

HiCrome® Salmonella HiVeg® Agar

MV1296

Intended Use:

Recommended for the isolation and differentiation of *Salmonella* species from coliforms by chromogenic method.

Composition**

Ingredients	g / L
HiVeg™ peptone	6.000
Yeast extract	2.500
Synthetic detergent No. I	1.000
Chromogenic mixture	5.400
Agar	13.000
Final pH (at 25°C)	7.7±0.2

**Formula adjusted, standardized to suit performance parameters

Directions

Suspend 27.9 grams in 1000 ml purified/distilled water. Gently 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

Salmonella species have been isolated from humans and almost all animals throughout the world. They cause many types of infections from mild, self-limiting gastroenteritis to life threatening typhoid fever. *Salmonella* Typhi and *Salmonella* Paratyphi A & B cause gastroenteritis, bacteremia and enteric fever, *Salmonella* Choleraesuis causes gastroenteritis and enteric fever, especially in children. *Salmonella* Typhimurium is the most frequently isolated serotype of *Salmonella*. *Salmonella* is a cause of food poisoning (1).

HiCrome® Salmonella HiVeg® Agar is a modification of the original formulation of Rambach (2) and is used for the differentiation of *Salmonella* species from other enteric bacteria. Rambach formulation differentiates *Salmonella* based on propylene glycol utilization and presence of a chromogenic indicator. However, HiCrome® Salmonella Agar medium uses only a chromogenic mixture for identification and differentiation of *Salmonella* species.

HiCrome® Salmonella HiVeg® Agar is prepared by completely replacing animal peptones with vegetable peptone to avoid BSE/TSE risks.

HiVeg® peptone and yeast extract provides nitrogenous, carbonaceous compounds and other essential growth nutrients. *Escherichia coli* and *Salmonella* are easily distinguishable due to their colony characteristics. *Salmonella* forms light purple coloured colonies with a purple halo.

E.coli and other β-glucuronidase positive organism exhibits a characteristic blue colour, due to presence of the enzyme β-glucuronidase. Other organisms form colourless colonies. The characteristic light purple and blue colour is due to the chromogenic mixture (3). Synthetic detergent No. I mixture inhibits gram-positive organisms.

Conventional method employees the H₂S production property for *Salmonella* detection which is also exhibited by other non *Salmonella* species such as *Citrobacter*, *Proteus*, etc. Hence further biochemical confirmation is required for further identification. *Salmonella* species isolated from food or clinical samples exhibit light purple colour with halo due to the specific enzyme substrate reaction.

Type of specimen

Food samples; Water samples

Specimen Collection and Handling

For food samples, follow appropriate techniques for sample collection and processing as per guidelines (4).

For water samples, follow appropriate techniques for sample collection, processing as per guidelines and local standards (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

1. The medium is selective for *Salmonella* may not support the growth of other microorganisms.
2. Most of the *Salmonella* strains show purple colonies except few which may show colorless colonies.
3. Due to nutritional variations, some strains may show poor growth.
4. Final confirmation of suspected colonies must be carried out by serological and biochemical tests.

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 1.3% Agar gel.

Colour and Clarity of prepared medium

Light amber coloured, slightly opalescent gel forms in Petri plates

Reaction

Reaction of 2.79% w/v aqueous solution at 25°C. pH : 7.7±0.2

pH

7.50-7.90

Cultural Response

Cultural characteristics observed after an incubation at 35-37°C for 24-48 hours.

Organism	Inoculum (CFU)	Growth	Recovery	Colour of Colony
<i>Staphylococcus aureus</i> subsp. <i>aureus</i> ATCC 25923 (00034*)	≥10 ⁴	inhibited	0%	
** <i>Bacillus spizizenii</i> ATCC 6633 (00003*)	≥10 ⁴	inhibited	0%	
<i>Escherichia coli</i> ATCC 25922 (00013*)	50-100	luxuriant	≥50%	blue
## <i>Proteus hauseri</i> ATCC 13315	50-100	good	40-50%	colourless
<i>Salmonella</i> Typhimurium ATCC 14028 (00031*)	50-100	luxuriant	≥50%	light purple
<i>Salmonella</i> Enteritidis ATCC 13076 (00030*)	50-100	luxuriant	≥50%	light purple
<i>Salmonella</i> Typhi ATCC 6539	50-100	good-luxuriant	≥50%	light purple

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

Storage and Shelf Life

Store dehydrated powder and prepared medium between 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 sample must be decontaminated and disposed of in accordance with current laboratory techniques (6,7).

Reference

1. Murray P. R., Baron J. H., Pfaller M. A., Jorgensen J. H. and Tenover F. C., (Ed.), 2003, Manual of Clinical Microbiology, 8th Ed., American Society for Microbiology, Washington, D.C.
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3. Greenwald R., Henderson R. W. and Yappan S., 1991, J. Clin. Microbiol., 29:2354.
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.
5. Baird R.B., Eaton A.D., and Rice E.W., (Eds.), 2015, Standard Methods for the Examination of Water and Wastewater, 23rd ed., APHA, 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.

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