



Salt Polymyxin Broth Base

M821

Intended use

Used for detection and enumeration of *Vibrio* species.

Composition**

Ingredients	g / L
Tryptone	10.000
Yeast extract	3.000
Sodium chloride	20.000
Final pH (at 25°C)	8.8±0.2

**Formula adjusted, standardized to suit performance parameters

Directions

Suspend 16.5 grams in 500 ml purified/distilled water. Heat if necessary to dissolve the medium completely. Sterilize by autoclaving at 15 lbs pressure (121°C) for 15 minutes. Cool to 45-50°C and aseptically add rehydrated contents of one vial of PolyB Selective Supplement (FD003). Mix well and dispense into sterile tubes or flasks as desired.

Principle And Interpretation

Vibrios are fairly easy to isolate from both clinical and environmental material, though some species may require growth factors and /or vitamins. Salt Polymyxin Broth is formulated as per the recommendation of APHA (1). Tryptone and yeast extract provide nitrogenous compounds, carbon, sulphur, trace elements, long chain amino acids and vitamin B complex, essential for the growth. Polymyxin B sulphate inhibits gram-positive organisms.

Type of specimen

Clinical sample : faeces; Food samples; Water samples.

Specimen Collection and Handling:

Weigh 50 grams of sample into a blender. Add 450 ml phosphate buffer saline dilution water and blend for 1 minute at 8000 rpm. This constitutes 1:10 dilution. Prepare 1:100, 1:1000, 1:10000 dilutions or higher, if necessary, in PCB. Inoculate 3 x 10 ml portion of the 1 : 10 dilutions into 3 tubes containing 10 ml of enrichment broth i.e. Salt Polymyxin Broth Base-2x concentration. This represents the 1 gram portion. Similarly inoculate 3 x 1 ml of dilutions into 10 ml of single strength Salt Polymyxin Broth Base. Incubate tubes at 35 ± 2°C for 24 hours.

After incubation a loopful is subcultured on solid medium such as TCBS Agar (M189) for further studies. *V. parahaemolyticus* appears as round, green or bluish colonies, 2-3 mm in diameter while *V. cholerae* forms yellow coloured colonies. After use, contaminated materials must be sterilized by autoclaving before discarding.

Warning and Precautions

In Vitro diagnostic use. For professional use only. 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 clinical specimens. Safety guidelines may be referred in individual safety data sheets.

Limitations

1. Subculturing on solid media is required for further studies.
2. Further serological and biochemical testing is required 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

Reaction of 3.3% w/v aqueous solution at 25°C. pH : 8.8±0.2

pH

8.60-9.00

Cultural Response

Cultural characteristics observed after an incubation at 35-37°C for 24-48 hours with added PolyB Selective Supplement (FD003).

Organism	Inoculum (CFU)	Growth
<i>Vibrio cholerae</i> ATCC 14035	50-100	luxuriant
<i>Vibrio parahaemolyticus</i> ATCC 17802	50-100	luxuriant

Storage and Shelf Life

Store between 10-30°C in a tightly closed container and the prepared medium at 15-30°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 clinical sample must be decontaminated and disposed of in accordance with current laboratory techniques (2,3).

Reference

1. 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.
2. Isenberg, H.D. Clinical Microbiology Procedures Handbook 2nd Edition.
3. 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.

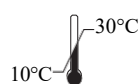
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