Tryptone Soya-Tryptose Broth

M1876

Intended Use:

Recommended for identification of *Salmonella* species from food samples in accordance with FDA BAM, 1998.

Composition**

Ingredients	Gms / Litre
Tryptone	8.500
Soya peptone	1.500
Sodium chloride	5.100
Dextrose (Glucose)	1.770
Dipotassium hydrogen phosphate	1.250
Tryptose	10.380
Yeast extract	3.000
Final pH (at 25°C)	7.2±0.2

**Formula adjusted, standardized to suit performance parameters

Directions

Suspend 31.50 grams in 1000 ml purified / distilled water. Heat if necessary to dissolve the medium completely. Dispense 5ml portions into 16×150mm test tube. Sterilize by autoclaving at 15 lbs pressure (121°C) for 15 minutes.

Principle And Interpretation

Salmonella is a genus of rod-shaped, Gram-negative, non-spore-forming, predominantly motile enterobacteria with peritrichous flagella. Most of the species are pathogenic, and the infections are mainly due to the ingestion of contaminated food (5). Tryptone Soya Tryptose Broth is used for serological identification of *Salmonella* species with respect to 'polyvalent flagellar (H) test' in accordance with FDA BAM, 1998 (1).

Add 25g of the food sample(s) suspected to be contaminated with *Salmonella* into 225ml culture broth (1:9 ratio) and incubate at 35 ± 2.0º C for 24 ± 2.0 hours in accordance with the BAM protocol. The incubated sample is processed for isolation of the species by inoculation into selective media such as Selenite broth (M052), Fluid Tetrathionate Medium w/o Iodine and BG, Modified (M032F) or Rappaport Vassiliadis Medium, Modified (M880F) and incubation for 24hrs at appropriate temperatures. Thoroughly mix and streak a 3 mm loopful of the incubated broth on Bismuth Sulphite Agar (M027), XLD agar (M031F), and Hektoen Enteric Agar, w/ 1.2% agar (M467F). Organism is identified by its colony characteristics in respective media. The organism can be confirmed through biochemical and serological tests. Serological tests include identification of polyvalent flagellar (H) antigen. Tryptone soya tryptose broth (M1876) is used for the initial inoculum preparation for this test (1,2).

Tryptone, Tryptose, Soya peptone and Yeast extract provide necessary carbon, nitrogen compounds, long chain amino acids, vitamins, and other trace mineral sources for the growth of microrganisms. Dextrose provide necessary carbon source to the medium. Sodium chloride maintains the osmotic equilibrium of the medium. Dipotassium hydrogen phosphate acts as the buffering agent.

Type of specimen

Food samples

Specimen Collection and Handling:

For food samples, follow appropriate techniques for sample collection and processing as per guidelines (6). After use, contaminated materials must be sterilized by autoclaving before discarding.

Warning and Precautions

Read the label before opening the container. Wear protective gloves/protective clothing/eye protection/ face protection. Follow good microbiological lab practices while handling specimens and culture. Standard precautions as per established guidelines should be followed while handling specimens. Safety guidelines may be referred in individual safety data sheets.

Limitations

1. Further biochemical and serological tests must be carried out for complete identification.

Performance and Evaluation

Performance of the medium is expected when used as per the direction on the label within the expiry period when stored at recommended temperature.

Quality Control

Appearance

Cream to yellow homogeneous free flowing powder

Colour and Clarity of prepared medium

Light yellow coloured clear solution without any precipitate.

Reaction

pH of 3.15% w/v aqueous solution at 25°C . pH : 7.2±0.2

pH

7.00-7.40

Cultural Response

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

Organism	Inoculum (CFU)	Growth
Staphylococcus aureus subsp. *aureus ATCC 25923* (00034*)	50 -100	luxuriant
Escherichia coli ATCC 8739 (00012*)	50 -100	luxuriant
Escherichia coli ATCC 25922 (00013*)	50 -100	luxuriant
Pseudomonas aeruginosa ATCC 27853 (00025*)	50 -100	luxuriant
Salmonella Typhi *ATCC 6539*	50 -100	luxuriant
Salmonella Enteritidis *ATCC 13076* (00030*)	50 -100	luxuriant
Salmonella Typhimurium *ATCC 14028* (00031*)	50 -100	luxuriant
Salmonella Abony *NCTC 6017* (00029*)	50 -100	luxuriant

Key : (*) Corresponding WDCM numbers.

Storage and Shelf Life

Store between 10-30°C in a tightly closed container and the prepared medium at 15-25°C. Use before expiry date on the label. On opening, product should be properly stored dry, after tightly capping the bottle in order to prevent lump formation due to the hygroscopic nature of the product. Improper storage of the product may lead to lump formation. Store in dry ventilated area protected from extremes of temperature and sources of ignition. Seal the container tightly after use. Product performance is best if used within stated expiry period.

Disposal

User must ensure safe disposal by autoclaving and/or incineration of used or unusable preparations of this product. Follow established laboratory procedures in disposing of infectious materials and material that comes into contact with sample must be decontaminated and disposed of in accordance with current laboratory techniques (3,4).

Reference

1. FDA, U.S. 1998. Bacteriological Analytical Manual. 8 ed. Gaithersburg, MD: AOAC International.
2. Forbes, B. A., Sahm, D. F . and Weissfield, A. S. 2002. Bailey and Scott's Diagnostic Microbiology. 11 ed. St Louis: The C.V. Mosby Co.
3. Isenberg, H.D. Clinical Microbiology Procedures Handbook 2nd Edition.
4. 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.
5. MacFaddin, J. F. 1985. Media for Isolation-Cultivation-Identification-Maintenance of Medical Bacteria vol. 1. Baltimore: Williams and Wilkins.
6. Salfinger Y., and Tortorello M.L. Fifth (Ed.), 2015, Compendium of Methods for the Microbiological Examination of Foods, 5th Ed., American Public Health Association, Washington, D.C.

Revision : 01 / 2019