HiCrome® Clostridial Agar Base

M2026

Intended Use:

Recommended for selective isolation and presumptive identification of *Clostridium* species.

Composition**

Ingredients	g / L
Tryptone	15.000
Yeast extract	10.000
Dextrose (Glucose)	1.000
Sodium chloride	5.000
Sodium thioglycollate	0.500
Chromogenic mixture	3.310
Agar	13.000
Final pH (at 25°C)	7.1±0.2

**Formula adjusted, standardized to suit performance parameters

Directions

Suspend 47.81 gram in 1000 ml purified / distilled water. Heat to boiling to dissolve the medium completely. Sterilize by autoclaving at 15 lbs pressure (121°C) for 15 minutes. Cool to 45-50°C. Aseptically add rehydrated contents of one vial of PS Selective Supplement II (FD012). Mix well and pour into sterile Petri plates.

Principle And Interpretation

One of the major species of anaerobic bacteria to cause disease in humans is *Clostridium*. *Clostridium* species cause tetanus and gas gangrene that ultimately leads to tissue damage. Another *Clostridium* species produces the lethal botulinum toxin, the causative agent of botulism (1). Clostridial Agar formulated by Vera is recommended for the selective isolation of pathogenic Clostridia form mixed flora (2). HiCrome® is the modification for chromogenic differentiation.

Tryptone and yeast extract provide the essential nutrients, mainly the nitrogen compounds, long chain amino acids and vitamins. Dextrose (Glucose) serves as the carbon or fermentable carbohydrate source. Sodium thioglycollate is the reducing agents that help to create low oxidation-reduction potential enabling the growth of Clostridia. Also the media is well supplemented to support luxuriant growth of *Clostridium* species. The selective supplements inhibits other enteric bacteria.

The ideal method of inoculation of Clostridial Agar is direct inoculation of sterile, cooled medium with the specimen (in tubes). Alternatively agar plates of the medium can also be inoculated by streaking.

Type of specimen

Clinical samples - stool, abscess, etc ; Food; Water samples.

Specimen Collection and Handling

For clinical samples follow appropriate techniques for handling specimens as per established guidelines (3,4).

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

For water samples, follow appropriate techniques for sample collection, processing as per guidelines and local standards (6).

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

Warning and Precautions

In Vitro diagnostic Use only. 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.Individual organisms differ in their growth requirement and may show variable growth patterns on the medium.

2.Each lot of the medium has been tested for the organisms specified on the COA. It is recommended to users to validate the medium for any specific microorganism other than mentioned in the COA based on the user's unique requirement.

3.Slight colour variation may be observed depending upon the utilization of the substrate by the organism.

4. Further biochemical and serological test 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 beige homogeneous free flowing powder

Gelling

Firm, comparable with 1.3% Agar gel

Colour and Clarity of prepared medium

Yellow coloured, clear to slightly opalescent gel forms in Petri plates

Reaction

Reaction of 4.78% w/v aqueous solution at 25°C. pH : 7.1±0.2

pH

6.90-7.30

Cultural Response

Cultural characteristics observed after an incubation at 35-37°C for 24-48hours (under anaerobic condition).

Organism	Inoculum (CFU)	Growth	Recovery	Colour of colony
Clostridium perfringens ATCC 13124 (00007*)	50-100	luxuriant	>=50%	Pale yellowish green
Clostridium sporogenes ATCC 11437	50-100	luxuriant	>=50%	Pale green-bluish green
Clostridium sporogenes ATCC 19404(00008*)	50-100	luxuriant	>=50%	Pale green-bluish green
Clostridium difficile ATCC 700057	50-100	luxuriant	>=50%	Pale yellowish green
Escherichia coli ATCC 25922 (00013*)	>=10^4	inhibited	0%	
Staphylococcus aureus subsp. *aureus* ATCC 25923 (00034*)	>=10^4	inhibited	0%	

Key : (*) Corresponding WDCM numbers.

Storage and Shelf Life

Store between 15-25°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 (3,4).

Reference

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3. Isenberg, H.D. Clinical Microbiology Procedures Handbook. 2nd Edition.
4. 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.
5. Salfinger Y., and Tortorello M.L. Fifth (Ed.), 2015, Compendium of Methods for the Microbiological Examination of Foods, 5th Ed., American Public Health Association, Washington, D.C.
6. Lipps WC, Braun-Howland EB, Baxter TE,eds. Standard methods for the Examination of Water and Wastewater, 24th ed. Washington DC:APHA Press; 2023.

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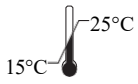
HiMedia Laboratories Pvt. Limited,
Plot No.C-40, Road No.21Y,
MIDC, Wagle Industrial Area,
Thane (W) -400604, MS, India

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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