



## MacCONKEY AGAR W/ SORBITOL (7320)

### **Intended Use**

**MacConkey Agar W/ Sorbitol** is used for the isolation of pathogenic *Escherichia coli*.

### **Product Summary and Explanation**

MacConkey Agar W/ Sorbitol is based on the formula by Rappaport and Henig.<sup>1</sup> Originally developed for isolating enteropathogenic (EPEC) *serotypes*, O11 and O55, this medium is recommended for the isolation and differentiation of enterohemorrhagic *E. coli* O157:H7. This organism causes hemorrhagic colitis, which results in bloody diarrhea and can lead to kidney failure and death.<sup>2</sup> Serotype O157 has been implicated in serious foodborne diseases.

MacConkey Agar W/ Sorbitol contains sorbitol instead of lactose for differentiating enteropathogenic *E. coli* serotypes; these strains are typically sorbitol negative. MacConkey Agar W/ Sorbitol is recommended for clinical and food testing.<sup>2-4</sup>

### **Principles of the Procedure**

Enzymatic Digest of Gelatin, Enzymatic Digest of Casein, and Enzymatic Digest of Animal Tissue are the nitrogen and vitamin sources in MacConkey Agar W/ Sorbitol. Sorbitol is the fermentable carbohydrate; typically enteropathogenic strains produce colorless colonies. Bile Salts Mixture and Crystal Violet are the selective agents, inhibiting Gram-positive cocci. Sodium Chloride maintains the osmotic environment, and Neutral Red is the pH indicator. Agar is the solidifying agent.

### **Formula / Liter**

|  |         |
|--|---------|
| Enzymatic Digest of Gelatin .....      | 17 g    |
| Enzymatic Digest of Casein .....       | 1.5 g   |
| Enzymatic Digest of Animal Tissue..... | 1.5 g   |
| Sorbitol.....                          | 10 g    |
| Bile Salts Mixture .....               | 1.5 g   |
| Sodium Chloride .....                  | 5 g     |
| Neutral Red.....                       | 0.03 g  |
| Crystal Violet.....                    | 0.001 g |
| Agar .....                             | 13.5 g  |
| Final pH: 7.1 ± 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, respiratory system, and skin.

### **Directions**

1. Suspend 50 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. Autoclave at 121°C for 15 minutes.
4. Cool to 45 - 50°C and dispense into sterile Petri dishes.

### **Quality Control Specifications**

**Dehydrated Appearance:** Powder is homogeneous, free flowing, and light pink-beige.

**Prepared Appearance:** Prepared medium is trace to slightly hazy, and medium to dark pink-purple.

**Expected Cultural Response:** Cultural response on MacConkey Agar W/ Sorbitol incubated at the appropriate atmosphere and temperature and examined for growth after 18 - 24 hours.

| Microorganism                            | Approx. Inoculum (CFU) | Expected Results                                      |
|--|------------------------|---|
| <i>Escherichia coli</i> ATCC® 25922      | 10 - 300               | Good growth, pink colonies are Sorbitol positive      |
| <i>Escherichia coli</i> ATCC® 35150      | 10 - 300               | Good growth, colorless colonies are Sorbitol negative |
| <i>Staphylococcus aureus</i> ATCC® 25923 | ~ 10 <sup>3</sup>      | Inhibited   |

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

### Test Procedure

Refer to the appropriate references for specific procedures using MacConkey Agar W/ Sorbitol.<sup>2-4</sup>

### Results

*E. coli* O157:H7, and other organisms that do not ferment sorbitol, are colorless on MacConkey Agar W/ Sorbitol. Sorbitol-fermenting organisms produce pink colonies. Confirmatory biochemical and serological testing should be performed on suspected colonies.

### Storage

Store dehydrated medium at 2 - 30°C. Once opened and recapped, place the 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 the appearance has changed from the original color. Expiry applies to medium in its intact container when stored as directed.

### Limitations of the Procedure

1. Due to nutritional variation, some strains may be encountered that grow poorly or fail to grow on this medium.
2. Colonies that are sorbitol positive can revert, and can be mistaken for sorbitol negative.<sup>5</sup>
3. *E. coli* O157:H7 can ferment sorbitol after prolonged incubation.<sup>5</sup>

### Packaging

|                                   |                 |              |              |
|-----------------------------------|-----------------|--------------|--------------|
| <b>MacConkey Agar W/ Sorbitol</b> | <b>Code No.</b> | <b>7320A</b> | <b>500 g</b> |
|                                   |                 | <b>7320B</b> | <b>2 kg</b>  |
|                                   |                 | <b>7320C</b> | <b>10 kg</b> |

### References

1. **Rappaport, F., and E. Henig.** 1952. Media for the isolation and differentiation of pathogenic *Escherichia coli* (serotypes 0111 and 055). J. Clin. Pathol. 5:361-362.
2. **www.fda.gov/Food/ScienceResearch/LaboratoryMethods/BacteriologicalAnalyticalManualBAM/default.htm.**
3. **Vanderzant, C., and D. F. Splittstoesser (eds.)**. 1992. Compendium of methods for the microbiological examination of food, 3<sup>rd</sup> ed. American Public Health Association, Washington, D.C.
4. **Murray, P. R., E. J. Baron, M. A. Pfaller, F. C. Tenover, and R. H. Tenover (eds.)**. 1995. Manual of clinical microbiology, 6<sup>th</sup> ed. American Society for Microbiology, Washington, D.C.
5. **Adams, S.** 1991. Screening for verotoxin-producing *Escherichia coli*. Clin. Lab. Sci. 4:19-20.

### 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.