Tryptose Cycloserine Azide Agar Base

M1279

Tryptose Cycloserine Azide Agar Base is recommended for enumeration of sulphite reducing anaerobes essentially Clostridia .

Composition**

Ingredients	Gms / Litre
Tryptose	15.000
Papaic digest of soyabean meal	5.000
Meat extract	5.000
Yeast extract	5.000
Glucose	2.000
Disodium disulphite	0.500
Ferric ammonium citrate	0.500
Sodium azide	0.050
Agar	14.000
Final pH (at 25°C)	7.4±0.2

**Formula adjusted, standardized to suit performance parameters

Directions

Suspend 23.52 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 aseptically add 1.5 ml rehydrated contents of 1 vial of T.S.C. Supplement (FD014) for 500 ml medium. Mix well and pour into sterile Petri plates.

Warning : Sodium azide has a tendency to form explosive metal azides with plumbing materials. It is advisable to use enough water to flush off the disposables.

Principle And Interpretation

Tryptose Cycloserine Azide Agar Base was originally formulated by Hauschild and Hilsheimer (1). This medium was later modified by decreasing the concentration of D-cycloserine, sulphite and iron and by the addition of sodium azide (2, 3). This medium utilizes the selective inhibitory properties of D-cycloserine and an indicator system involving sulphite and iron. Growth of non-mesophillic organisms are suppressed while *C. perfringens* and related species will reduces the sulphite and form black colonies due to the production of ferrous sulphide (4).

Tryptose, papaic digest of soyabean meal, meat extract and yeast extract provide essential nitrogenous compounds and vitamins needed for the growth of anaerobes. Glucose serves as carbon source. Disodium disulphite is reduced to hydrogen sulphide which combines with ferric ions of ferric salts to produce the insoluble black precipitate of ferrous sulphide. D-Cycloserine (FD014) and sodium azide inhibit a number of organisms including *Bacillus* species, enteric bacilli, *Proteus* , *Pseudomonas* and most of the cocci. Some anaerobes reduce sulphite to hydrogen sulphide (H2S) which is indicated by blackening of the colonies due to presence of ferric ammonium citrate.

Quality Control

Appearance
Cream to brownish yellow homogeneous free flowing powder

Gelling
Firm, comparable with 1.4% Agar gel.

Colour and Clarity of prepared medium
Yellow to amber coloured clear to slightly opalescent gel forms in Petri plates.

Reaction
Reaction of 4.7% w/v aqueous solution at 25°C. pH : 7.4±0.2

pH

7.20-7.60

Cultural Response

M1279: Cultural response observed after an incubation at 35-37°C for 18-24 hours with added T.S.C. Supplement (FD014).

Organism	Inoculum (CFU)	Growth	Recovery	Colour of colony
Clostridium perfringens ATCC 12924	50-100	good	>=50%	black
Clostridium sporogenes ATCC 11437	50-100	good	>=50%	black
Escherichia coli ATCC 25922	>=10^3	inhibited	0%	
Staphylococcus aureus ATCC 25923	>=10^3	inhibited	0%	

Storage and Shelf Life

Store below 30°C in tightly closed container and the prepared medium at 2-8°C.Use before expiry date on the label.

Reference

1. Hauschild A. H. W. and Hilsheimer R., 1974, Appl. Microbiol., 27, 521-527.
2. Eisgruber H., 1986, Vet Med. Diss. FU Berlin.
3. Eisgruber H. and Reuter G., 1991, Arch. Lebensmittelhyg, 42,125-129.
4. Corry J. E. L., Curtis G. D. W. and Baird R. M., 1995, Culture Media for Food Microbiology, Vol. 34, ELSEVIER, Amsterdam.

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