Glucose Salt Teepol Broth (Twin Pack)

M621

Intended Use:

Recommended for enrichment of *Vibrio parahaemolyticus* and marine isolates from food.

Composition**

Ingredients	Gms / Litre
Part A	-
Peptone	10.000
HM peptone B #	3.000
Sodium chloride	30.000
Dextrose (Glucose)	5.000
Methyl violet	0.002
Part B	-
Teepol	4.000
Final pH (at 25°C)	8.8±0.2

**Formula adjusted, standardized to suit performance parameters
Equivalent to Beef extract

Directions

Suspend 48.0 grams of Part A in 1000 ml purified/distilled water containing 4.0 ml of Part B. Heat gently to dissolve the medium completely. Dispense in tubes as desired and sterilize by autoclaving at 15 lbs pressure (121°C) for 15 minutes. Cool to 45-50°C.

Principle And Interpretation

Glucose Salt Teepol Broth is a special media used to enrich *Vibrio parahaemolyticus* from sea foods (1) and also used to enumerate the bacteria by MPN technique (2).

V. parahaemolyticus is a gram-negative marine bacterium, which causes seafood-borne gastroenteritis in humans (3). Fujino and co-workers were the first to isolate *Vibrio parahaemolyticus* as a causative agent of food-borne gastroenteritis, following a large outbreak in Japan (4).

Peptone and HM peptone B provide essential nitrogenous nutrients and the high percentage of sodium chloride (3%) helps for the better enrichment of halophilic *V.parahaemolyticus*. Glucose is utilized while teepol inhibits the growth of gram-positive organisms. The test sample should be held under moderate refrigeration (about 7 to 10°C) and should be analyzed as soon as possible, after collection as possible. This maximizes the survival and recovery of *Vibrio's* and reduces the tendency for overgrowth by indigenous marine microflora.

Type of specimen

Clinical samples - Stool sample, wound swab; Food samples

Specimen Collection and Handling:

Weigh 50 gram of seafood sample into a blender. Add 450 ml of PBS (Phosphate Buffer Saline) dilution water and blend for 1 min at 8000 rpm. This constitutes the 1:10 dilution. Prepare 1:100, 1:1000, 1:10000 dilutions or higher if necessary in PBS. Inoculate 3 x 10 ml portion of the 1:10 dilution into 3 tubes containing 10 ml of enrichment broth i.e. Glucose Salt Teepol Broth in 2x concentration. This represents the 1-gram portion. Similarly inoculate 10 ml of single strength enrichment broth as above. If high numbers of *V.parahaemolyticus* are expected, the examination may start at the 1:10 dilution of the product (5). After overnight incubation of Glucose Salt Teepol Broth at 35 ± 2°C, a loopful of culture from top 1 cm of the broth showing growth is streaked onto TCBS Agar (M189). After overnight incubation at 35 ± 2°C, *V.parahaemolyticus* colonies on TCBS Agar appear as round, green or bluish measuring 2.3 mm in diameter, while *V.alginolyticus* colonies are larger and yellow coloured. These colonies are further identified by biochemical characterization.For biochemical tests in identification of *V.parahaemolyticus, V.cholera,* and *V.vulnificus,* appropriate positive control organisms have to be inoculated. When the blue green colonies are finally identified as *V.parahaemolyticus,* refer to the original positive dilution in the enrichment broth and apply the 3 tube MPN tables for final enumeration of the organism.

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. Further biochemical and serological tests must be carried out for further 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

Part A : Cream to yellow homogeneous free flowing powder Part B : Colourless viscous liquid

Colour and Clarity of prepared medium

Yellow coloured, clear solution with a very slight precipitate.

Reaction

Reaction of 4.8% w/v aqueous solution with 0.4% Teepol 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 18-24 hours .

Organism	Inoculum (CFU)	Growth
Vibrio alginolyticus ATCC 17749	50-100	good-luxuriant
Vibrio parahaemolyticus ATCC 17802 (00037*)	50-100	good-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 clinical sample must be decontaminated and disposed of in accordance with current laboratory techniques (3,4).

Reference

1.Akiyama S., Takizawa K., and Obara Y., 1964, Ann. Rep. Kanagawa Pref. Inst. Public Health, 13:7
2.Salfinger Y., and Tortorello M.L. Fifth (Ed.), 2015, Compendium of Methods for the Microbiological Examination of Foods, American Public Health Association, Washington, D.C.
3.Thompson F. L., T. Iida and Swings J., 2004, Biodiversity of Vibrios, Microbiol. Mol. Biol. Rev., 68: 403-431.
4.Fujino T., Okuno Y., Nakada D., Aoyama A., Fukai K., Mukai T. and Ueho T., 1953, Med. J. Osaka Univ., 4:299-304.
5.Isenberg, H.D. Clinical Microbiology Procedures Handbook 2nd Edition.
6.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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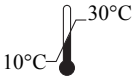
IVD	In vitro diagnostic medical device
CE	CE Marking
	Storage temperature
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