DNase Test Agar Base w/ methyl green

M1419

DNase Test Agar is recommended for the detection of deoxyribonuclease activity of bacteria and fungi, and especially for identification of pathogenic Staphylococci.

Composition**

Ingredients	Gms / Litre
Tryptose	20.000
Deoxyribonucleic acid (DNA)	2.000
Sodium chloride	5.000
Methyl green	0.050
Agar	15.000
Final pH (at 25°C)	7.3±0.2

**Formula adjusted, standardized to suit performance parameters

Directions

Suspend 42.05 grams in 1000 ml distilled water. Heat to boiling to dissolve the medium completely. Sterilize by autoclaving at 15 lbs pressure (121°C) for 15 minutes. Mix well and pour into sterile Petri plates.

Principle And Interpretation

DNase test Agar is used for detecting deoxyribonuclease activity of bacteria and fungi and particularly for identification of pathogenic Staphylococci (1). DNase producing organisms exhibit clear zone around growth against green background. Reagent addition is not required (2). This medium is based on modification of the procedure for detecting DNase-producing bacteria as per Smith, Hanoch, and Rhoden (4) and Jefferies, Holtman and Guse (3). The medium supports growth of both gram positive and gram-negative bacteria.

Tryptose serves as nitrogenous source for the organisms. DNase produced by microorganisms depolymerizes the DNA substrate in the medium. Methyl green fades into a colourless compound producing distinct clear zones surrounding colonies (or band/ spot inocula) in an otherwise green coloured medium. Methyl green requires a highly polymerized DNA substrate (5) and it combines with polymerized DNA forming a stable, green complex at pH 7.5 (6,7,8). As hydrolysis progresses, methyl green is released and when not combined at this pH it fades and becomes a colourless compound. Therefore clear zones are observed (7,9).

Quality Control

Appearance
Light yellow to greeenish yellow homogeneous free flowing powder

Gelling
Firm, comparable with 1.5% Agar gel

Colour and Clarity of prepared medium
Green coloured, clear to slightly opalescent gel forms in Petri plates

Reaction
Reaction of 4.2% w/v aqueous solution at 25°C. pH : 7.3±0.2

pH
7.10-7.50

Cultural Response
M1419: Cultural characteristics observed after an incubation at 35-37°C for 18-24 hours

Organism	Inoculum (CFU)	Growth	DNase Activity

Serratia marcescens ATCC 8100	50-100	luxuriant	positive, clear halo around the growth.
Staphylococcus aureus ATCC 25923	50-100	luxuriant	positive, clear halo around the growth.
Staphylococcus epidermidis ATCC 12228	50-100	luxuriant	negative reaction
Streptococcus pyogenes ATCC 19615	50-100	luxuriant	positive, clear halo around the growth.

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

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