

Bacillus Cereus Selective Agar Base (MYP) ISO

Cat. 1343

For the presumptive enumeration of Bacillus cereus.

Practical information

Applications	Categories
Selective enumeration	Bacillus cereus

Industry: Food

Regulations: ISO 11133 / ISO 21871 / ISO 7932

Principles and uses

Bacillus Cereus Selective Agar Base (MYP) is a medium recommended by the ISO normative 7932 for the presumptive enumeration of Bacillus cereus with the count plate technique at 30 °C.

Beef extract and enzymatic digest of casein provide nitrogen, vitamins, minerals and amino acids essential for growth. Mannitol is the fermentable carbohydrate providing carbon and energy, Bacillus cereus is mannitol-negative. The mannitol content allows the identification of the accompanying mannitol positive flora, which are characterized by a yellow color. Phenol red is the pH indicator. Bacteriological agar is the solidifying agent.

Bacillus cereus is resistant to certain concentrations of polymyxin, which inhibits the accompanying flora, and it is effective mainly against gram-negative organisms. Bacillus cereus produces lecithinases. The insoluble degradation products of the lecithin from egg yolk, are accumulated around the Bacillus cereus colonies, forming a white precipitate.

The presumptive colonies are large, pink (indicating that mannitol fermentation has not occurred) and generally surrounded by a zone of precipitation (indicating the production of lecithinase).

Formula in g/L

Enzymatic digest of casein	10	Bacteriological agar	15
D-mannitol	10	Beef extract	1
Phenol red	0,025	Sodium chloride	10

Preparation

Suspend 46 grams of the medium in 900 ml of distilled water. Mix well and dissolve by heating with frequent agitation. Boil for one minute until complete dissolution. Sterilize in autoclave at 121 °C for 15 minutes. Cool to 44-47 °C and aseptically add 100 ml of Egg Yolk Emulsion (Cat. 5152) and 2 vials of Bacillus Cereus Supplement (Cat. 6021) reconstituted in 5 ml of sterile distilled water. Homogenize gently and dispense into appropriate containers.

Instructions for use

For the enumeration of presumptive Bacillus cereus according to ISO 7932:

- Transfer 0,1 ml of the test sample or of the initial suspension to each of two agar plates. Make decimal dilutions if necessary.
- When, for certain products, it is desirable to estimate low numbers of B. cereus, the limits of detection may be raised by a factor of 10, by examining 1,0 ml of the test sample if the initial product is liquid, or 1,0 ml of the initial suspension for the other products.
- Distribute the 1 ml of inoculum either on the surface of a large Petri dish (140 mm) or over the surface of three small dishes (90 mm) using a sterile spreader. In both cases, prepare duplicates by using two large plates or six small plates.
- Inoculated plates should be incubated for 18 hours to 24 hours at 30 °C. If colonies are not clearly visible, incubate the plates for an additional 24 hours before counting.

Quality control

Solubility	Appearance	Color of the dehydrated medium	Color of the prepared medium	Final pH (25°C)
w/o rests	Fine powder	Pink-cream	Red-orange	7,2±0,2

Microbiological test

According to ISO 11133:

Incubation conditions: Productivity (24±3-44±4 h/ 30±1 °C) / Specificity, Selectivity (44±4h/ 30±1 °C).

Inoculation conditions: Productivity quantitative (100±20. Min.50 CFU) / Selectivity (10⁴-10⁶ CFU) / Specificity (10³-10⁴ CFU).

Reference media: TSA

Microrganisms	Specification	Characteristic reaction
Bacillus cereus ATCC 11778	Good growth >50%	Pink colonies with precipitation
Escherichia coli ATCC 25922	Total inhibition (0)	
Bacillus subtilis ATCC 6633		Yellow colonies without precipitation

Storage

Temp. Min.:2 °C

Temp. Max.:25 °C

Bibliography

International Standard ISO 7932 Microbiology of food and animal feeding stuffs — Horizontal method for the enumeration of presumptive *Bacillus cereus* — Colony-count technique at 30 °C.

Mossel, D.A.A. Koopman, M.J. a Jongerius, E.: Enumeration of *Bacillus cereus* in Foods. *Appl. Microbiol.*, 1 5; 650:653 (1967)