TM 1120 – BACILLUS CEREUS AGAR BASE

INTENDED USE

Selective isolation and enumeration of *Bacillus cereus.*

PRODUCT SUMMARY AND EXPLANATION

Bacillus cereus causes food poisoning due to the consumption of contaminated rice, eye infections and a wide range of other clinical conditions like abscess formation, meningitis, septicemia and wound infection. *Bacillus cereus* is a known cause of disease mastitis, especially in ewes and heifers among the veterinarians. Holbrook and Anderson developed Bacillus Cereus Agar, which is a highly specific and selective medium for the isolation and enumeration of Bacillus cereus from foods. It supports the growth of even a small number of *Bacillus cereus* cells and spores in the presence of large number of other food contaminants. The typical colonies of *Bacillus cereus* are crenate, about 5 mm in diameter and have a distinctive turquoise to peacock blue colour surrounded by a good egg yolk precipitate of the same colour. The bacteria do not ferment mannitol and thus there is no change in colour of the indicator dye around the colonies. Addition of polymyxin-B sulphate at a final concentration of 100 units per ml of medium is sufficient to make the medium selective for the isolation of *Bacillus cereus*. It suppresses the growth of accompanying bacterial flora. If moulds are suspected in the inoculum, 40 mcg per ml filter-sterilized cycloheximide may be incorporated to suppress the moulds contamination. Some strains of *Bacillus cereus* have very weak egg yolk reaction. Moreover, on this medium *Bacillus cereus* is indistinguishable from *Bacillus thuringiensis*.

For the isolation and enumeration of Bacillus cereus in foodstuffs the following method is recommended. Distribute 0.1ml of the homogenized specimen diluted in Peptone Water onto the surface of the medium. Incubate at 37°C under aerobic conditions for 24-48 hours. Possible growth of contaminants is greatly reduced by incubation for 24 hours. Report the results as the number of *Bacillus cereus* colonies per gram weight of the food sample. Confirmatory tests should be carried out before interpretation.

COMPOSITION

Ingredients	Gms / Ltr
Peptone	1.000
Mannitol	10.000
Sodium chloride	2.000
Magnesium sulphate	0.100
Disodium hydrogen phosphate	2.500
Potassium dihydrogen phosphate	0.250
Sodium pyruvate	10.000
Bromo thymol blue	0.120
Agar	15.000

PRINCIPLE

Peptone provides and sodium pyruvate improve egg yolk precipitation and enhance sporulation. Bromothymol blue acts as pH indicator to detect mannitol fermentation.

INSTRUCTION FOR USE

- Dissolve 20.5 grams in 475 ml purified/distilled water.
- Heat to boiling to dissolve the medium completely.
- Sterilize by autoclaving at 15 psi pressure (121°C) for 15 minutes.

- Cool to 45-50°C and aseptically add rehydrated contents of 1 vial of Polymyxin B Selective Supplement and 25 ml of sterile Egg Yolk Emulsion.
- Mix well and pour into sterile Petri plates.

QUALITY CONTROL SPECIFICATIONS

Appearance of Powder : Cream to greenish yellow homogeneous free flowing powder.

Appearance of prepared medium : Basal medium: Green coloured clear to slightly opalescent gel. After addition of egg yolk emulsion : Yellowish green coloured opaque gel forms in Petri plates

pH (at 25°C) : 7.2±0.2

INTERPRETATION

Cultural characteristics observed after incubation with added Polymyxin B Selective Supplement and Egg Yolk Emulsion.

Microorganism	ATCC	Inoculum (CFU/ml)	Growth	Recovery	Incubation Temperature	Incubation Period
Bacillus cereus	10876	50-100	Good-luxuriant	>=50%	35-37°C	24-48 Hours
Escherichia coli	25922	>=10^4	Inhibited	0%	35-37°C	24-48 Hours
Proteus vulgaris	13315	50-100	Good-luxuriant	>=50%	35-37°C	24-48 Hours
Serratia marcescens	8100	50-100	Good-luxuriant	>=50%	35-37°C	24-48 Hours
Staphylococcus aureus subsp. aureus	25923	50-100	Good-luxuriant	>=50%	35-37°C	24-48 Hours

PACKAGING:

In pack size of 100 gm and 500 gm bottles.

STORAGE

Dehydrated powder, hygroscopic in nature, store in a dry place, in tightly-sealed containers between 25-30°C and protect from direct sunlight. Under optimal conditions, the medium has a shelf life of 4 years. When the container is opened for the first time, note the time and date on the label space provided on the container. After the desired amount of medium has been taken out replace the cap tightly to protect from hydration.

Product Deterioration: Do not use if they show evidence of microbial contamination, discoloration, drying or any other signs of deterioration.

DISPOSAL

After use, prepared plates, specimen/sample containers and other contaminated materials must be sterilized before discarding.

REFERENCES

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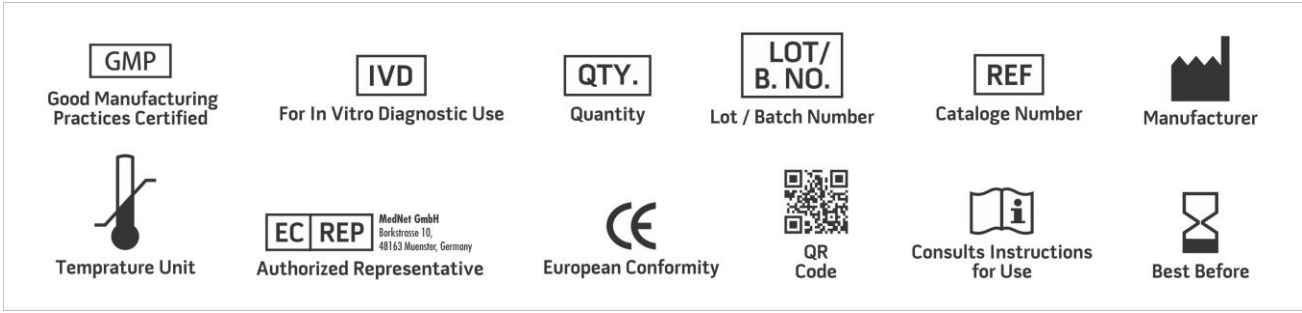

NOTE: Please consult the Material Safety Data Sheet for information regarding hazards and safe handling Practices.

***For Lab Use Only**

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