DHL AGAR (7457)

Intended Use
DHL Agar is used for the detection and isolation of pathogenic Enterobacteriaceae.

Product Summary and Explanation
Sakazaki et. al.\(^1,2\) proposed a modification of Deoxycholate Agar, referred to as DHL Agar. DHL Agar is also known as Deoxycholate Hydrogen Sulfide Lactose Agar. This medium provides contains a rich nitrogen base to permit growth of fastidious strains of Salmonella and Shigella. The colonies formed are considerably larger than those found on similar selective media.

Principles of the Procedure
Enzymatic Digest of Casein and Enzymatic Digest of Animal Tissue are the nitrogen and vitamin sources in DHL Agar. Sucrose permits differentiation of weakly lactose-positive, lactose-negative, or sucrose-positive species from sucrose and lactose-negative Enterobacteriaceae. H\(_2\)S production is indicated by a blackening of the colonies due to formation of iron sulfide. Sodium Deoxycholate, Sodium Thiosulfate, Ammonium Iron Citrate, and Sodium Citrate inhibit growth of Gram-positive bacteria. Neutral Red is a pH indicator. Agar is the solidifying agent.

Formula / Liter

<table>
<thead>
<tr>
<th>Component</th>
<th>Amount</th>
</tr>
</thead>
<tbody>
<tr>
<td>Enzymatic Digest of Casein</td>
<td>5 g</td>
</tr>
<tr>
<td>Enzymatic Digest of Animal Tissue</td>
<td>5 g</td>
</tr>
<tr>
<td>Lactose</td>
<td>10 g</td>
</tr>
<tr>
<td>Sodium Deoxycholate</td>
<td>1 g</td>
</tr>
<tr>
<td>Sodium Chloride</td>
<td>5 g</td>
</tr>
<tr>
<td>Dipotassium Phosphate</td>
<td>2 g</td>
</tr>
<tr>
<td>Ferric Citrate</td>
<td>1 g</td>
</tr>
<tr>
<td>Sodium Citrate</td>
<td>1 g</td>
</tr>
<tr>
<td>Neutral Red</td>
<td>0.03 g</td>
</tr>
<tr>
<td>Agar</td>
<td>16 g</td>
</tr>
</tbody>
</table>

Final pH: 7.2 ± 0.2 at 25°C
Formula may be adjusted and/or supplemented as required to meet performance specifications.

Precautions
1. For Laboratory Use.
2. IRRITANT. Irritating to eyes, skin, and respiratory system. May be harmful if swallowed or inhaled.

Directions
1. Suspend 46 g of the medium in one liter of purified water.
2. Heat with frequent agitation and boil for one minute to completely dissolve the medium.
3. DO NOT AUTOCLAVE.

Quality Control Specifications
Dehydrated Appearance: Powder is homogeneous, free flowing and beige to pink-beige.

Prepared Appearance: Prepared medium is light to medium pinkish red and trace to slightly hazy
Expected Cultural Response: Cultural response on DHL Agar incubated at 35 ± 2.0°C and examined for growth at 18 – 24 hours incubation.

<table>
<thead>
<tr>
<th>Microorganism</th>
<th>Response</th>
<th>Reactions</th>
</tr>
</thead>
<tbody>
<tr>
<td>Enterococcus faecalis ATCC® 29212</td>
<td>partial to complete inhibition</td>
<td>colorless colonies</td>
</tr>
<tr>
<td>Escherichia coli ATCC® 25922</td>
<td>growth</td>
<td>red colonies</td>
</tr>
<tr>
<td>Klebsiella pneumoniae ATCC® 13883</td>
<td>growth</td>
<td>pink colonies</td>
</tr>
<tr>
<td>Klebsiella pneumoniae ATCC® 10031</td>
<td>growth</td>
<td>pink colonies</td>
</tr>
<tr>
<td>Proteus vulgaris ATCC® 13315</td>
<td>growth</td>
<td>pink colonies</td>
</tr>
<tr>
<td>Proteus mirabilis ATCC® 12453</td>
<td>growth</td>
<td>colorless ± black centers</td>
</tr>
<tr>
<td>Salmonella typhi ATCC® 19430</td>
<td>growth</td>
<td>colorless ± black centers</td>
</tr>
<tr>
<td>Salmonella choleraesuis ATCC® 13076</td>
<td>growth</td>
<td>colorless colonies w/ black centers</td>
</tr>
<tr>
<td>Salmonella typhimurium ATCC® 14028</td>
<td>growth</td>
<td>colorless colonies w/ black centers</td>
</tr>
<tr>
<td>Shigella flexneri ATCC® 12022</td>
<td>growth</td>
<td>colorless colonies</td>
</tr>
<tr>
<td>Staphylococcus aureus ATCC® 25923</td>
<td>inhibition</td>
<td>colorless colonies</td>
</tr>
</tbody>
</table>

The organisms listed are the minimum that should be used for quality control testing.

Test Procedure
For a complete discussion on the isolation and identification of enteric bacilli refer to appropriate references.

Results
Differentiation of enteric bacilli is based on fermentation of lactose. Bacteria that ferment lactose produce acid and, in the presence of Neutral Red, form pink to red colonies. Bacteria that do not ferment lactose form colorless colonies. The majority of normal intestinal bacteria ferment lactose (red colonies) while Salmonella spp. and Shigella spp. do not ferment lactose (colorless colonies).

Storage
Store dehydrated medium at 2 - 30°C. Once opened and recapped, place container in a low humidity environment at the same storage temperature. Protect from moisture and light by keeping container tightly closed.

Expiration
Refer to expiration date stamped on the container. The dehydrated medium should be discarded if not free flowing, or if appearance has changed from the original color. Expiry applies to medium in its intact container when stored as directed.

Limitation of the Procedure
Due to nutritional variation, some strains may be encountered that grow poorly or fail to grow on this medium.

Packaging
DHL Agar    Code No.    7457A  500 g
            7457B  2 kg
            7457C  10 kg

References

Technical Information
Contact Acumedia Manufacturers, Inc. for Technical Service or questions involving dehydrated culture media preparation or performance at (517)372-9200 or fax us at (517)372-2006.