiga-Toxin E.coli (STEC).

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>30.8 g/L</td>
<td>10ml/L</td>
</tr>
<tr>
<td>Composition g/L</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Agar 15.0 Peptones and yeast extract 8.0</td>
<td>Selective mix</td>
<td></td>
</tr>
<tr>
<td>Salts 5.2 Chromogenic mix 2.6</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Aspect</td>
<td>Powder Form</td>
<td>freeze dried vials</td>
</tr>
<tr>
<td>STORAGE</td>
<td>15/30°C</td>
<td>15/30°C</td>
</tr>
<tr>
<td>FINAL MEDIA pH</td>
<td>6.9 +/- 0.2</td>
<td></td>
</tr>
</tbody>
</table>

PREPARATION (Calculation for 1L)

**Step 1**
Preparation of the base CHROMagar STEC base (B)
- Disperse slowly 30.8g of powder base in 1L of purified water.
- Stir until agar is well thickened.
- Heat and bring to boil (100°C) while swirling or stirring regularly. DO NOT HEAT TO MORE THAN 100°C. DO NOT AUTOCLAVE AT 121°C.
  - Warning 1: If using an autoclave, do so without pressure.
  - Advice 1: For the 100°C heating step, mixture may also be brought to a boil in a microwave oven after initial boiling, remove from oven, stir gently, then return to oven for short repeated bursts of heating until complete fusion of the agar grains has taken place (large bubbles replacing foam).
- Cool in a water bath to 45-50°C. Swirl or stir gently to homogenize.

**Step 2**
Preparation of the Supplement (S) and Mix of the prepared mix (B)
- Aseptically rehydrate TWO freeze dried vials of CHROMagar STEC supplement, ref ST160(S) with 5ml of sterile water each.
- Swirl well until complete dissolution.
- Add these 2x5 ml solutions of CHROMagar STEC supplement to the CHROMagar STEC base cooled at 45-50°C.
- Swirl gently to homogenize.
  - OR
- Using Supplement Reference ST162(S)
  - Aseptically rehydrate ONE vial with 10ml of sterile water.
  - Swirl well until complete dissolution.
  - Add this rehydrated solution to the CHROMagar STEC base cooled at 45-50°C.
  - Swirl gently to homogenize.

**Step 3**
Pour plates
- 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.
  - Advice 2: Plates can be stored for up to one month under refrigeration (2/8°C) if properly prepared and protected from light and dehydration.
  - Advice 3: If not fully used, rehydrated CHROMagar STEC supplement can be stored up to 2 months at 2/8°C.

INOCULATION
- If the agar plate has been refrigerated, allow to warm to room temperature before inoculation.
- Streak sample onto plate.
- Incubate in aerobic conditions at 37°C for 18-24 hours.

Final Media HELPING CALCULATION
- 500ml use ONE freeze dried vial
- 1 L use both freeze dried vials
- 1 vial -> qsf 500ml

Final Media HELPING CALCULATION
- 1 L use one vial
- 5 L use 5 vials
- 1 vial -> qsf 1 liter

Typical Samples
- All types of samples
- Possible enrichment step
  - Direct streaking or spreading technique
INTERPRETATION

<table>
<thead>
<tr>
<th>Microorganism</th>
<th>Typical colony appearance</th>
</tr>
</thead>
<tbody>
<tr>
<td>STEC E.coli</td>
<td>mauve</td>
</tr>
<tr>
<td>Other Enterobacteriaceae</td>
<td>colourless, blue or inhibited</td>
</tr>
<tr>
<td>Gram positive bacteria</td>
<td>inhibited</td>
</tr>
<tr>
<td>Note: fluorescence under UV lamp (365nm.) :</td>
<td></td>
</tr>
<tr>
<td>STEC E.coli O157</td>
<td>non fluorescent</td>
</tr>
<tr>
<td>STEC E.coli non O157</td>
<td>+/- fluorescent</td>
</tr>
</tbody>
</table>

**Typical colony appearance**

![Image](image-url)

The pictures shown are not contractual.

LIMITATIONS

- Final confirmation as STEC E.coli must be done by appropriate methods.
- Some STEC E.coli could have a poor or no growth on the media.
- Some rare strains of non-STEC E.coli could appear as mauve colonies w/o fluorescence.
- Rare O157 are fluo +.
- Serotypes with agglutination tests can be performed directly from the colony.

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 ATCC strains below:

<table>
<thead>
<tr>
<th>Microorganism</th>
<th>Typical colony appearance</th>
</tr>
</thead>
<tbody>
<tr>
<td>E.coli O157 ATCC® 35150</td>
<td>mauve</td>
</tr>
<tr>
<td>E.cloacae ATCC® 13047</td>
<td>metallic blue</td>
</tr>
<tr>
<td>E.coli ATCC® 25922</td>
<td>inhibited</td>
</tr>
</tbody>
</table>

WARNINGS

- Do not use plates if they show any evidence of contamination or any sign of deterioration.
- Do not use the product beyond its expiry date or if product shows any evidence of contamination or any sign of deterioration.
- For Research Use Only. Not for use in Diagnostic Procedure. Performance has not been established. This laboratory product should be used only by trained personnel in compliance with good laboratory practices.
- 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.
- 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.

REFERENCES

Please refer to our website page «Publications» for scientific publications about this particular product.

Web link: http://www.chromagar.com/publication.php

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.

IFU/LABEL INDEX

- Quantity of powder sufficient for X liters of media
- Expiry date
- Required storage temperature
- Store away from humidity

Pack Size

<table>
<thead>
<tr>
<th>1000 ml</th>
<th>50 Tests of 20ml = ST160</th>
</tr>
</thead>
<tbody>
<tr>
<td>5000 ml</td>
<td>250 Tests of 20ml = ST162</td>
</tr>
</tbody>
</table>

Microorganism | Typical colony appearance |
--------------|---------------------------|
E.faecalis ATCC® 29212 | inhibited |

Need some Technical Documents?

- Available for download on www.CHROMagar.com
  - Certificate of Analysis (CoA) → One per Lot
  - Material Safety Data Sheet (MSDS)