

## 75605 m-CP Agar Base (Membrane Clostridium perfringens Agar Base)

The m-CP Agar Base with selective supplement is recommended by the Directive of the Council of the European Union 98/83/EC for isolation and enumeration of *Clostridium perfringens* from water sample using membrane filtration technique.

### Composition:

Ingredients	Grams/Litre
Tryptose	30.0
Yeast extract	20.0
Sucrose	5.0
L-Cysteine hydrochloride	1.0
Magnesium sulphate. 7H <sub>2</sub> O	0.1
Bromo cresol purple	0.04
Ferric chloride. 6H <sub>2</sub> O	0.09
Indoxyl-β-D-Glucoside	0.06
Agar	15.0

Final pH (at 25 °C) 7.6 +/- 0.2

Store prepared media below 4 °C, protected from direct light. Store dehydrated powder in a dry place in tightly-sealed containers at 4 °C.

### Directions:

Suspend 35.6 g in 485 ml distilled water. Heat to boiling to dissolve the medium completely. Sterilize by autoclaving at 1 bar pressure (121 °C) for 15 minutes. Cool to 50 °C. Aseptically add the rehydrated contents of 1 vial of M-CP Selective Supplement I (Cat. No. 51962) and 1 vial of M-CP Selective Supplement II (Cat. No. 82265). Mix well and pour into sterile petri plates.

### Principle and Interpretation:

M-CP Agar Base is prepared as per the formula of Armon and Payment [1]. It is also recommended by the Directive of the Council of the European Union 98/83/EC [2] for isolation and enumeration of *Clostridium perfringens* from water sample using membrane filtration technique.

Tryptose and yeast extract provide nitrogenous compounds. Sucrose is the fermentable carbohydrate. Bromo cresol purple serves as a pH indicator, indoxyl-β-D-glucoside is a chromogenic substrate for β-D-glucosidase or cellobiase and phenolphthalein diphosphate for the detection of acid phosphatase. The addition of D-Cycloserine and Polymyxin B makes the medium inhibitory to accompanying non clostridial flora and thus allows analysis of both vegetative cells and spores of *Clostridium*. Further selectivity is provided by incubation under anaerobic conditions. In case of the presence of β-D-glucosidase or cellobiase the colonies are colored blue or green in combination of the ability to ferment sucrose. This would show that the colony is not *Clostridium perfringens*. Yellow (cellobiase-negative) colonies becoming old rose to pink-red upon exposure to ammonia fumes for 30 seconds are considered to be presumptive *Clostridium perfringens*. Color differentiation on m-CP Agar Base is sometimes difficult, so typical colonies (yellow turning into pink) as well as atypical colonies (green or those that remained yellow upon exposure to ammonia fumes) are picked for confirmation. For further confirmation of *Clostridium perfringens* it is suggested to carry out following biochemical test [3]: Sulphite reduction, gram-reaction, sporulating rods, motility, reduction of nitrate, gelatin liquefaction and lactose fermentation.



Cultural characteristics after 24-48 hours incubation under anaerobic conditions at 44 °C.

Organisms (ATCC)	Growth	Color of colony	$\beta$ -D-glucosidase	acid phosphatase
<i>Clostridium perfringens</i> (12924)	+++	Yellow (sucrose positive)	-	+ (rose to pink-red upon exposure to ammonia fumes)
<i>Clostridium sordellii</i> (9714)	+++	dark blue (sucrose negative)	+	+ (rose to pink-red upon exposure to ammonia fumes)
<i>Clostridium bifermentans</i> (638)	-/+	blue (sucrose negative)	+	- (no change upon exposure to ammonia fumes)
<i>Enterococcus faecalis</i> (29212)	-/+	blue	+	- (no change upon exposure to ammonia fumes)
<i>Escherichia coli</i> (25922)	-	-	-	-
<i>Pseudomonas aeruginosa</i> (27853)	-	-	-	-
<i>Staphylococcus aureus</i> (25923)	-	-	-	-
<i>Bacillus subtilis</i> (6633)	-	-	-	-
<i>Salmonella serotype Typhi</i> (6539)	-	-	-	-

#### References:

1. R. Armon, P. Payment, Can. J. Microbiol., 34:78-79 (1988)
2. Directive of the Council of the European Union 98/83/EC
3. D.P. Sartory, M. Field, S.M. Curbishley, A.M. Pritchard, Lett. Appl. Microbiol., 27, 323-327 (1998)
4. J.W. Bisson, V.J. Cabelli, Appl. Environ. Microbiol. 37, 55-66 (1979)

#### Precautions and Disclaimer

This product is for R&D use only, not for drug, household, or other uses. Please consult the Material Safety Data Sheet for information regarding hazards and safe handling practices.

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