

Instructions for Use

MACCONKEY AGAR WITH SORBITOL (SMAC)

Cat. no. G36	MacConkey Agar with Sorbitol, 15x100mm Plate, 18ml	10 plates/bag
Cat. no. J49	CIN / MacConkey Agar with Sorbitol, 15x100mm Biplate, 10ml/10ml	10 plates/bag

INTENDED USE

Hardy Diagnostics MacConkey Agar with Sorbitol is to be used as a selective and differential medium for the detection of enterohemorrhagic *Escherichia coli* O157:H7.

SUMMARY

E. coli O157:H7 is an enteric pathogen that typically causes hemorrhagic colitis and bloody diarrheal illnesses. It may be followed by hemolytic uremic syndrome, especially in young children. MacConkey Agar with Sorbitol is recommended for isolation of this organism. Rappaport and Henig first described the formulation of the medium, and it was later confirmed by March and Ratham who reported MacConkey Agar with Sorbitol to have a sensitivity of 100% and a specificity of 85%. This medium has proved to be an inexpensive, rapid, simple yet reliable means for the detection of *E. coli* O157:H7.

FORMULA

Ingredients per liter of deionized water:*

Pancreatic Digest of Gelatin	17.0gm
Sorbitol	10.0gm
Sodium Chloride	5.0gm
Pancreatic Digest of Casein	1.5gm
Peptic Digest of Animal Tissue	1.5gm
Bile Salts Mixture	1.5gm
Neutral Red	30.0mg
Crystal Violet	1.0mg
Agar	13.5gm

Final pH 7.1 +/- 0.2 at 25°C.

* Adjusted and/or supplemented as required to meet performance criteria.

STORAGE AND SHELF LIFE

Storage: Upon receipt store at 2-8°C. away from direct light. Media should not be used if there are any signs of deterioration (shrinking, cracking, or discoloration), contamination, or if the expiration date has passed. Product is light and temperature sensitive; protect from light, excessive heat, moisture, and freezing.

The expiration date on the product label applies to the product in its intact packaging when stored as directed. The product may be used and tested up to the expiration date on the product label and incubated for the recommended incubation times as stated below.

Refer to the document "[Storage](#)" for more information.

PRECAUTIONS

This product may contain components of animal origin. Certified knowledge of the origin and/or sanitary state of the animals does not guarantee the absence of transmissible pathogenic agents. Therefore, it is recommended that these products be treated as potentially infectious, and handle observing the usual Universal Precautions for blood. Do not ingest, inhale, or allow to come into contact with skin.

This product is for *in vitro* diagnostic use only. It is to be used only by adequately trained and qualified laboratory personnel. Observe approved biohazard precautions and aseptic techniques. All laboratory specimens should be considered infectious and handled according to "standard precautions." Refer to the document "[Guidelines for Isolation Precautions](#)" from the Centers for Disease Control and Prevention.

For additional information regarding specific precautions for the prevention of the transmission of all infectious agents from laboratory instruments and materials, and for recommendations for the management of exposure to infectious disease, refer to CLSI document M29: *Protection of Laboratory Workers from Occupationally Acquired Infections*.

Sterilize all biohazard waste before disposal.

Refer to the document "[Precautions When Using Media](#)" for more information.

PROCEDURE

Specimen Collection: Consult appropriate references to determine how to correctly collect the specimen to be tested (stool, food, etc.).

Method of Use: Allow plates to warm to room temperature. The agar surface should be dry before inoculating. Inoculate the plates with a suspension of the sample to be tested in a dilution small enough to produce isolated colonies. Incubate plates aerobically at 37°C. for 24 hours. Examine media macroscopically for typical colonies.

INTERPRETATION OF RESULTS

Colony Morphology: *E. coli* O157:H7 forms colorless colonies, but otherwise typical of that produced by other *E. coli* species. Any bacteria capable of fermenting sorbitol (including other *E. coli* species) form pink colonies on MacConkey Agar with Sorbitol.

LIMITATIONS

The MacConkey Agar with Sorbitol may be used as an aid in the identification of bacteria. Additional biochemical and/or serological testing using pure culture is recommended for complete identification.

Reading of MacConkey Agar with Sorbitol beyond 24 hours should be avoided since the pink color fades in sorbitol-fermenting colonies. Likewise, an extremely heavy inoculum of sorbitol-fermenters may prematurely exhaust the sorbitol and yield white to off-white colonies.

Other gram-negative organisms are able to grow on MacConkey Agar with Sorbitol . However, colony appearance is generally enough to differentiate these organisms from *E. coli* O157:H7.

