



HiCrome™ Vibrio Agar

M1682

Intended use

HiCrome™ Vibrio Agar is recommended for the isolation, and selective chromogenic differentiation of *Vibrio* species from seafood.

Composition**

Ingredients	Gms / Litre
Peptone	10.000
Sodium chloride	25.000
Sodium thiosulphate	5.000
Sodium citrate	6.000
Sodium cholate	1.000
Chromogenic mixture	5.500
Agar	15.000
Final pH (at 25°C)	8.5±0.2

**Formula adjusted, standardized to suit performance parameters

Directions

Suspend 67.5 grams in 1000 ml distilled water. Heat to boiling to dissolve the medium completely. DO NOT AUTOCLAVE. Cool to 45-50°C. Mix well before pouring into sterile Petri plates.

Principle And Interpretation

Vibrio's have played a significant role in human history. Outbreaks of cholera, caused by *Vibrio cholerae*, can be traced back in time to early recorded descriptions of enteric infections. The *Vibrio*'s have also received the attention of marine microbiologists who observed that the readily cultured bacterial population in near-shore waters and those associated with fish and shell fish were predominantly *Vibrio* species (1). *Vibrio* species are mainly responsible for causing cholera and food poisoning in humans. *Vibrio cholerae* causes cholera due to the intake of contaminated food such as raw oysters. *Vibrio parahaemolyticus* is a major cause of food borne infections, causing food poisoning (2). Since *Vibrio* species naturally occur in sea water, worth special mention is their need for sodium chloride, although some species can grow with minimum sodium chloride concentration (1). The widely used media for *Vibrio* isolation are TCBS Agar and Alkaline Peptone Water (3). However accompanying sucrose-fermenting bacteria pose a problem in the identification of *Vibrio* species on TCBS Agar. On HiCrome™ Vibrio Agar, the colour development by *Vibrio* species is not affected by the presence of colonies of other bacteria. This is because, the amount of colour developed depends on the reaction of the bacterial beta-galactosidase with the substrate contained in the media (4).

Peptone provides carbonaceous, nitrogenous and essential nutrients to the organisms. High concentration of sodium chloride in addition to maintaining the osmotic equilibrium also has an inhibitory action on the accompanying microflora. Sodium thiosulphate, sodium citrate and sodium cholate are used in the formulation because they can inhibit the growth of gram positive and some gram negative bacteria, but not members of *Enterobacteriaceae*. The proprietary chromogenic mixture incorporated in the medium helps in the chromogenic differentiation of *Vibrio cholerae* and *Vibrio parahaemolyticus*. The high (alkaline) pH of the medium helps in selective isolation of *Vibrio* species.

Type of specimen

Food samples

Specimen Collection and Handling

For food samples, follow appropriate techniques for sample collection and processing as per guidelines (5)

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

Not applicable

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

Light yellow to light tan homogeneous free flowing powder

Gelling

Firm, comparable with 1.5% Agar gel

Colour and Clarity of prepared medium

Light yellow coloured, clear to slightly opalescent gel forms in Petri plates

Reaction

Reaction of 6.75% w/v aqueous solution at 25°C. pH : 8.5±0.2

pH

8.30-8.70

Cultural Response

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

Cultural Response

Organism	Inoculum (CFU)	Growth	Recovery	Colour of colony
<i>Enterococcus faecalis</i> ATCC 29212 (00087*)	≥10 ³	inhibited	0%	
<i>Escherichia coli</i> ATCC 25922 (00013*)	≥10 ³	inhibited	0%	
<i>Staphylococcus aureus</i> ATCC 25923 (00034*)	≥10 ³	inhibited	0%	
<i>Vibrio cholerae</i> ATCC 15748	50-100	good-luxuriant	≥50%	purple
<i>Vibrio parahaemolyticus</i> ATCC 17802 (00037*)	50-100	good-luxuriant	≥50%	bluish green

Key : *Corresponding WDCM numbers.

Storage and Shelf Life

Store below 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. Use before expiry date on the label. 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 (6,7).

Reference

- 1.Thompson et al (ed.). 2006. The Biology of Vibrios, ASM Press, chapter 1, pg 3.
- 2.Alcamo. E.I, 2001. Fundamentals of Microbiology, 6th ed, Jones and Bartlett Publishers, Inc. pg 254, 244.
- 3.Clesceri, Greenberg and Eaton (ed.). 1998. Standard Method for the examination of Water and Waste water, 20th ed. American Public Health Association, Washington, D. C.
- 4.Kudo. H. Y et al, 2001. Improved Method for Detection of ! Vibrio parahaemolyticus @ in Seafood. ASM. Vol 67, 12ppg 5819-5823.
5. Downes F. P. and Ito K., (Eds.), 2001, Compendium of Methods for the Microbiological Examination of Foods, 4th Ed., American Public Health Association, Washington, D.C.
6. Murray P. R., Baron J. H., Pfaller M. A., Jorgensen J. H. and Tenover F. C., (Eds.), 2003, Manual of Clinical Microbiology, 8th Ed., American Society for Microbiology, Washington, D.C.
7. Isenberg, H. . Clinical Microbiology Procedures Handbook. 2nd Edition.

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