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# Bacillus cereus Agar Base (Mossel)

Basal medium for the isolation and enumeration of *Bacillus cereus*, according to ISO 7932, ISO 21871 and FDA-BAM.

| TYPICAL FORMULA | (g/l) |
|---|---|
| Enzymatic Digest of Casein | 10.0 |
| Meat Extract | 1.0 |
| D-Mannitol | 10.0 |
| Sodium Chloride | 10.0 |
| Phenol Red | 0.025 |
| Agar | 14.0 |
| Final pH 7.2 ± 0.2 | |

## DESCRIPTION

Bacillus cereus Agar Base is a medium used with supplements for the selective detection of *Bacillus cereus* in food.

The complete medium (MYP) complies with the recommendations of ISO 7932, ISO 21871 and FDA-BAM for the identification of *B. cereus* on the basis of polymyxin resistance, ability to ferment mannitol and production of lecithinase.

Mossel's formulation makes this medium sufficiently selective to detect even small numbers of *Bacillus cereus* cells and spores in the presence of large numbers of other contaminants. Therefore, it is also suitable for the examination of stool specimens.

## PRINCIPLE

Enzymatic digest of casein and beef extract provide amino acids, nitrogen, carbon, vitamins and minerals. Mannitol is the fermentable carbohydrate. Sodium chloride maintain the osmotic balance of the medium. Phenol red is the pH indicator. Agar is the solidifying agent.

Egg Yolk Emulsion (ref. 80219) is added to the medium to determine lecithinase activity, whereas Bacillus cereus Supplement (ref. 81016) which contains Polymyxin B, is incorporated to confer selectivity.

## PREPARATION

Suspend 45 g of powder in 950 milliliter of deionized or distilled water. Bring to boil and shake until completely dissolved. Sterilize at 121°C for 15 minutes. Cool up to 45-50°C. Aseptically, add rehydrated content of 2 vials (10 ml) of Bacillus cereus Supplement generating a final concentration of 100,000 units of polymyxin B per liter of medium. Also add 50 ml of Egg Yolk Emulsion. Mix well and pour in Petri dishes.

## TECHNIQUE

Inoculate the medium, directly with the sample (if liquid) or its initial suspension by using the spread plate method. Repeat with further decimal dilution. Incubate at 30 ± 1°C for 24 hours under aerobic atmosphere. If colonies are not clearly visible, extend incubation for other 24 hours.

## INTERPRETATION OF RESULTS

*Bacillus cereus* forms large colonies of about 5 mm diameter, pink coloured (mannitol neg.) and typically surrounded by an opaque halo of egg yolk precipitation (lecithinase pos.). Further tests should be performed for purposes of identification.

**Note**: Other egg yolk reacting organisms such as *Staphylococcus aureus*, *Serratia marcescens* and *Proteus vulgaris*, are able to growth on this medium but are distinguished from *Bacillus cereus* by colony form and colour. In addition, these organisms produce an egg yolk-clearing reaction in contrast to egg yolk precipitate produced by *Bacillus cereus*.

## STORAGE

The powder is very hygroscopic, store the powder at 10-30°C, in a dry environment, in its original container tightly closed and use it before the expiry date on the label or until sings of deterioration or contamination are evident. Store prepared plates at 2-8°C away from light.

## WARNING AND PRECAUTIONS

The product does not contain hazardous substances in concentrations exceeding the limits set by current legislation and therefore is not classified as dangerous. it is nevertheless recommended to consult the safety data sheet for its correct use. The product is designed for *in vitro* diagnostic use only and must be used by properly trained operators.

## DISPOSAL OF WASTE

Disposal of waste must be carried out according to the national and local regulations in force.

## REFERENCES

1. EN ISO 11133:2014. Microbiology of food, animal feed and water – Preparation, production, storage and performance testing of culture media.
2. FDA-BAM Chapter 14 (2012): *Bacillus cereus.*
3. ISO 21871:2006: Microbiology of food and animal feeding stuffs – Horizontal method for the determination of low numbers of presumptive *Bacillus cereus* – Most probable number technique and detection method.
4. ISO 7932:2004: Microbiology of food and animal feeding stuffs – Horizontal method for the enumeration of presumptive *Bacillus cereus* – Colony-count technique at 30 °C
5. FDA-BAM Media M95 (1998): Mannitol-Egg Yolk-Polymyxin (MYP) Agar.
6. Mossel, D.A.A., Koopman, M.J. and Jongerius, E. (1967): Enumeration of Bacillus cereus in foods. Appl. Microbiol. 15: 650-653.
7. Donovan, K.O. (1958): A selective medium for Bacillus cereus in milk. J. Appl. Bacteriol. 21: 100-103.

# PRODUCT SPECIFICATIONS

## NAME
Bacillus cereus Agar Base (Mossel)

## PRESENTATION
Dehydrated medium

## STORAGE
10-30°C

## PACKAGING

| Ref. | Content | Packaging |
|---|---|---|
| 610114 | 500 g | 500 g of powder in plastic bottle |
| 620114 | 100 g | 100 g of powder in plastic bottle |

## pH OF THE MEDIUM
7.2 ± 0.2

## USE
Bacillus cereus Agar Base is a medium used with supplements for the selective detection of *Bacillus cereus* in food and other materials, according to ISO 7932, ISO 21871 and FDA-BAM

## TECHNIQUE
Refer to technical sheet of the product

## APPEARANCE OF THE MEDIUM
Powder medium
Appearance: free-flowing, homogeneous
Colour: pinkish-beige
Ready-to-use medium
Appearance: opaque
Colour: pink-orange

## SHELFLIFE
4 years

## QUALITY CONTROL
1. Control of general characteristics, label and print
2. Microbiological control
   Inoculum for productivity: 50-100 CFU
   Inoculum for selectivity: $10^4$-$10^6$ CFU
   Inoculum for selectivity: $10^3$-$10^4$ CFU
   Incubation Conditions: 21-48 h at 30 ± 1°C, in aerobiosis

| Microorganism | | Growth | Appearance of the colonies |
|---|---|---|---|
| *Bacillus cereus* | WDCM 00001 | Good | Pink colonies with precipitation halo |
| *Escherichia coli* | WDCM 00012 | Inhibited | --- |
| *Bacillus subtilis* | WDCM 00003 | Good | Yellow colonies without precipitation halo |

## TABLE OF SYMBOLS

| | | | | |
|---|---|---|---|---|
| LOT Batch code | IVD *In vitro* Diagnostic Medical Device | Manufacturer | Use by | Fragile, handle with care |
| REF Catalogue number | Temperature limitation | Contains sufficient for <n> tests | Caution, consult instructions for use | Do not reuse |

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