Ready Prepared Media

Technical Information

MiCrome Bacillus Agar Plate

Product Code: PM 2651

Application: Recommended for isolation and differentiation between various species of *Bacillus* from a mixed culture in foods, clinical and non-clinical samples by chromogenic method .

Composition**

Ingredients	Gms / Litre
Peptone	10.000
HM extract #	1.000
D-Mannitol	10.000
Sodium chloride	10.000
Chromogenic mixture	3.200
Phenol red	0.025
Agar	15.000
Bacillus Selective supplement (MS2324)	
Polymyxin sulphate	10.000mg
Bacitracin	10.000mg
Final pH (at 25°C)	7.1±0.2

**Formula adjusted, standardized to suit performance parameters

#-Equivalent to Meat extract

Principle & Interpretation

Majority of *Bacillus* species apparently have little or no pathogenic potential and are rarely associated with disease in human or lower animals. The principal exception to this are *Bacillus anthracis,* the agent of anthrax, and *Bacillus cereus* , but as mall number of other species, particularly those of the *B.subtilis* group, have been implicated in food poisoning and other human and animal infections (7). *Bacillus cereus* causes food poisoning due to consumption of contaminated rice (2,5,10), other starchy foods such as potato, pasta and cheese have also been implicated, eye infections and a wide range of other clinical conditions like abscess formation, meningitis, septicemia and wound infection.

HiCrome™ Bacillus Agar is based on the formulation of MYP Agar formulated by Mossel et al (6) used for enumeration of *Bacillus cereus* and *Bacillus thuringiensis* when present in large number in certain foodstuffs

The medium contains peptone and HM extract, which provide nitrogenous, carbonaceous compounds, long chain aminoacids, vitamins and other essential growth nutrients. Mannitol serves as the fermentable carbohydrate, fermentation of which can be detected by phenol red. The chromogenic mixture present in the medium is cleaved by the enzyme beta-glucosidase found in *B.cereus* resulting in the formation of blue colonies. *B.thuringiensis* also grows as blue/green colonies on this medium as *B.cereus* and *B.thuringiensis* are biochemically identical, however *B.cereus* shows flat colonies with distinct blue centers, while *B.thuringiensis* shows irregular margins. Bacillus Selective supplement (MS2324). inhibits the growth of other *Bacillus* and contaminating microflora.

Type of specimen

Food and dairy samples.

Specimen Collection and Handling

For food and dairy samples, follow appropriate techniques for sample collection and processing as per guidelines (1,8,9).After use, contaminated materials must be sterilized by autoclaving before discarding.

Warning and Precautions

Read the label before opening the pack. 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. Due to variable nutritional requirements, some strains may show poor growth on this medium.
2. Slight colour variation may be observed depending upon the utilization of the substrate by the organism.
3. Individual organisms differ in their growth requirement and may show variable growth patterns on the medium
4. 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.

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.

Methodology

Either streak, inoculate or surface spread the test inoculum (50-100 CFU) aseptically on the plate.

Quality Control

Appearance
Sterile MiCrome Bacillus Agar in 90 mm disposable plates.
Colour of medium
Red coloured medium
Quantity of medium
25 ml of medium in 90 mm disposable plates.
pH
6.90-7.30
Sterility Test
Passes release criteria
Cultural Response
Cultural characteristics observed after an incubation at 30°C for 24-48 hours.

Organism	Inoculum(CFU)	Growth	Recovery	Colour of Colony
Bacillus subtilis subsp.spizizenii ATCC 6633 (00003*)	50-100	Inhibited	0%	
Bacillus cereus ATCC 10876	50-100	good luxuriant	>=50%	light blue,large,flat colonies with blue center
Bacillus thuringiensis ATCC 10792	50-100	good luxuriant	>=50%	light blue,large,flat colonies with irregular margins
Bacillus megaterium ATCC 14581	50-100	Inhibited	0%	
Bacillus coagulans ATCC 7050 (00002*)	50-100	Inhibited	0%	
Bacillus pumilis ATCC 14884	50-100	Poor	10-20%	light green to green colonies
Staphylococcus aureussubsp. aureus ATCC 25923 (00034*)	50-100	Inhibited	0%	
Enterococcus faecalis ATCC 29212 (00087*)	50-100	Inhibited	0%	

(*) - Corresponding WDCM numbers

Storage and Shelf Life

- On receipt store between 2-8°C 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 (3,4).

Further Reading

1. American Public Health Association, Standard Methods for the Examination of Dairy Products, 1978, 14th Ed., Washington D.C.
2. Bouza E., Grant S., Jordan C. et al, 1979, Arch. Ophthamol., 97:488.
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. Mortimer P. R. and McCann G., 1974, Lancet, 1043.
6. Mossel D. A. A., Koopman M. J. and Jongerium E., 1967, Appl. Microbiol., 15:650.
7. Murray P. R., Baron J. H., Pfaller M. A., Jorgensen J. H. and Yolken R. H., (Eds.), 2003, Manual of Clinical Microbiology, 8th Ed. American Society for Microbiology, Washington, D.C.
8. 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.
9. Wehr H. M. and Frank J. H., 2004, Standard Methods for the Microbiological Examination of Dairy Products, 17th Ed.,APHA Inc., Washington, D.C.
10. Wohlgemuth K., Kirkbride C. A., Bicknell E. J. and Ellis R. P., 1972 Am. Vet. Met, Ass. 161:1691.

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Disclaimer :

- User must ensure suitability of the product(s) in their application prior to use.
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