M-TEC Agar

M1391

M-TEC Agar is recommended for isolation, differentiation and rapid enumeration of thermotolerant *Escherichia coli* from water by membrane filtration.

Composition**

Ingredients	Gms / Litre
Proteose peptone	5.000
Yeast extract	3.000
Lactose	10.000
Sodium chloride	7.500
Potassium dihydrogen phosphate	1.000
Dipotassium hydrogen phosphate	3.300
Sodium lauryl sulphate	0.200
Sodium deoxycholate	0.100
Bromocresol purple	0.080
Bromphenol red	0.080
Agar	15.000
Final pH (at 25°C)	7.3±0.2

**Formula adjusted, standardized to suit performance parameters

Directions

Suspend 45.26 grams in 1000 ml distilled water. Heat to boiling to dissolve the medium completely. Sterilize by autoclaving at 15 lbs pressure (121°C) for 15 minutes. Cool to 45°C and pour into sterile Petri plates.

Principle And Interpretation

M-TEC Agar is recommended for rapid isolation, differentiation and rapid enumeration of thermotolerant *E. coli* from water by membrane filtration. TEC stands for thermotolerant *E. coli* , the presence of which is widely used as an indicator of faecal contamination in water. There are many procedures for enumerating *E. coli* based on its ability to grow at elevated temperatures and produce indole from tryptophan (1, 2). The determination of indole production along with MPN procedures requires the use of additional medium and additional incubation time. Dufour et al (3) developed a simple membrane filtration technique for rapid enumeration of E. coli, which quantified *E. coli* within 24 hours without requiring subculturing and identification of isolates.

M-TEC Agar and urea substrate are recommended for use in the detection of *E. coli* when evaluating microbiological quality of recreational water (2).

Proteose peptone and yeast extract act as source of nitrogen, carbon, amino acids and vitamins. Potassium phosphate salts help in buffering the medium. Lactose is the source of fermentable carbohydrate. Bromocresol purple and bromophenol red serve as indicator. Sodium lauryl sulphate and sodium deoxycholate inhibit gram-positive bacteria.

Membrane filters that are used for filtration are aseptically placed with face upwards on the surface of M-TEC Agar. These plates are then incubated at 44.5 ± 0.5°C. Following incubation, these filters are aseptically placed on sterile absorbent cotton pads saturated with urease substrate i.e. urea (approx. 2 ml). Urease substrate is prepared by dissolving 2 grams urea and 0.01 gram phenol red in 100 ml distilled water with the pH adjusted to 5.0 ± 0.2 (2). Urease-negative reaction or formation of yellow to yellow brown colonies observed after 15-20 minutes is confirmatory for presence of thermotolerant *E. coli* .

Quality Control

Appearance

Cream to greenish yellow homogeneous free flowing powder

Gelling

Firm, comparable with 1.5% Agar gel

Colour and Clarity of prepared medium
Dark purple coloured with red cast clear to slightly opalescent gel forms in Petri plates

Reaction
Reaction of 4.53% w/v aqueous solution at 25°C. pH : 7.3±0.2

pH
7.10-7.50

Cultural Response
M1391: Cultural characteristics observed after an incubation at 35-37°C for 2 hours and at 44.5°±0.5°C for 22 hours.

Organism	Inoculum (CFU)	Growth
Escherichia coli ATCC 25922	50-100	good (further testing using urease substrate should be performed)

Storage and Shelf Life
Store below 30°C in tightly closed container and the prepared medium at 2 - 8°C. Use before expiry date on the label.

Reference
1. Mara D. D., 1973, J. Hyg. 71: 783.
2. Clesceri L. S., Greenberg A. E. and Eaton A. D., (Ed.), 1998, Standard Methods for the Examination of Water and Waste water, 20th Ed., American Public Health Association, Washington, D.C.
3. Dufour A. P., Strickland E. R. and Cabelli V. J., 1981, Appl. Environ. Microbiol., 41: 1152

Revision : 2 / 2015