Modified MYP Agar Base

M1139

Intended Use:

Recommended for isolation and identification of *Bacillus* species and pathogenic Staphylococci.

Composition**

Ingredients	g/ L
Peptone	10.000
HM extract#	1.000
D-Mannitol	10.000
Sodium chloride	10.000
Phenol red	0.025
Agar	12.000
Final pH (at 25°C)	7.1±0.2

**Formula adjusted, standardized to suit performance parameters
Equivalent to Meat extract

Directions

Suspend 43.02 grams in 900 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 2 vials of sterile PolyB Selective Supplement (FD003) solution to a final concentration of 100 units per ml and 100 ml sterile Egg Yolk Emulsion (FD045) per 1000 ml medium. Mix well and pour into sterile Petri plates.

Principle And Interpretation

Bacillus cereus is ubiquitously present in soil, vegetation water and dust. It has been isolated from a large variety of foods, including vegetables, meat, cereals, pasteurized fresh milk and powdered milk (1,2,3) and processed foods. Under favorable conditions, the organism multiplies and causes gastrointestinal illness (3). It is implicated in two different forms of food poisoning; an emetic illness and a diarrheal illness. The emetic illness is mediated by a highly stable toxin that survives high temperature, exposure to trypsin, pepsin and pH extremes. The diarrheal illness is mediated by a heat and acid labile enterotoxin.

Lecithinase activity is the key reaction in the differential identification of *B.cereus*, the most commonly encountered and important species in clinical laboratories, from the majority of the other *Bacillus* species. If unknown isolate produces lecithinase, *Bacillus cereus* can be presumptively identified by also observing colonial morphology, hemolytic reactivity and motility tests. When present in large numbers in certain foodstuffs, *B. cereus* can produce metabolites responsible for the clinical symptoms of food poisoning (4). This medium differentiates *B.cereus* from other bacteria based on the basis of lecithinase activity, mannitol fermentation and resistance to polymyxin (FD003). Modified MYP Agar has similar composition to MYP Agar except agar concentration.

Modified MYP Agar Base contains peptone and HM extract, which provide nitrogen and carbon source, long chain amino acids, vitamins and other essential growth nutrients. Mannitol fermentation can be detected by phenol red, which yields yellow colour to the mannitol fermenting colonies due to acid production. Added egg yolk emulsion helps in differentiation of lecithinase producing colonies, which are surrounded by a zone of white precipitate. Addition of PolyB Selective Supplement (FD003) helps to restrict growth of gram-negative bacteria such as *Escherichia coli* and *Pseudomonas aeruginosa.* These differentiating media allow differentiation of *B.cereus* from other *Bacillus* species by its inability to ferment mannitol and poor sporulation. *B.cereus* dissimilates egg yolk and gives rise to typical bacilli form colonies with reddish zones and white halos. Acid produced by organisms other than *B.cereus* often diffuse through the medium, making it difficult to distinguish between mannitol fermenters and non-fermenters. So it is advised to transfer the suspected colonies to a fresh medium to visualize the true reaction. Colonies from Modified MYP Agar Base are subcultured on Nutrient Agar and incubated at 30°C for 24 hours to observe/determine vegetative cells, sporangium and spore morphology and lipid globules within vegetative cell.

Type of specimen

Clinical samples - Skin lesions, respiratory secretions.; Food samples; Water samples

Specimen Collection and Handling:

For clinical samples follow appropriate techniques for handling specimens as per established guidelines (5,6).
For food samples, follow appropriate techniques for sample collection and processing as per guidelines (7).
For water samples, follow appropriate techniques for sample collection, processing as per guidelines and local standards (9). After use, contaminated materials must be sterilized by autoclaving before discarding.

Warning and Precautions :

In Vitro diagnostic Use. 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.If unknown isolate produces lecithinase, *Bacillus cereus* can be presumptively identified by also observing colonial morphology, hemolytic reactivity and motility tests.
2.Individual organisms differ in their growth requirement and may show variable growth patterns on the medium.
3.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.

Quality Control

Appearance

Light yellow to pinkish purple homogeneous free flowing powder

Gelling

Firm, comparable with 1.2% Agar gel

Colour and Clarity of prepared medium

Basal medium :Red coloured clear to slightly opalescent gel. After Addition of Egg Yolk Emulsion (FD045) : Light orange coloured opaque gel forms in Petri plates

Reaction

Reaction of 4.3% w/v aqueous solution at 25°C. pH : 7.1±0.2

pH

6.90-7.30

Cultural Response

Cultural characteristics observed with added Egg Yolk Emulsion (FD045) and PolyB Selective Supplement FD003) after an incubation at 32°C for 18-40 hours.

Organism	Inoculum (CFU)	Growth	Recovery	Colour of colony	Lecithinase activity
Bacillus cereus ATCC 10876	50-100	luxuriant	>=50%	red	positive, opaque zone around the colony
#*Bacillus spizizenii* ATCC 6633 (00003*)	50-100	luxuriant	>=50%	yellow	negative
Escherichia coli ATCC 25922 (00013*)	50-100	none-poor	<=10%		Negative
Proteus mirabilis ATCC 25933	50-100	luxuriant	>=50%	red	negative
Pseudomonas aeruginosa ATCC 27853 (00025*)	50-100	none-poor	<=10%		Negative
Staphylococcus aureus subsp. *aureus* ATCC 25923 (00034*)	50-100	luxuriant	>=50%	yellow	positive, opaque zone around the colony

Key : (*) Corresponding WDCM numbers
Formerly known as *Bacillus subtilis* subsp. *spizizenii*

Storage and Shelf Life

Store between 10-30°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. 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 (5,6).

Reference

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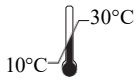
IVD ***In vitro* diagnostic medical device**



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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