Perfringens Agar Base (T.S.C.)

M837I

Intended use

Recommended for the enumeration of *Clostridium perfringens* from food .The composition and performance criteria of this medium are as per the specifications laid down in ISO 7937:1985.

Composition**

Ingredients	Gms / Litre
Tryptose	15.000
Soya peptone	5.000
Yeast extract	5.000
Sodium metabisulphite	1.000
Ferric ammonium citrate	1.000
Agar	15.000
Final pH (at 25°C)	7.6±0.2

**Formula adjusted, standardized to suit performance parameters

Directions

Suspend 21 grams in 500 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 50°C and add rehydrated contents of one vial of TSC Supplement (FD014). Alternatively if fluorogenic detection is desired add rehydrated contents of Clostridium perfringens supplements (FD243). When used for membrane filtration methods do not use FD014. Mix well before pouring into sterile Petri plates.

Principle And Interpretation

Tryptose Sulphite Cycloserine Agar (TSC) was originally formulated by Harmon et al (1) for the enumeration of *C.perfringens* from food. TSC Agar has been documented as one of the most useful media for the quantitative recovery of *C. perfringens* while suppressing growth of other facultative anaerobes (2). Perfringens Agar Base is also recommended by APHA (3). Perfringens Agar Base (M837I) is recommended for enumeration of *C.perfringens* from foods by ISO Committee (4) .

Tryptose, soya peptone, yeast extract, provide nitrogenous and carbonaceous compounds, long chain amino acids, vitamin B complex and trace elements essential for clostridial growth. Sodium metabisulphite and ferric ammonium citrate act as an indicator of sulphite reduction, indicated by black coloured colonies. D-cycloserine (FD014) help in the selective isolation of *C.perfringens* by inhibiting accompanying flora. Homogenized food samples can be directly streaked on the surface of plates or can be pre-enriched in Cooked Meat Medium (M149) before streaking.

Type of specimen

Food and animal feed samples.

Specimen Collection and Handling:

For food and animal feed samples, follow appropriate techniques for sample collection and processing as per guidelines (4). After use, contaminated materials must be sterilized by autoclaving before discarding.

Warning and Precautions :

Read the label before opening the container. Wear protective gloves/protective clothing/eye protection/ face protection. Follow good microbiological lab practices while handling specimens and culture. Standard precautions as per established guidelines should be followed while handling specimens. Safety guidelines may be referred in individualsafety data sheets

Limitations :

1. Some species of Clostridia may show poor growth. Preenrichment may be required.

Performance and Evaluation

Performance of the medium is expected when used as per the directions on the label within the expiry period when stored at recommended temperature.

Quality Control

Appearance
Light yellow to brownish yellow homogeneous free flowing powder

Gelling
Firm, comparable with 1.5% Agar gel

Colour and Clarity of prepared medium
Amber coloured clear to slightly opalescent gel.

Reaction
Reaction of 4.2% w/v aqueous solution at 25°C. pH : 7.6±0.2

pH
7.40-7.80

Cultural Response
Cultural characteristics observed under anaerobic condition with added TSC Supplement (FD014) and Egg Yolk Emulsion (FD045), after an incubation at 35-37°C for 18-24 hours.

Cultural Response

Organism	Inoculum (CFU)	Growth	Recovery	Sulphite Reduction	Fluorescence
Cultural Response					
Clostridium perfringensATCC 12924	50-100	luxuriant	>=50%	positive, blackening of medium	Positive Reaction
Clostridium sordellii ATCC 9714	>=10^3	inhibited	0%		

Storage and Shelf Life

Store between 10-30°C in a tightly closed container and the prepared medium at 2-8°C. Use before expiry date on the label. On opening, product should be properly stored dry, after tightly capping the bottle inorder to prevent lump formation due to the hygroscopic nature of the product. Improper storage of the product may lead to lump formation. Store in dry ventilated area protected from extremes of temperature and sources of ignition Seal the container tightly after use. Use before expiry date on the label.
Product performance is best if used within stated expiry period.

Disposal

User must ensure safe disposal by autoclaving and/or incineration of used or unusable preparations of this product. Follow established laboratory procedures in disposing of infectious materials and material that comes into contact with sample must be decontaminated and disposed of in accordance with current laboratory techniques (5,6).

Reference

1.Harmon S. M., Kauttar D.A. and Peiler J. T., 1971, Appl. Microbiol., 22:688.
2.Harmon S. M. and Kautter D.A., 1987, J. Asso. Off. Anal. Chem., 70: 994.
3.Downes F. P. and Ito K., (Eds.), 2001, Compendium of Methods for the Microbiological Examination of Foods, 4th Ed., American Public Health Association, Washington, D.C.
4.International Organization for Standardization (ISO- 7937:2004) : Microbiology of food and animal feeding stuffs- Horizontal method for the enumeration of Clostridium perfringens- Colony count technique
5.Isenberg, H.D. Clinical Microbiology Procedures Handbook. 2nd Edition.
6.Jorgensen,J.H., Pfaller , M.A., Carroll, K.C., Funke, G., Landry, M.L., Richter, S.S and Warnock., D.W. (2015) Manual of Clinical Microbiology, 11th Edition. Vol. 1.

Revision : 03 / 2018