



Salt Polymyxin Broth Base

M821

Salt Polymyxin Broth Base is recommended for detection and enumeration of *Vibrio* species.

Composition**

Ingredients	Gms / Litre
Casein enzymic hydrolysate	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 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 1 vial of Polymyxin B Selective Supplement (FD003). Mix well and dispense 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).

Casein enzymic hydrolysate and yeast extract provide nitrogenous compounds, carbon, sulphur, trace elements and vitamin B complex, essential for the growth. Polymyxin B sulphate inhibits gram-positive organisms.

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.

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

M821: Cultural characteristics observed after an incubation at 35-37°C for 24-48 hours with added Polymyxin B 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 below 30°C in tightly closed container and the prepared medium at 2 - 8°C. Use before expiry date on the label.

Reference

Downes F. P. and Ito K., (Eds.), 2001, Compendium of Methods for the Microbiological Examination of Foods, 4th Ed., APHA, Washington, D.C.

Revision : 1 / 2011



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