

TECHNICAL DATA SHEET

Article No. 9716

Clostridium Perfringens Supplement (MUP + Cycloserine Selective Supplement)

SPECIFICATION

Sterile selective supplement used for isolation and presumptive identification of *Clostridium perfringens* by using fluorogenic substrates.

COMPOSITION (G/VIAL)

D-Cycloserine.....0.100
4-Methylumbelliferyl phosphate..... 0.025

Reconstitute the original freeze-dried vial by adding:

Sterile Distilled Water.....6 ml

Each vial is sufficient to supplement 200 ml of TSC Agar (base) (Art. no. 8032).
10 vials with freeze-dried supplement per box.

DESCRIPTION

Clostridium Perfringens Selective Supplement is added to TSC Agar (Base) in order to obtain a final selective medium which has the advantage to simplify the counting of plates with high numbers of colonies because smaller colonies of *C. perfringens* are formed. Sodium metabisulfite and ferric ammonium citrate are used as an indicator of sulfite reduction made by *C. perfringens* that produce black colonies in TSC agar (44 ±1 °C). The addition of MUP (4-methylumbelliferyl phosphate) demonstrates the ability of *C. perfringens* using this fluorogenic substrate.

TECHNIQUE

Collect, dilute and prepare samples and volumes as required according to specifications, directives, official standard regulations and/or expected results. Reconstitute the vial with 6 ml of sterile diluent in aseptic conditions and add it to 200 ml of melted TSC Agar cooled to 50 °C. Do not overheat once supplemented.

Pour the complete medium into Petri dishes and inoculate by MF Methods. Incubate the plates in anaerobic atmosphere at 44 ±1 °C for 21 ±3 h.

After incubation, count all the colonies that have appeared onto the surface of MF. *C. perfringens* grows in black colonies, due to the iron sulfide precipitation and is fluorescence positive under UV light (365 nm).

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Amtsgericht Stuttgart / HRB-Nr. 252035
Geschäftsführer: Lutz-Alexander Geyer / Thomas Roth

QUALITY CONTROL

- Physical/chemical control: Colour white grey
pH at 25 °C
- Microbiological control: Reconstitute 1 vial as indicated, shake and dissolve completely.
Distribute the complete medium, cooled at 50 °C, in filtration plates.
Anaerobiosis. Incubation at 44 ±1.0 °C, 20-24 h.

Microorganism	Growth	Remarks
<i>Clostridium perfringens</i> ATCC® 13124	Good - black colonies, fluorescent	None
<i>Clostridium perfringens</i> ATCC® 10543	Good - black colonies, fluorescent	None
<i>Bacillus subtilis</i> ATCC® 6633	Inhibited	None
<i>Clostridium sporogenes</i> ATCC® 11437	Good – no fluorescence	None

- Sterility control: Incubation 48 h at 30-35 °C and 48 h at 20-25 °C: No growth.
Check at 7 days after incubation at the same conditions.

REFERENCES

- ADCOCK, P.W. & C.P. SAINT (2001) Rapid Confirmation of *Clostridium perfringens* by Using Chromogenic and Fluorogenic Substrates. *App. Environm. Microbiol.* 67(9):4382-4384.
- ARAUJO, M. R.A. SUEIRO, M.J. GÓMEZ & M.J. GARRIDO (2001) Evaluation of fluorogenic TSC agar for recovering *Clostridium perfringens* in groundwater samples. *Water Sci. Technol.* 43:201-204.
- ARAUJO, M. R.A. SUEIRO, M.J. GÓMEZ & M.J. GARRIDO (2004) Enumeration of *Clostridium perfringens* spores in groundwater samples: comparison of six culture media. *J. Microbiol. Methods* 57:175-180.
- SARTORY, D.P., R. WALDOCK, C.E. DAVIES & A.M. FIELD (2006) Evaluation of acid phosphatase as confirmation test for *Clostridium perfringens* isolated from water. *Letter App. Microbiol.* 42:418-424.
- ISO 7937:2004. Microbiology of Food and Animal Feeding Stuffs. Horizontal Method for Enumeration of *C. perfringens*. Colony-count technique.
- ISO 6461-2 : 1986 Water Quality.- Detection and enumeration of the spores of sulfite-reducing anaerobes (Clostridia).- Part 2: Method by Membrane Filtration.
- ISO 11133:2014. Microbiology of food, animal feed and water. Preparation, production, storage and performance testing of culture media.
- ISO 14189:2013. Water quality. Enumeration of *Clostridium perfringens* — Method using membrane filtration.

STORAGE

2-25 °C

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SHELF LIFE

49 months from date of production.

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