MACCONKEY AGAR w/ SORBITOL

INTENDED USE
Remel MacConkey Agar w/ Sorbitol is a solid medium recommended for use in qualitative procedures as a differential and selective medium for the detection of sorbitol-negative *Escherichia coli*.

SUMMARY AND EXPLANATION
Escherichia coli serotype O157:H7 (VTEC) produces a verotoxin which causes hemorrhagic colitis outbreaks, with symptoms ranging from mild diarrhea to idiopathic hemolytic uremic syndrome.[1,2] On routine isolation media, colonies of VTEC are indistinguishable from those of non-VTEC *E. coli.* In 1986, March and Ratnam introduced MacConkey Agar w/ Sorbitol for isolation of verotoxin-producing *E.coli* O157:H7 from clinical specimens. Unlike most serotypes of *E.coli*, VTEC does not ferment sorbitol. Haldane et al. implemented the use of MacConkey Agar w/ Sorbitol as a primary isolation medium to distinguish verotoxin-producing strains of *E. coli* which are sorbitol-negative from non-VTEC *E. coli* which is sorbitol-positive.[3]

PRINCIPLE
Gelatin, casein, and meat peptones provide nitrogen, amino acids, and peptides essential for bacterial growth. Sodium chloride is a source of essential electrolytes and maintains osmotic equilibrium. Bile salts and crystal violet are selective agents which inhibit gram-positive organisms. Lactose is omitted from this formulation and is substituted with D-sorbitol. Verotoxin-producing strains of *E. coli* ferment lactose but not D-sorbitol and form colorless colonies on MacConkey Agar w/ Sorbitol.

REAGENTS (CLASSICAL FORMULA)*

Gelatin Peptone	17.0	g	Meat Peptone	1.5	g
D-Sorbitol	10.0	g	Neutral Red	30.0	mg
Sodium Chloride	5.0	g	Crystal Violet	1.0	mg
Bile Salts	1.5	g	Agar	13.5	g
Casein Peptone	1.5	g	Demineralized Water	1000.0	ml

pH 7.1 ± 0.2 @ 25°C

*Adjusted as required to meet performance standards.

PROCEDURE
1. Inoculate and streak the specimen as soon as possible after it is received in the laboratory.
2. If material is being cultured directly from a swab, roll the swab over a small area of the agar surface and streak for isolation.
3. Selective and nonselective media should be inoculated to increase the chance of recovering enteric pathogens when the number of gram-negative organisms is low and to provide isolation of other organisms present in the specimen.
4. Incubate plates in ambient air at 33-37°C for 18-24 hours.
5. Observe plates for typical colonial morphology.

Pour Tube: Melt the pour tube in a boiling water bath and cool to 45-50°C. Mix and dispense into a sterile petri dish and proceed with the instructions above.

QUALITY CONTROL
All lot numbers of MacConkey Agar w/ Sorbitol have been tested using the following quality control organisms and have been found to be acceptable. Testing of control organisms should be performed in accordance with established laboratory quality control procedures. If aberrant quality control results are noted, patient results should not be reported.

CONTROL	INCUBATION	RESULTS
Escherichia coli ATCC® 43888	Ambient, 18-24 h @ 33-37°C	Growth, colorless colonies
Escherichia coli ATCC® 25922	Ambient, 18-24 h @ 33-37°C	Growth, pink colonies
Proteus mirabilis ATCC® 12453	Ambient, 18-24 h @ 33-37°C	Growth, colorless colonies, swarming inhibited
Salmonella enterica serovar Typhimurium ATCC® 14028	Ambient, 18-24 h @ 33-37°C	Growth, pink colonies
Enterococcus faecalis ATCC® 29212	Ambient, 18-24 h @ 33-37°C	Inhibition (partial to complete)
Staphylococcus aureus ATCC® 25923	Ambient, 18-24 h @ 33-37°C	Inhibition (partial to complete)

LIMITATIONS
1. Sorbitol-negative isolates should be confirmed as *E. coli* by current accepted methodologies. Confirmed *E. coli* isolates should be serotyped using an *E. coli* O157 and H7 antiserum. These strains are reportable to State Public Health Departments.[4]

BIBLIOGRAPHY

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Refer to the front of Remel *Technical Manual of Microbiological Media* for **General Information** regarding precautions, product storage and deterioration, specimen collection, storage and transportation, materials required, quality control, and limitations.

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