Sorbitol-negative colonies may be presumptively identified as *E. coli* O157 using our *E. coli*PRO™ O157 Kit (Cat. no. PL070HD). Further serotyping with H7 antiserum is necessary for definitive identification (Cat. no. 221591).

Refer to the document "[Limitations of Procedures and Warranty](#)" for more information.

MATERIALS REQUIRED BUT NOT PROVIDED

Standard microbiological supplies and equipment such as loops, other culture media, swabs, applicator sticks, incinerators, and incubators, etc., as well as serological and biochemical reagents, are not provided.

QUALITY CONTROL

Hardy Diagnostics tests each lot of commercially manufactured media using appropriate quality control microorganisms and quality specifications as outlined on the Certificate of Analysis (CofA) and the CLSI document M22-A3 *Quality Assurance for Commercially Prepared Microbiological Culture Media*. The following microorganisms are routinely used for testing at Hardy Diagnostics:

Test Organisms	Inoculation Method*	Incubation			Results
		Time	Temperature	Atmosphere	
<i>Escherichia coli</i> O157:H7 ATCC® 35150	A	24hr	35°C	Aerobic	Clear colonies seen; no fermentation of sorbitol
<i>Escherichia coli</i> ATCC® 25922	A	24hr	35°C	Aerobic	Pink colonies seen; fermentation of sorbitol
<i>Enterococcus faecalis</i> ATCC® 29212	B	24hr	35°C	Aerobic	Partial to complete inhibition

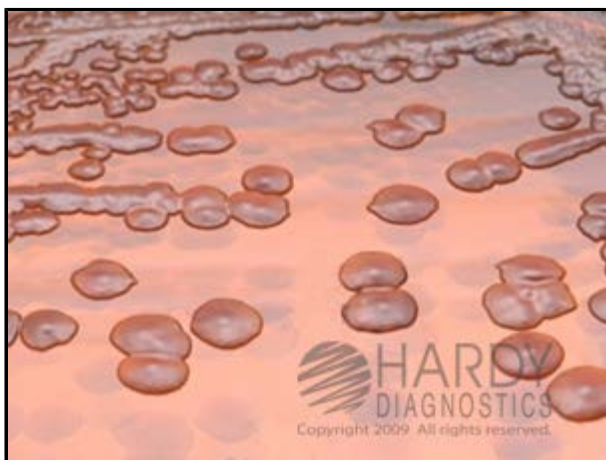
* Refer to the document "[Inoculation Procedures for Media QC](#)" for more information.

USER QUALITY CONTROL

End users of commercially prepared culture media should perform QC testing in accordance with applicable government regulatory agencies, and in compliance with accreditation requirements. Hardy Diagnostics recommends end users check for signs of contamination and deterioration and, if dictated by laboratory quality control procedures or regulation, perform quality control testing to demonstrate growth or a positive reaction and to demonstrate inhibition or a negative reaction, if applicable. Hardy Diagnostics quality control testing is documented on the certificate of analysis (CofA) available from Hardy Diagnostics [Certificate of Analysis](#) website. Also refer to the document "[Finished Product Quality Control Procedures](#)," and the CLSI document M22-A3 *Quality Assurance for Commercially Prepared Microbiological Culture Media* for more information on the appropriate QC procedures. See the references below.

PHYSICAL APPEARANCE

MacConkey Agar with Sorbitol should appear slightly opalescent, and reddish-purple in color.



Escherichia coli O157:H7 (ATCC® 35150) colonies growing on MacConkey Agar with Sorbitol (Cat. no. G36). Incubated aerobically for 24 hours at 35°C.



Escherichia coli (ATCC® 25922) colonies growing on MacConkey Agar with Sorbitol (Cat. no. G36). Incubated aerobically for 24 hours at 35°C.



Uninoculated plate of MacConkey Agar with Sorbitol (Cat. no. G36).

REFERENCES

1. Jorgensen., et al. *Manual of Clinical Microbiology*, American Society for Microbiology, Washington, D.C.
2. Anderson, N.L., et al. *Cumitech 3B; Quality Systems in the Clinical Microbiology Laboratory*, Coordinating ed., A.S. Weissfeld. American Society for Microbiology, Washington, D.C.
3. Rappaport, F. and E. Henig. 1952. *J. Clin. Path.*; 5:361.
4. March, S.B., S. Ratnam, et al. 1986. Sorbitol-MacConkey medium for detection of *Escherichia coli* O157:H7 associated with hemorrhagic colitis. *J. Clin. Microbiol.*; 23:869-872.
5. March, S.B., S. Ratnam, et al. 1988. Characterization of *Escherichia coli* serotype O157:H7, *J. Clin. Microbiol.*;

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ATCC is a registered trademark of the American Type Culture Collection.

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