## **PRODUCT DATA SHEET**

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# TM 1207 - KG AGAR BASE

## **INTENDED USE**

For differentiation of *Bacillus cereus* and *Bacillus thuringiensis* based on fast sporulation.

## **PRODUCT SUMMARY AND EXPLANATION**

*Bacillus cereus*, a gram-positive rod shaped and beta haemolytic bacteria, is widely distributed in nature and can be isolated from a variety of foods. *B. cereus* is classified as a large-celled species of Group I bacilli (species with a cell width greater than 0.9µm and whose spores do not appreciably swell the sporangium). KG Agar Base formulated by Kim and Goepfert and recommended by APHA is used to promote free spore formation of *B. cereus*, *Bacillus thuringiensis* within an incubation period of 20-24 hours. This feature allows a) direct confirmation of zone forming organisms as Group I bacilli by means of microscopic examination and b) immediate differentiation of *B. cereus* from *B. thuringiensis* by visualization of the endotoxin crystal in sporulated cells of the latter organism. Additionally, Group 2 bacilli such as *Bacillus polymyxa*, which produce lecithinase, are unable to form lecithinase under the rather nutritionally poor conditions imposed by KG Agar Base.

## COMPOSITION

Ingredients	Gms / Ltr
Peptone	1.000
Yeastextract	0.500
Phenolred	0.025
Agar	18.000

## PRINCIPLE

Peptone and yeast extract in the medium supports the growth of *B. cereus, B. thuringiensis*. Lecithinase activity is observed as an opaque zone surrounding the individual colony. *B. cereus* is resistant to Polymyxin B, which restricts gram-negative organisms. *B. cereus* and *B. thuringiensis* can be distinguished by means of microscopic examination of stained cells. *B. thuringiensis* shows endotoxin crystals in sporulated cells.

## **INSTRUCTION FOR USE**

- Dissolve 19.53 grams in 900 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 100 ml sterile, Egg Yolk Emulsion and sterile contents of 2 vials of reconstituted Polymyxin B Selective Supplement.
- Mix well and pour into sterile Petri plates.

# QUALITY CONTROL SPECIFICATIONS

Appearance of Powder	: Light yellow to light pink homogeneous free flowing powder.
Appearance of prepared medium	: Basal medium: Orange coloured clear to slightly opalescent gel After addition of
	Egg Yolk Emulsion : Light orange coloured opaque gel forms in Petri plates.
pH (at 25°C)	: 6.8±0.2

#### **INTERPRETATION**

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



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Microorganism	ATCC	Inoculum (CFU/ml)	Growth	Recovery	Lecithinase	Incubation Temperature	Incubation Period
Bacillus cereus	14579	50-100	Good- luxuriant	>=50%	Positive, opaque zone around the colony	30-35°C	24Hours
Bacillus thuringiensis	10792	50-100	Good	40-50%	Positive, opaque zone around the colony	30-35°C	24Hours
Escherichia coli	25922	50-100	None- poor	0=10%	Negative	30-35°C	24Hours

## PACKAGING:

In pack size of 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

- 1. Isenberg, H.D. Clinical Microbiology Procedures Handbook 2nd Edition.
- 2. Jorgensen, J.H., Pfaller, M.A., Carroll, K.C., Funke, G., Landry, M.L., Richter, S.S and Warnock., D.W. (2015) Manual of Clinical Microbiology, 11th Edition. Vol. 1.
- 3. Kim H. V. and Goepfert J. M., 1971, Appl. Microbiol., 22:581.
- 4. Salfinger Y., and Tortorello M.L. Fifth (Ed.), 2015, Compendium of Methods for the Microbiological Examination of Foods, 5th Ed., American Public Health Association, Washington, D.C.



NOTE: Please consult the Material Safety Data Sheet for information regarding hazards and safe handling Practices. \*For Lab Use Only Revision: 08 Nov., 2019





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