

TECHNICAL DATA SHEET

EMB Broth (Eosine Methylene Blue Broth)

Principle

Eosin Methylene Blue Broth composed of peptone, lactose, sucrose, dipotassium phosphate, Eosin and methylene blue. Peptone as a source of carbon, nitrogen, vitamins and minerals. Lactose and sucrose are fermentable carbohydrates. Dipotassium phosphate acts as buffer. Eosin and methylene blue are dyes provide selectivity to media and act as pH indicator and inhibitors. The media also used for cultivation of lactose fermenters and non-lactose fermenters. Some gram-negative bacteria can not able to metabolize lactose or metabolize lactose very slowly. Hence, the sucrose is used as secondary carbohydrate source. While the lactose non-fermenters such as *Salmonella sp.* raise the pH of surrounding by deamination of protein thus resulting in change of pH, which solubilizes the methylene blue-eosin complex resulting, color of media changes from light purple to pink or colorless. While the coliforms or lactose fermenters produce acid and decrease pH and media color changes from purple to dark purple with greenish sheen, such as *Escherichia coli* forms characteristic green colored metallic sheen. Gram-positive bacteria are partially inhibited on the medium.

Use: For the isolation of gram-negative enteric bacteria from clinical and non-clinical samples.

Contents*

Ingredients	Gram/Litre
Peptone	10.000
Lactose	5.000
Sucrose	5.000
Dipotassium Phosphate	2.000
Eosin Y	0.400
Methylene Blue	0.065
pH at 25°C	7.2 ±0.2

*** Formula adjusted for optimum performance and parameters**

Directions: Dissolve 22.50 grams in 1000 ml distilled water check. Boil to dissolve the medium completely and distribute aseptically. Sterilize by autoclaving at 15 lbs pressure (121 °C) for 15 min, cool it to 42-45 °C and inoculate test sample aseptically.

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Specimens types analyzed

The water samples, soil samples, food and dairy samples, pharmaceutical samples, clinical samples etc.

Precautions to be taken

These microbial media are intended for the in-vitro use only. All the handling, experiments, storage, and discarding should be performed with the help of skilled and knowledgeable technicians and as per the established guidelines. The material should be disposed only after proper sterilization by autoclaving. Please go through the MSDS of the media to avoid any accidents or in emergency.

Performance and Evaluation

The expected performance of the medium is liable to use as per the direction on the label when stored at optimum conditions and within expiry date.

Quality Control

Appearance	Light pink colored free flowing, homogeneous powder
Reaction of 2.25% solution	7.2 $\pm$ 0.2 at 25 °C
pH	7.00- 7.40
Color and clarity of ready medium	Reddish purple colored opalescent solution
Growth Promotion properties	Best at $\leq$ 100 CFU at 32-37 °C for 18-72 h
Indicative properties	Optimum at $\leq$ 100 CFU at 32-37 °C for 18-48 h
Negative control	Performed using sterile distilled water

Different Microbial Response:

Organism	ATCC	Inoculum	Growth	Colony Color	Incubation
<i>Escherichia coli</i>	8739	50-100	Luxuriant	Dark purple with green ting	33-37°C, 18-24 hours
<i>Salmonella typhimurium</i>	14028	50-100	Good	Colorless	33-37°C, 18-24 hours
<i>Klebsiella aerogenes</i>	13048	50-100	Good	Pink	33-37°C, 18-24 hours
<i>Staphylococcus aureus</i>	25923	50-100	Inhibited	--	33-37°C, 18-24 hours

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Storage and Shelf Life: The product is highly hygroscopic; keep the container tightly closed at all times and store it properly as per the conditions mentioned on the label. The declared expiry is valid only when stored as per the conditions mentioned on the label. Note: Sterilize media immediately after reconstitution.

Disposal: To avoid the contamination or propagation of any hazardous microbes the used, unusable or modified preparation of this product must be disposed after autoclaving after completion of task.

Reference

1. Atlas, R. M. (2004). *Handbook of microbiological media*. CRC press.
2. Atlas, R. M. (2005). *Handbook of media for environmental microbiology*. CRC press.
3. *Difco Manual* (1998). 11th Edition. Difco Laboratories., Division of Becton Dickinson and Company, Sparks, Maryland, USA.
4. Rand, M. C., Arnold E. Greenberg, and Michael J. Taras. *Standard methods for the examination of water and wastewater*. Prepared and published jointly by American Public Health Association, American Water Works Association, and Water Pollution Control Federation., 1976.

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