# Cholera Medium Base

M558

## Intended Use:

Recommended for selective isolation of *Vibrio* species from specimens heavily contaminated with *Enterobacteriaceae.*

## Composition**

| Ingredients | g / L |
|---|---|
| Peptone | 10.000 |
| HM peptone B # | 10.000 |
| Sucrose | 10.000 |
| Sodium lauryl sulphate | 0.100 |
| Sodium chloride | 20.000 |
| Sodium carbonate | 5.000 |
| Agar | 10.000 |
| Final pH ( at 25°C) | 8.5±0.2 |

**Formula adjusted, standardized to suit performance parameters

## Directions

Suspend 65.1 grams in 1000 ml purified/distilled water. Heat to boiling to dissolve the medium completely. **DO NOT AUTOCLAVE.** Cool to 70°C and add 2 ml of sterile 1% Potassium Tellurite Solution (FD052) and 5 ml of sterile defibrinated blood. Maintain at 70°C for a few minutes. Cool to 45-50°C. Mix well and pour into sterile Petri plates.

## Principle And Interpretation

*Vibrio cholerae* is the etiological agent of cholera in humans in which the disease is caused not by tissue invasion of microorganisms but through the production of toxins that interrupt normal intra-intestinal exchanges of water and electrolytes. *Vibrios* grow readily on most isolation media. Adding sodium chloride to the medium enhances growth of all species. Cholera Medium Base is a selective medium used for the isolation of *Vibrio* species from specimens contaminated with enteric bacteria. It is based on the formulation described by Felsenfeld and Watanabe (1) for the isolation of *V. cholerae* and similar *Vibrios* from specimens contaminated with *Enterobacteriaceae*.

Peptone and HM peptone B provide nitrogenous nutrients whereas sucrose serves as the fermentable carbohydrate source for the metabolism of *Vibrios*. Sodium lauryl sulphate inhibits many contaminating organisms. Potassium tellurite also inhibits many gram-positive and gram-negative bacteria except *Vibrios*. Sodium chloride maintains osmotic equilibrium.

## Type of specimen

Clinical samples- faeces; Food samples.

## Specimen Collection and Handling

For clinical samples follow appropriate techniques for handling specimens as per established guidelines (2,3).
For food samples, follow appropriate techniques for sample collection and processing as per guidelines (4).
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.Slight colour variation may be observed depending upon strains.
2.Further biochemical tests must be carried out for confirmation.

## 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

**Gelling**

Firm, comparable with 1.0% Agar gel.

**Colour and Clarity of prepared medium**

Basal medium: Yellow coloured clear to slightly opalescent gel. After Addition of blood & Tellurite and on heating : Brownish red coloured opaque gel forms in Petri plates.

**Reaction**

Reaction of 6.5% w/v aqueous solution at 25°C. pH : 8.5±0.2

**pH**

8.30-8.70

**Cultural Response**

Cultural characteristics observed with added 1% Potassium Tellurite Solution(FD052) and sterile defibrinated blood, after an incubation at 35-37°C for 18-24 hours.

| Organism | Inoculum (CFU) | Growth | Recovery |
|---|---|---|---|
| ** *Bacillus spizizenii* ATCC 6633 (00003*) | >=$10^4$ | inhibited | 0% |
| *Escherichia coli* ATCC 25922 (00013*) | >=$10^4$ | inhibited | 0% |
| *Proteus mirabilis* ATCC 25933 | >=$10^4$ | inhibited | 0% |
| *Pseudomonas aeruginosa* ATCC 27853 (00025*) | >=$10^4$ | inhibited | 0% |
| *Vibrio cholerae* ATCC 15748 | 50-100 | luxuriant | >=50% |
| *Vibrio parahaemolyticus* ATCC 17802 (00037*) | 50-100 | luxuriant | >=50% |

Key : *Corresponding WDCM numbers. **Formerly known as *Bacillus subtilis* subsp. *spizizenii*

## Storage and Shelf Life

Store between 10-30°C in a tightly closed container and the prepared medium at 2 - 8°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. Felsenfeld O. and Watanabe Y., 1958, U.S. Armed Forces Med. J., 9 (7) : 975.
2. Isenberg, H. 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.
4. Salfinger Y., and Tortorello M.L., 2015, Compendium of Methods for the Microbiological Examination of, Foods, 5th Ed., American Public Health Association, Washington, D.C.

Revision : 04 / 2024

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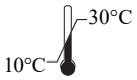
IVD ***In vitro* diagnostic medical device**




**Storage temperature**

EC REP CEpartner4U, Esdoornlaan 13,
3951DB Maarn, NL
www.cepartner4u.eu

**CE Marking**

**Do not use if package is damaged**

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