

Clostridium Perfringens Agar Base (m-CP)

Cat. 1132

For the enumeration and isolation of Clostridium perfringens in water samples.

Practical information

Applications	Categories
Selective enumeration	Clostridium perfringens
Selective isolation	Clostridium perfringens

Industry: Water

Regulations: ISO 11133 / Council Directive 98/83/EC



Principles and uses

Clostridium Perfringens Agar Base (m-CP) is a medium used for the rapid isolation and presumptive identification of Clostridium perfringens from water samples. m-CP Agar was first described by Bisson and Cabelli for the rapid quantisation of Clostridium perfringens from various water samples (seawater, drinking water and sewage) for human consumption and from environmental water samples. It is recommended in European Council Directive 98/83/EC for testing the quality of water intended for human consumption by the membrane filtration technique.

The medium was shown to give better recovery of Clostridium perfringens from water and sewage samples. It can be used for monitoring all types of waters. C. perfringens is present in large numbers in water and sewage and its spores are resistant to wastewater treatment practices, extremes in temperature, and environmental stress. The medium has been recommended for the examination of chlorinated waters and untreated water containing industrial wastes lethal to non-spore forming bacteria, sewage sludge, and situations in which the detection of remote as well as recent pollution is desirable.

Tryptose provides nitrogen, vitamins, minerals and amino acids essential for growth. Yeast extract is a source of vitamins, particularly of the B-group essential for bacterial growth. Sucrose is a complex carbohydrate energy source. L-Cysteine hydrochloride is the reducing agent and Bromocresol purple is the pH indicator. Bacteriological Agar is the solidifying agent.

Formula in g/L

Bacteriological agar	15	Bromocresol purple	0,04
L-Cysteine hydrochloride	1	Magnesium sulfate heptahydrated	0,1
Sucrose	5	Tryptose	30
Yeast extract	20		

Preparation

Suspend 35,57 grams of the medium in 500 ml of distilled water. Mix well and dissolve by heating with frequent agitation. Boil for one minute until complete dissolution. Sterilize in autoclave at 121 °C for 15 minutes. Cool to 45-50 °C and aseptically add the three vials of m-CP Supplement (Cat. 6073) per 500 ml of medium, previously reconstituted in 2 ml of sterile distilled water each. Mix well and distribute into sterile containers.

Instructions for use

Membrane filtration method:

- Filter through a membrane with a pore of 45 micrometers, a measured volume of the sample or a dilution of it.
- For water intended for human consumption, it is usual to filter a volume of 100 ml.
- Place the membrane grid face upwards on a m-CP agar plate ensuring that no air bubbles are trapped under the filter.
- Incubate the plates with the filters, anaerobically at 44±1 °C for 21±3 hours, inverted to avoid interference with the condensing water.

Quality control

Solubility	Appareance	Color of the dehydrated medium	Color of the prepared medium	Final pH (25°C)
w/o rests	Fine powder	Beige	Purple	7,6±0,2

Microbiological test

According tpo ISO 11133:

Incubation conditions: Productivity, Selectivity, Specifity (44±1 °C / 21±3 h).

Inoculation conditions: Productivity quantitative (100±20. Min. 50 CFU) / Selectivity (10³-10⁴ CFU) / Specificity (10⁴-10⁶ CFU).

Reference media: TSA or other non selective media for anaerobes.

Microrganisms	Specification	Characteristic reaction
Clostridium perfringens ATCC 13124	Good growth, >50 %	Yellow colonies; Phophatase test positive
Clostridium bifermentans CECT 550		Blue colonies; Phosphatase test negative
Escherichia coli ATCC 8739	Total inhibition (0)	

Storage

Temp. Min.:2 °C

Temp. Max.:25 °C

Bibliography

Armon, R., and Payment, P., 1988, A modified m-CP medium for enumerating Clostridium perfringens from water samples: Canadian Journal of Microbiology, v.34. p.78-79.

Bisson, J.W., and Cabelli, V.J., 1979, Membrane filter enumeration method for Clostridium perfringens: Applied and Environmental Microbiology, v. 37. no.1. p. 55-66

E.U. (1998) 98/83/EC of Council of 3rd of November 1998 on the quality of water intended for human consumption. Off. J. Eur. Commun., L330, 32-54