



## Polymyxin Pyruvate Egg Yolk Mannitol Bromothymol Blue Agar Base (PEMBA)

M1484

Polymyxin Pyruvate Egg Yolk Mannitol Bromothymol Blue Agar Base (PEMBA) is used for the cultivation of *Bacillus cereus*.

### Composition\*\*

Ingredients	Gms / 100 ml
Peptic digest of animal tissue	0.100
Mannitol	1.000
Sodium pyruvate	1.000
Disodium phosphate	0.250
Sodium chloride	0.200
Potassium dihydrogen phosphate	0.025
Magnesium sulphate. heptahydrate	0.010
Bromo thymol blue	0.010
Agar	1.800
Final pH ( at 25°C)	7.4±0.2

\*\*Formula adjusted, standardized to suit performance parameters

### Directions

Suspend 4.39 grams in 90 ml 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 sterile rehydrated contents of 1 vial PEMBA Supplement (FD200) and 5 ml of sterile Egg Yolk Emulsion (FD045). Mix well and pour into sterile Petri plates.

### Principle And Interpretation

*Bacillus cereus* is an aerobic spore-forming bacteria commonly found in soil and isolated from different vegetables, raw and processed foods. *B. cereus* causes food poisoning due to the consumption of contaminated raw vegetables, sprouts, meat, custards, soups, boiled or fried rice (1, 2, 3). It also causes eye infections (2) and a wide range of other clinical conditions like abscess formation, meningitis, septicaemia and wound infection. *B. cereus* is a known cause of mastitis, especially in ewes and heifers (3). Polymyxin Pyruvate Egg Yolk Mannitol Bromothymol Blue Agar (PEMBA), formulated as per Holbrook and Anderson (4), is a highly specific, selective medium for the isolation and enumeration of *B. cereus* from foods (5, 6). Selectivity is attained with polymyxin and a critical concentration of nutrients (9). It supports the growth of even a small number of *B. cereus* cells and spores from foods having large number of microbial load.

The low peptone content in the medium promotes sporulation and sodium pyruvate reduces the colony size of the organisms. Egg yolk emulsion demonstrates the strong lecithinase opacity reaction (5). Bromothymol blue acts as pH indicator to detect mannitol fermentation.

Addition of polymyxin B sulphate (7, 8) at a final concentration of 100 units per ml of medium is sufficient to make the medium selective for the isolation of *B. cereus*. Cycloheximide (4mg/ l) may be used to inhibit the growth of moulds (9). Some strains of *B. cereus* have very weak egg yolk reaction. Moreover, on this medium *B. cereus* is indistinguishable from *B. thuringiensis*. *B. cereus* forms crenated blue colonies surrounded by a zone of opacity in the medium.

### Quality Control

#### Appearance

Cream to greenish yellow homogeneous free flowing powder

#### Gelling

Firm, comparable with 1.8% Agar gel

#### Colour and Clarity of prepared medium

Basal medium :Green coloured clear to slightly opalescent gel. After addition of Egg Yolk Emulsion : Forest green coloured, opaque gel forms in Petri plates

#### Reaction

Reaction of 4.4% w/v aqueous solution (basal medium) at 25°C. pH : 7.4±0.2

#### pH

7.20-7.60

#### Cultural Response

M1484: Cultural characteristics observed with added PEMBA (FD200) and Egg Yolk Emulsion (FD045), after an incubation at 35-37°C for 24-48 hours.

Organism	Inoculum (CFU)	Growth	Recovery	Colour of colony	Egg Yolk Reaction
<i>Bacillus cereus</i> ATCC 10876	50-100	good-luxuriant	≥50%	blue	positive,precipitation
<i>Escherichia coli</i> ATCC 25922	≥10 <sup>3</sup>	inhibited	0%		
<i>Proteus vulgaris</i> ATCC 13315	50-100	good-luxuriant	≥50%	green	negative
<i>Staphylococcus aureus</i> ATCC 25923	50-100	good-luxuriant	≥50%	yellow	positive, clearing

#### Storage and Shelf Life

Store below 30°C in tightly closed container and prepared media at 2 8°C. Use before expiry date on label.

#### Reference

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