Reference: DSHB3084

Product:

CHAPMAN TTC AGAR (TERGITOL® 7 AGAR)

### Also known as

Tergitol 7 Agar; T7 Agar

## **Specification**

Medium for the detection of coliforms by membrane filtration in water analyses according to ISO 9308-1:2000 standard.

Formula * in g/L	
Meat peptone	10,000
Meat extract	
Lactose	20,000
Yeast extract	6,000
Bromothymol blue	0,050
Tergitol ® 7	0,100
Agar	15,000

Final pH 7,2 ±0,1 at 25 °C

#### Directions

Suspend 56,2 g in 1 L of distilled water and bring to the boil. Distribute into suitable containers and sterilize in the autoclave at 121°C for 15 minutes. Cool to 45-50°C. Add 2-3 mL/L of filtered sterile 1% aqueous 2, 3, 5-triphenyltetrazolium chloride (TTC) (Art. No DSHB3074) and pour plates. Do not reheat.

## **Description**

This medium is formulated for the presumptive identification of coliforms in drinking water, by membrane filtration according to ISO 9308-1:2000. Due to the instability of the triphenyltetrazolium it is provided in a separate container, sterilized and ready to use (Art. No. DSHB3074). Poured plates can be stored refrigerated for up to 8 days without losing their effectiveness. They should not be used if any signs of dehydration or drying appear.

### Technique

While using the membrane filter technique for the presumptive identification of coliforms in water, it should be kept in mind that the minimum volume to be filtered depends on the type of water being tested. If necessary dilute with sterile phosphate buffer in order to obtain the number of colonies on the membrane appropriate for counting.

For every water sample two volumes must be filtered over two different membranes and incubated on Tergitol ® 7 agar at 35°C and 44°C respectively.

After 48 hours typical colonies have the appearance as follow:

- Escherichia coli, Citrobacter spp.: Yellow with a centred orange nucleus under the membrane filter (MF).
- Klebsiella spp.: Brick red or yellow without a nucleus. The medium under the (MF) is yellow.
- Enterobacter spp.: Dark yellow or brick red with an orange nucleus. The medium is also yellow.
- Non lactose-fermenters: Violet or indigo colonies. The medium turns blue.

Most coliforms can not grow on this medium when incubated at 44°C, except E. coli which forms a colony with a characteristic appearance.

Results are always expressed per 100 ml sample including any applied dilutions. Estimation is done by taking typical colonies which have grown at 35°C as faecal coliforms, together with those grown at 44°C as E. coli. Nevertheless, according to legislation and despite the selectivity of medium, results can only be considered as presumptive and all coliform colonies have to be confirmed by following the criteria below:

Typical appearance in EMB Agar or Endo Agar Base and characteristic reactions in Kligler Iron Agar.

For the confirmation of faecal E. coli, the following characteristics are used for verification: a motile, Gram negative bacillus and lactose fermenter with acid and gas production, which gives negative results on the citrate test and indole production positive.

# **Quality control**

Incubation temperature: 36°C ±2,0 Incubation time: 44±4 h

Inoculum: Practical range 100 ± 20 CFU. Min. 50 CFU (Productivity) / 10<sup>4</sup>-10<sup>6</sup> CFU (Selectivity) / 10<sup>3</sup>-10<sup>4</sup>CFU

(Specificity) according to ISO 11133:2014/Amd 1:2018 . MF methods. **Growth Remarks** 

Microorganism Escherichia coli ATCC® 8739 Productivity > 0.50 Yellow-orange colonies under MF Escherichia coli ATCC® 11775 Productivity > 0.50 Yellow-orange colonies under MF Escherichia coli ATCC® 25922 Productivity > 0.50 Yellow-orange colonies under MF Enterococcus faecalis ATCC® 19433 Selectivity Total inhibition Citrobacter freundii ATCC® 43864 Productivity > 0.50 Yellow-orange colonies under MF Pseudomonas aeruginosa ATCC® 9027 Good (specificity) Dark red colonies, blue center E. coli NCTC® 13167 Productivity > 0.50 Yellow-orange colonies under MF

<sup>\*</sup> Adjusted and /or supplemented as required to meet performance criteria

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### References

- · ATLAS, R.M., L.C. PARKS (1993) Handbook of Microbiological Media. CRC Press, Inc. London.
- · CHAPMAN G.H. (1951) A culturé medium for detecting and confirming E. coli in ten hours. Am. J. Publ. Hlth 41:1381 -1386.
- · DOWNES, F.P. & K. ITO (2001) Compendium of Methods for the Microbiological Examination of Foods. 3rd ed. APHA. Washington.
- · GUINEA, SANCHO, PARES (1979) Análisis Microbiológico de Aguas. Ed. Omega. Barcelona.
- · ISO 9308-1:2000 Standard. Water Quality Detection and enumeration of Escherichia coli and coliform bacteria Part 1: Membrane filtration method.
- . ISO 11133:2014/ Adm 1:2018. Microbiology of food, animal feed and water. Preparation, production, storage and performance testing of culture media.
- · SPECK, M (Ed.) (1982) Compendium of Methods for the Microbiological Examination of Foods. 2nd ed. APHA. Washington.

## **Storage**

For laboratory use only. Keep tightly closed, away from bright light, in a cool dry place (+4 °C to 30 °C).