

TM 2400 – TRYPTONE YEAST SODIUM SULPHITE AGAR BASE (ISO 14189:2013)

INTENDED USE

Recommended for the enumeration of Clostridium perfringens from water.

PRODUCT SUMMARY AND EXPLANATION

Tryptose Sulphite Cycloserine Agar (TSC) was originally formulated by Harmon et al 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. Perfringens Agar Base is also recommended by APHA. Tryptone Yeast Sodium sulphite Agar Base has been recommended by the ISO Committee for the isolation of *C. perfringens* from water samples using membrane filtration technique.

D-Cycloserine help in the selective isolation of *C.perfringens* by inhibiting accompanying flora. The water sample to be tested is filtered through 0.45 micron filter membrane and the membrane filter is then placed on Tryptone Yeast Sodium sulphite Agar Base and incubated anaerobically at 43-45°C for 18-24 hours. Sulfite reacts with ferric salt to produce sulfide which results in production of black or grey to yellow brown colonies. Confirmatory test: Smear some growth of 24 hours old culture of *Clostridium perfringens* from Blood Agar /Columbia Agar Base/ Tryptone Soya Agar (incubated anaerobically at 34-38°C) on the filter paper. Add 2-3 drops of Acid phosphatase Reagent on to the colonies of filter paper, observe for appearance of strong purplish colour developed within 3-4 min which is positive reaction for *Clostridium perfringens*.

COMPOSITION

Ingredients	Gms / Ltr		
Tryptone	15.000		
Soya peptone	5.000		
Yeast extract	5.000		
Sodium metabisulphite	1.000		
Ferric ammonium citrate	1.000		
Agar	15.000		

PRINCIPLE

Tryptone, Soya peptone and yeast extract provide nitrogenous compounds, carbon, long chain amino acids, sulphur, 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.

INSTRUCTION FOR USE

- Dissolve 21.0 grams in 500 ml purified / distilled water.
- Heat to boiling to dissolve the medium completely.
- Sterilize by autoclaving at 15 psi pressure (121°C) for 15 minutes. Cool to 45-50°C.
- Aseptically add the rehydrated contents of one vial of TSC Supplement.
- Mix well and pour into sterile Petri plates.

QUALITY CONTROL SPECIFICATIONS













Appearance of Powder : Light yellow to brownish yellow homogeneous free flowing powder. **Appearance of prepared medium** : Amber coloured clear to slightly opalescent gel forms in Petri plates.

pH (at 25°C) : 7.6±0.2

INTERPRETATION

Cultural characteristics observed after incubation under anaerobic condition with added TSC Supplement.

Microorganis m	ATCC	Inoculu m (CFU/ml)	Growth	Recove ry	Sulphite reduction	Acid phosphatas e test	Incubation Temperatur e	Incubati on Period
Clostridium perfringens	1292 4	50-100	Luxurian t	>=70%	Positive, blackening of medium	Positive reaction	43-45°C	18-24 Hours
Clostridium perfringens	1312 4	50-100	Luxurian t	>=70%	Positive, blackening of medium	Positive reaction	43-45°C	18-24 Hours

PACKAGING:

In pack size of 500 gm bottles.

STORAGE

Dehydrated powder, hygroscopic in nature, store in a dry place, in tightly-sealed containers between 25-30°C and protect from direct sunlight. Under optimal conditions, the medium has a shelf life of 4 years. When the container is opened for the first time, note the time and date on the label space provided on the container. After the desired amount of medium has been taken out replace the cap tightly to protect from hydration.

Product Deterioration: Do not use if they show evidence of microbial contamination, discoloration, drying or any other signs of deterioration.

DISPOSAL

After use, prepared plates, specimen/sample containers and other contaminated materials must be sterilized before discarding.

REFERENCES

- 1. Baird R.B., Eaton A.D., and Rice E.W., (Eds.), 2015, Standard Methods for the Examination of Water and Wastewater, 23rd ed., APHA, Washington, D.C.
- 2. Harmon S. M., Kauttar D.A. and Peiler J. T., 1971, Appl. Microbiol., 22:688.
- 3. Harmon S. M. and Kautter D.A., 1987, J. Asso. Off. Anal. Chem., 70: 994.
- 4. International Organization for Standardization (ISO- 14189:2013) Water quality Enumeration of Clostridium perfringens Method using membrane filtration
- 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.





































NOTE: Please consult the Material Safety Data Sheet for information regarding hazards and safe handling Practices. *For Lab Use Only Revision: 08 Nov., 2019







