

## Technical Information

### Bacillus Differentiation Agar

**Product Code: DM 2394**

**Application:** - This medium is used for the differentiation between *Bacillus cereus* and *Bacillus subtilis* based on mannitol fermentation.

#### Composition\*\*

| Ingredients                     | Gms / Litre |
|---------------------------------|-------------|
| Yeast autolysate                | 0.200       |
| Mannitol                        | 5.000       |
| Monohydrogen ammonium phosphate | 1.000       |
| Potassium chloride              | 0.200       |
| Magnesium sulphate              | 0.200       |
| Bromo cresol purple             | 0.0075      |
| Agar                            | 15.400      |
| Final pH (at 25°C)              | 7.2±0.2     |

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

#### Principle & Interpretation

*Bacillus* is Gram positive, rod-shaped bacteria; can be obligate aerobes or facultative anaerobes <sup>(1)</sup>. Under unfavorable/ stressful environmental conditions they produce oval endospores, that can be dormant for longer periods <sup>(2)</sup>. *Bacillus cereus* causes food-borne illness and *Bacillus subtilis* is involved in food spoilage like ropiness in bread and other related foods.

Bacillus Differentiation Agar is recommended for differentiation between *Bacillus cereus* and *Bacillus subtilis* based on mannitol fermentation. Yeast autolysate provide necessary nitrogenous source for growth of *Bacillus*. Magnesium sulphate and Potassium chloride supports sporulation. Ammonium phosphate maintains buffering action. Bromocresol purple act as a pH indicator to detect mannitol fermentation.

#### Methodology

Suspend 22.0 grams of powder media in 1000 ml distilled water. Shake well & heat to dissolve the medium completely. Sterilize by autoclaving at 15 lbs pressure (121°C) for 15 minutes.

#### Quality Control

##### Physical Appearance

Light yellow to light green homogeneous free flowing powder.

##### Gelling

Firm, comparable with 1.54 % Agar gel.

##### Colour and Clarity of prepared medium

Light purple coloured clear to slightly opalescent gel forms in Petri plates.

##### Reaction

Reaction of 2.2% w/v aqueous solution at 25°C. pH : 7.2±0.2

pH Range 7.00-7.40

##### Cultural Response/ characteristics

**DM 2394:** Cultural characteristics observed after an incubation at 35-37°C for 18-24 hours.



Dehydrated Culture Media  
Bases / Media Supplements

| Organism                           | Inoculum (CFU) | Growth    | Recovery | Colour     |
|------------------------------------|----------------|-----------|----------|------------|
| <i>Bacillus cereus</i> ATCC 10876  | 50-100         | luxuriant | >=70%    | colourless |
| <i>Bacillus subtilis</i> ATCC 6633 | 50-100         | luxuriant | >=70%    | yellow     |

## Storage and Shelf Life

**Dried Media:** Store below 30°C in tightly closed container and use before expiry date as mentioned on the label.

**Prepared Media:** 2-8° in sealable plastic bags for 2-5 days.

## Further Reading

1. Turnbull PCB (1996). *Bacillus*. In: Barron's Medical Microbiology (Baron S et al., eds.) (4th ed.). Univ of Texas Medical Branch.
2. Madigan M; Martinko J (editors). (2005). Brock Biology of Microorganisms (11th ed.). Prentice Hall.

## Disclaimer :

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