Perfringens Agar Base (TSC)

(Tryptose Sulphite Cycloserine (TSC) Agar)

LAB 194

Description
Perfringens Agar Base is a nutrient medium to which egg yolk emulsion (X073) and cycloserine (X194) are added for the preparation of Tryptose Sulphite Cycloserine (TSC) Agar. Sodium metabisulphite and ferric ammonium citrate are used as an indicator of sulphite reduction by *Clostridium perfringens*. The reduction of sulphite by *Cl. perfringens* produces black colonies and the egg yolk emulsion incorporated into the media detects the lecithinase activity of this bacteria. However not all strains produce lecithinase and therefore black lecithinase positive and black lecithinase negative colonies should be considered as presumptive *Cl. perfringens*.

Formula	g/litre
Tryptose	15.0
Soy Peptone	5.0
Beef extract	5.0
Yeast extract	5.0
Sodium metabisulphite	1.0
Ferric ammonium citrate	1.0
Agar	14.0

Method for reconstitution
Weigh 46.0 grams of powder and disperse in 1 litre of deionised water. Allow the mixture to soak for 10 minutes, swirl to mix and sterilise by autoclaving at 121°C for 10 minutes. Allow the medium to cool to 47°C and supplement with 2 vials of X194 (cycloserine) and 50ml of egg yolk emulsion (X073), mix well and pour into sterile Petri dishes. The egg yolk emulsion is omitted for preparation of Egg Yolk Free TSC Agar and Egg Yolk Free TSC Agar should be used for an overlay medium.

Appearance: Straw, clear gel or pale yellow opaque gel.

pH: 7.6 ± 0.2

Minimum Q.C. organisms: *Clostridium perfringens* NCIMB 50027
Escherichia coli NCIMB 50034 (inhibition)

Storage of Prepared Medium: Plates can be stored up to 7 days at 2-8°C in the dark.

Inoculation: For a spread plate inoculate the agar plate with 0.1ml aliquots of an appropriate serial dilution of the homogenised test sample and overlay if required. For a pour plate mix 1ml aliquots of an appropriate serial dilution of the homogenised test sample with approximately 20 ml of TSC plus egg yolk emulsion. For full details refer to appropriate references and standard method protocols.

Incubation: 35°C ± 2°C anaerobically for 18-24 hours.

Interpretation: Count all black colonies with or without a halo as presumptive *C. perfringens*. Further confirmation should be carried out according to standard method protocols e.g. nitrate reduction, lactose fermentation, gelatin liquefaction and absence of motility.

References

Shahidi, S.A. and Furguson, A.R. (1971). Appl. Microbiol. 21. 500-506.

Harmon, S.M., Kauttar, D.A. and Peeler, J.T. (1971). Appl. Microbiol. 22. 688-692.

Hauschild, A. H. W. and Hilsheimar R. (1973). Appl. Microbiol. 27. 78-82.

Hauschild A.H.W. and Hilsheimar, R. (1973). Appl. Microbiol. 27. 521-526.

Hauschild, A.H.W. *et al* (1977). Can. J. Microbiol. 23. 884-892.

Labbe, R. G. and Harmon, S.M. (1992). Compendium of methods for the microbiological examination of foods, 3rd ed 623-635. American Public Health Association, Washington, D.C.

Rhodehamel, E.J. and Harmon, S.M. (1995). Bacteriological Analytical Manual 8th ed. 16.01-16.06 AOAC International, Gaithersberg, MD.

Andrews, W. (1995) Official methods of analysis AOAC International 16th ed. 1-119. AOAC International, Arlington, VA.