# MacCONKEY SORBITOL AGAR w/ CEFIXIME & TELLURITE (CTSMAC)

- For in vitro use only - Catalogue No. PM22 (K)

Our MacConkey Sorbitol Agar with Cefixime and Tellurite (CTSMAC) is a selective medium used for the isolation and differentiation of *Escherichia coli* 0157:H7.

The majority of outbreaks of hemorrhagic colitis have been caused by *Escherichia coli* serotype O157:H7. MacConkey Sorbitol Agar is based on the work of Rappaport and Henig and Its usefulness for differentiating *E. coli* O157:H7 from other non-hemorrhagic *E. coli* was later confirmed by March and Ratnam. The substitution of sorbitol in place of lactose allows for differentiation: *E. coli* 0157:H7 does not ferment sorbitol and forms colorless colonies while other *E. coli* strains ferment sorbitol and form typical pink colonies. MacConkey Sorbitol Agar is not very selective and therefore ineffective for food testing, since the normal bacterial flora present can easily outgrow O157:H7 strains. It was later discovered that the addition of cefixime and tellurite produced a highly selective medium suitable for isolating O157:H7.

Both the FDA and the Canadian Health Protection Branch recommend the use of CTSMAC for the isolation of *E. coli* O157:H7 from foods. In testing foods, an enrichment step in EHEC Enrichment Broth (EEB) is prescribed.

**Formula per Litre of Medium**

| | |
|---|---|
| Pancreatic digest of gelatin | 15.5 g |
| Pancreatic digest of animal tissue | 3.0 g |
| D-Sorbitol | 10.0 g |
| Bile Salts | 1.5 g |
| Sodium chloride | 5.0 g |
| Agar | 15.0 g |
| Neutral red | 0.03 g |
| Crystal violet | 0.001 g |
| Potassium tellurite | 2.5 mg |
| Cefixime | 0.05 mg |

pH 7.1 ± 0.2

**Recommended Procedure (FDA)**

1. Weigh 25 g of food into 225 mL of EEB, blend or stomach briefly as necessary.
2. Incubate at 37 ± 0.5°C with shaking for 24 h.
3. After incubation, spread 0.1 mL of appropriately diluted enrichment broth onto CTSMAC plates to obtain 100 to 300 isolated colonies.
4. Streak one loop-full of the enrichment to one additional CTSMAC plate.
5. Incubate plates at 35-37°C for 18 - 24 h.
6. Pick up to 5 typical O157:H7 colonies from TCSMAC onto Trytic Soy Agar Yeast Extract (TSAYE) plates and incubate at 35°C for 18 to 24 h. Refer to FDA literature for more details.

**Interpretation of Results**

*Escherichia coli* serotype O157:H7 does not ferment sorbitol and therefore produces colorless colonies.

Due to the inhibitory nature of the medium other *E. coli* strains may be inhibited on this medium. *E. coli* strains able to grow on this medium will ferment sorbitol and produce pink colonies.

Additional biochemical and/or serological tests should be performed on isolated colonies from pure culture in order to complete identification.

- *Prolonged incubation can result in fading of pink-colored sorbitol-positive colonies*

- *Although most non-O157:H7 E. coli ferment sorbitol, about 6% of the isolates do not. These atypical strains along with other sorbitol non-fermenting bacteria such as Morganella and Hafnia appear identical to O157:H7 colonies on CTSMAC agar and therefore confirmatory tests may be necessary*

## Quality Control

After checking for correct pH, colour, depth, and sterility, the following organisms are used to determine the growth performance of the completed medium.

| Organism | Expected Result |
|---|---|
| *Escherichia coli* (O157:H7) ATCC 35150 | Growth, colorless colonies |
| *Staphylococcus aureus* ATCC 25923 | Inhibition |

## Storage and Shelf Life

Our MacConkey Sorbitol Agar with Cefixime and Tellurite should be stored away from direct light at 4 to 8°C. The medium side should be uppermost to prevent excessive accumulation of moisture on the agar surface. Under these conditions this medium has a shelf life of 6 weeks from the date of manufacture.

## Ordering Information

| Cat# | Description | Format |
|---|---|---|
| PM22 | MacConkey Sorbitol Agar with Cefixime and Tellurite [Standard 15x100-mm plate] | 10/pkg |
| PM22K | MacConkey Sorbitol Agar with Cefixime and Tellurite [Kirby 15x150-mm plate] | 5/pkg |

## References


1. Rappaport F, Henig E. Media for the isolation and differentiation of pathogenic *Escherichia coli* (serotypes 0111 and 055). J Clin Path 1952; 5:361-2.
2. Mehlman IJ. Coliforms, fecal coliforms, *Escherichia coli* and enteropathogenic *E. coli*. pp 265-85. In Speck ML, ed. Compendium of methods for the microbiological examination of foods, 2nd ed. Washington, DC: APHA, 1984.
3. March SB, Ratnam S. Sorbitol-MacConkey medium for detection of *Escherichia coli* O157:H7 associated with hemorrhagic colitis. J Clin Micro 1986; 23:869-72.
4. Zadik PM, Chapman PA, Siddons CA. Use of tellurite for the selection of verocytotoxigenic *Escherichia coli* O157. J Med Microbiol 1993; 39:155-8.
5. Fey PD, Wickert RS, Rupp ME, Safranek TJ, Hinrichs SH. Prevalence of non-O157:H7 shiga toxin-producing *Escherichia coli* in diarrheal stool samples from Nebraska. Emerg Infect Dis 2000; 6:530-3.
6. Warburton D. Isolation of *E. coli* O157:H7 in foods, MFLP-80. Government of Canada, March 2001. Retrieved November 1, 2002, from Food Directorate's (Health Canada's) website: http://www.hc-sc.gc.ca/food-ailment.
7. Feng P, Weagant SD. Diarrheagenic Escherichia coli. In FDA: Bacteriological analytical manual, September 2002. Retrieved January 27, 2003, from FDA website: http://www.cfsan.fda.gov/~ebam/bam-toc.html



Original: January 2003
Revised / Reviewed: October 2014