HiCrome™ Vibrio Agar

Recommended for the isolation and selective chromogenic differentiation of *Vibrio* species from seafood.

M1682

Composition **

Ingredients	Grams/Litre
Peptone	10.00
Sodium chloride	25.00
Sodium thiosulphate	5.00
Sodium citrate	6.00
Sodium cholate	1.00
Chromgenic mixture	5.50
Agar	15.00

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 and pour 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 (4). *Vibrio* species are mainly responsible for causing cholera and food poisoning in humans. *Vibrio cholerae* causes cholerae due to the intake of contaminated food such as raw oysters. *Vibrio parahaemolyticus* is a major cause of food borne infections, causing food poisoning (1). 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 (4). The widely used media for *Vibrio* isolation are TCBS Agar and Alkaline Peptone Water (2). 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 (3).

Peptone provides carbonaceous and nitrogeneous compounds, long chain amino acids 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.

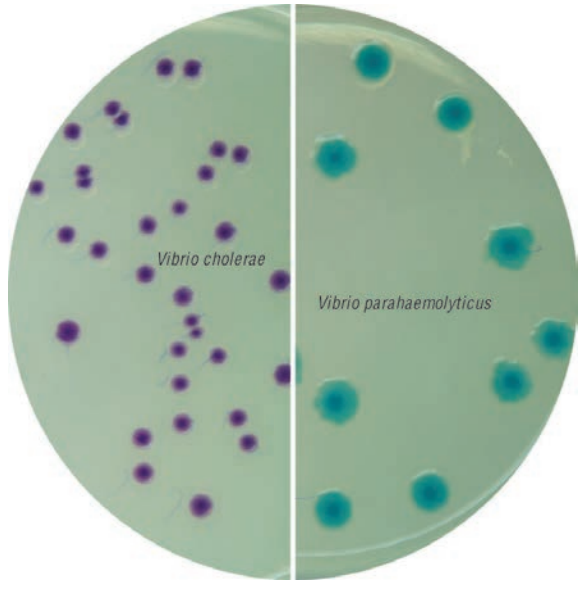
Type of specimen

Food samples

Specimen Collection and Handling

For food samples, follow appropriate techniques for sample collection, processing as per guidelines and local standards (1).

After use, contaminated materials must be sterilized by autoclaving before discarding.



M1682 HiCrome™ Vibrio Agar

HiCromeVeg™ Single Streak Rapid Differentiation Series *Freedom from BSE / TSE worries*

HiCrome™ ***Vibrio*** Agar (M1682) is also available as HiCrome™ ***Vibrio*** HiVeg™ Agar (MV1682) & HiCrome™ ***Vibrio*** HiCynth™ Agar (MCD1682) wherein all the animal origin nutrients have been replaced by vegetable based nutrients and chemically defined peptones respectively.

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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

1. Being highly selective, some species may show poor growth due to nutritional variations.
2. Slight colour variation may be observed depending upon strains.
3. 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 recommended temperature.

Quality Control

Appearance of Powder : Light yellow to light tan coloured, 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.
Cultural Response : Cultural characteristics observed after an incubation at 35-37°C for 18-24 hours.

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

Key * = corresponding WDCM Numbers

Storage and Shelf-life

Store between 2-8°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 inorder 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 sample must be decontaminated and disposed of in accordance with current laboratory techniques (6, 7).

References

1. Alcamo. E.I, 2001. Fundamentals of Microbiology, 6th ed, Jones and Bartlett Publishers, Inc. pg 254, 244.
2. 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.
3. Kudo. H. Y et al, 2001. Improved Method for Detection of Vibrio parahaemolyticus in Seafood. ASM. Vol 67, No. 12, pg 5819-5823.
4. Thompson et al (ed.). 2006. The Biology of Vibrios, ASM Press, chapter1, pg 3.
5. Salfinger Y., and Tortorello M.L. Fifth (Ed.), 2001, Compendium of Methods for the Microbiological Examination of Foods, 5th Ed., American Public Health Association, Washington, D.C.
6. Isenberg, H.D. Clinical Microbiology Procedures Handbook. 2nd Edition.
7. 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.