Semisolid Nutrient Agar

M1191

Semisolid Nutrient Agar is recommended for detection of *Salmonella* species on the basis of motility and hydrogen sulphide (H2S) production.

Composition**

Ingredients	Gms / Litre
Peptic digest of animal tissue	5.000
Beef extract	3.000
Agar	4.000
Final pH (at 25°C)	7.0±0.2

**Formula adjusted, standardized to suit performance parameters

Directions

Suspend 12 grams in 1000 ml distilled water. Heat to boiling to dissolve the medium completely. Dispense in tubes. Sterilize by autoclaving at 15 lbs pressure (121°C) for 15 minutes. Allow the tubes to cool in an upright position.

Principle And Interpretation

Nutrient Agar is a basic culture medium used in water and food studies. It is generally used for maintenance purpose or to check the purity of subcultures (1). Bacterial motility is an important determinant in making a final species identification. Tubes containing semisolid media are most often used to determine motility. Motile organisms form a diffuse zone of growth flaring out from the line of inoculation. Non-motile organisms grow along the stabline. Certain bacterial species have ability to liberate sulphur from sulphur-containing amino acids or other compounds in the form of H2S (hydrogen sulphide). Lead acetate paper strips serve as the H2S indicators (2). Semisolid Nutrient Agar couples these two tests in a single medium. It is also recommended by ISO Committee (3) for the detection of *Salmonella* species.

Peptic digest of animal tissue and beef extract provide essential growth nutrients. The motile cultures grow away from stabline while non-motile grow along the stabline. After inoculation, with the test organism, insert a lead acetate paper strip (DD034) between the plug and inner wall of tube. Lead acetate strip incorporation helps to detect H2S production.

Quality Control

Appearance
Cream to yellow homogeneous free flowing powder

Gelling
Semisolid, comparable with 0.4% Agar gel.

Colour and Clarity of prepared medium
Light yellow coloured clear gel forms in tubes as butts

Reaction
Reaction of 1.2% w/v aqueous solution at 25°C. pH : 7.0±0.2

pH
6.80-7.20

Cultural Response
M1191: Cultural characteristics observed after an incubation at 35-37°C for 18-24 hours.

Organism	Inoculum (CFU)	Growth	Motility	H2S (with lead acetate strip)
Cultural Response				
Escherichia coli ATCC 25922	50-100	luxuriant	positive, growth away from stabline causing turbidity	negative reaction, no blackening

Salmonella Typhi ATCC 6539	50-100	luxuriant	positive, growth away from stabline causing turbidity	positive reaction, blackening of the lower portion of the strip
Salmonella Enteritidis ATCC 13076	50-100	luxuriant	positive, growth away from stabline causing turbidity	positive reaction, blackening of the lower portion of the strip

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. Lapage S. P., Shelton J. E. and Mitchell T. G., 1970, Methods in Microbiology, Norris J. R. and Ribbons D. W., (Eds.), Vol. 3A, Academic Press, London.
2. Koneman E. W., Allen S. D., Janda W. M., Schreckenberger P. C., Winn W. C. Jr., 1992, Colour Atlas and Textbook of Diagnostic Microbiology, 4th Ed., J. B. Lippinccott Company.
3. International Organization for Standardization (ISO), 1993, Draft ISO/DIS 6579.

Revision : 02 / 2015