

## Sheep Blood Agar, Modified Plate

MP1956

### Intended Use:

Recommended for cultivation and studying haemolytic reactions of *Bacillus cereus*. The composition and performance of this media are as per the specification laid down in ISO 21871 :2006

### Composition\*\*

Ingredients	Gms / Litre
Tryptone ##	15.000
Soya peptone#	5.000
Sodium chloride	5.000
Agar	12.500
Blood	50ml
Final pH ( at 25°C)	7.3±0.2

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

#- Equivalent to Enzymatic digest of soya

##- Equivalent to Enzymatic digest of casein

### Directions

Either streak, inoculate or surface spread the test inoculum (50-100 CFU) aseptically on the plate.

### Principle And Interpretation

Haemolysins are exotoxins produced by bacteria that lyse red blood cells. The haemolytic reaction can be visualized on blood agar plates. On blood agar plates colonies of haemolytic bacteria may be surrounded by clear, colourless zone where the red blood cells have been lysed and the haemoglobin destroyed to a colourless compound. This is beta haemolysis. Other types of bacteria can reduce haemoglobin to methaemoglobin which produces a greenish zone around the colonies and is called alpha haemolysis (5). Gamma haemolysis is no haemolysis where no change in the medium is observed (3). *Bacillus cereus* is Gram -positive aerobic or facultatively anaerobic, motile, spore forming, rod shaped bacterium that is widely distributed environmentally. *B.cereus* is associated mainly with food poisoning it is increasingly reported to be cause of serious and fatal non- gastrointestinal-tract infections.

Sheep Blood Agar Base, Modified with added sheep blood was developed to allow maximum recovery of *B.cereus* without interfering with their haemolytic reactions. This medium is formulated in accordance with ISO (4). It was formulated to be compatible with sheep blood and give improved haemolytic reactions of organisms. Tryptone and Soya peptone provide nitrogen, carbon, amino acids and vitamins. Sodium chloride maintains the osmotic balance.

### Type of specimen

Food and animal feeding stuff samples.

### Specimen Collection and Handling:

For food samples, follow appropriate techniques for sample collection and processing as per guidelines (4,6).

After use, contaminated materials must be sterilized by autoclaving before discarding.

### Warning and Precautions :

Read the label before opening the container. Wear protective gloves/protective clothing/eye protection/ face protection. Follow good microbiological lab practices while handling specimens and culture. Standard precautions as per established guidelines should be followed while handling specimens. Safety guidelines may be referred in individual safety data sheets.

### Limitations :

1. Individual organisms differ in their growth requirement and may show variable growth patterns on the medium
2. Each lot of the medium has been tested for the organisms specified on the COA. It is recommended to users to validate the medium for any specific microorganism other than mentioned in the COA based on the user's unique requirement.

## Performance and Evaluation

Performance of the medium is expected when used as per the direction on the label within the expiry period when stored at recommended temperature.

## Quality Control

### Appearance

Sterile Sheep Blood Agar, Modified in 90 mm disposable plates.

### Colour of medium

Cherry red coloured

### Quantity of medium

25 ml of medium in 90 mm disposable plates.

### Reaction

7.10-7.50

### Sterility Test

Passes release criteria

### Cultural Response

Cultural characteristics observed after an incubation at 35-37°C for 18-48 hours.

Organism	Growth	Inoculum (CFU)	Recovery	Haemolysis
<i>Bacillus cereus</i> ATCC 10876 luxuriant		50-100	≥70%	beta

## Storage and Shelf Life

On receipt store between 2-8°C Use before expiry date on the label. Product performance is best if used within stated expiry period.

## Disposal

User must ensure safe disposal by autoclaving and/or incineration of used or unusable preparations of this product. Follow established laboratory procedures in disposing of infectious materials and material that comes into contact with sample must be decontaminated and disposed of in accordance with current laboratory techniques (1,2).

## Reference

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. Koneman E. W., Allen S. D., Janda W. M., Schreckenberger P. C., Winn W. C. Jr., 1992, Colour Atlas and Textbook of Diagnostic Microbiology, 4th Ed., J. B. Lippincott Company.
4. Microbiology of Food & Animal feeding stuffs. Horizontal method for the determination of low numbers of presumptive *Bacillus cereus*-Most probable number technique and detection methods International Organization for Standardization (ISO),Draft ISO 21871:2006 .
5. Pelczar M. J. Jr., Reid R. D., Chan E. C. S., 1977, Microbiology, 4th Ed., Tata McGraw-Hill Publishing Company Ltd, New Delhi.

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