



**CHROMOGENIC BACILLUS AGAR**

**TM 1523**

**INTENDED USE**

For isolation & differentiation between various species of *Bacillus* using chromogenic substrates.

**COMPOSITION**

| Ingredients                    | Gms/Ltr |
|--------------------------------|---------|
| Agar                           | 15.000  |
| Sodium chloride                | 10.000  |
| D-Mannitol                     | 10.000  |
| Peptic digest of animal tissue | 10.000  |
| Chromogenic mixture            | 3.200   |
| Meat extract                   | 1.000   |
| Phenol red                     | 0.025   |

**PRODUCT SUMMARY AND EXPLANATION**

CHROMOGENIC BACILLUS AGAR is used for isolation & differentiation between various species of *Bacillus* using chromogenic substrates. The selectivity of the medium has been optimized to prevent the over-growth of the interfering flora and to allow an easy interpretation of the results even when highly contaminated samples with competitive flora are analyzed. Chromogenic Bacillus Agar is based on the formulation of MYP Agar formulated by Mossel et al used for enumeration of *Bacillus cereus* and *Bacillus thuringiensis* when present in large number in certain foodstuffs.

**PRINCIPLE**

The medium contains peptic digest of animal tissues and Meat extract which provides nitrogenous compounds. Mannitol serves as the fermentable carbohydrate, fermentation of which can be detected by the pH indicator phenol red. Mannitol fermenting organisms like *B. megaterium* yield yellow coloured colonies. The chromogenic mixture present in the medium is cleaved by the enzyme  $\beta$ -glucosidase found in *B. cereus* resulting in the formation of blue colonies. *B. thuringiensis* will also grow as blue/green colonies on this medium as *B. cereus* and *B. thuringiensis* are biochemically identical. If selective isolation of *B. cereus* or *B. thuringiensis* is required aseptically add polymyxin B.

**INSTRUCTIONS FOR USE**

1. Dissolve 49.22 gm in 1000 ml distilled water.
2. Boil to dissolve the medium completely.
3. Sterilize by autoclaving at 15 psi pressure (121°C) for 15 minutes.



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4. Cool to 45-50°C and aseptically add rehydrated contents of 1 vial of Polymyxin B Selective Supplement (TS 058) if desired.
5. Mix well and pour into sterile Petri plates.

### QUALITY CONTROL SPECIFICATIONS

**Appearance Dehydrated powder:** Light yellow to pink colour, free flowing powder.

**Appearance of the prepared medium:** Red coloured, clear to slightly opalescent gel.

**pH (at 25°C):** 7.1 ± 0.2

### CULTURE RESPONSE

Cultural characteristics observed after inoculating incubation at 30° C and 44°C for 24 - 48 hours.

| Organism                     | ATCC  | Inoculum (CFU/ml) | Growth with supplement | Growth without supplement | Colour of Colony                                  |
|------------------------------|-------|-------------------|------------------------|---------------------------|---------------------------------------------------|
| <i>Bacillus cereus</i>       | 10876 | 50 - 100          | Good - Luxuriant       | Good-Luxuriant            | Light blue, large, flat colonies with blue centre |
| <i>Bacillus subtilis</i>     | 6633  | 50 - 100          | Inhibited              | Fair                      | yellowish green to green colonies                 |
| <i>Bacillus coagulans</i>    | 7050  | 50 - 100          | Inhibited              | Good-Luxuriant            | pink, small, raised colonies                      |
| <i>Bacillus megaterium</i>   | 14581 | 50 - 100          | Inhibited              | Good-Luxuriant            | yellow, mucoid colonies                           |
| <i>Staphylococcus aureus</i> | 25923 | 50 - 100          | Inhibited              | Good-Luxuriant            | yellow colonies                                   |

### STORAGE & STABILITY

Dehydrated powder, hygroscopic in nature, store in a dry place, in tightly-sealed containers below 8°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.

### REFERENCES

1. P.R. Mortimer, G. McCann, Lancet, 1043. (1974).
2. E. Bouza, S. Grant, C. Jordan, et. al, Arch. Ophthalmol., 97, 488. (1979).
3. K. Wohlgemuth, C.A. Kirkbride, E.J. Bicknell, R.P. Ellis, Am. Vet. Met, Ass., 161, 1691. (1972)

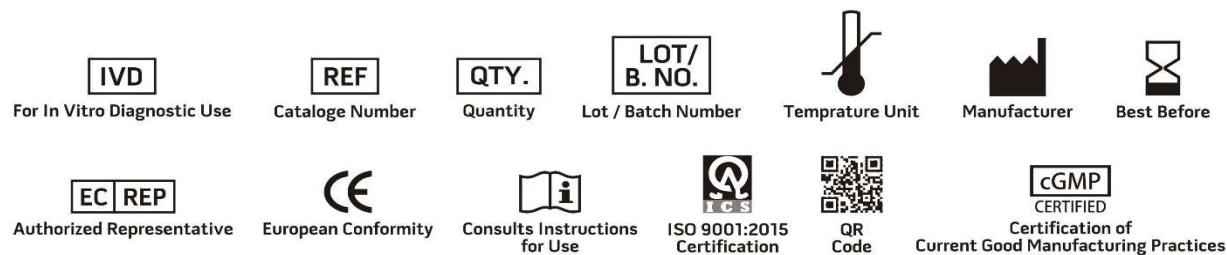
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## PRODUCT DATA SHEET



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