



Eosine Methylene Blue Agar

RDM-EMBA-01

Principle

Eosine methylene blue agar composed of peptone, lactose, sucrose, dipotassium phosphate, Eosin Y, methylene blue and agar. Peptone as a source of carbon, nitrogen, vitamins and minerals. Lactose and sucrose are fermentable carbohydrates. Dipotassium phosphate act as buffer. Eosin and methylene blue are dyes provide selectivity to media and act as pH indicator and inhibitors. The media also differentiate between the 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, solubilize the Eosin Methyl red complex and grow as colorless colonies. While the coliforms or lactose fermenters produce acid and decrease pH around the colonies, result in precipitation of both the dyes at acidic pH. produce black purple colonies, such as *Escherichia coli* forms characteristic green colored metallic sheen. Gram-positive bacteria are partially inhibited on the medium. Agar is a solidifying agent.

Use: Recommended for the differential isolation of the gram-negative Enteric Bacilli 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
Agar	13.500
pH at 25°C	7.2 ±0.2

* Formula adjusted for optimum performance and parameters

Directions: Dissolve 36.00 grams in 1000 ml distilled water check. Boil to dissolve the medium completely and sterilize by autoclaving at 15 lbs pressure (121 °C) for 15 min, cool it to 42-45 °C and distribute aseptically. Ensure complete solidification and inoculate test sample aseptically.

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	Pinkish purple colored free flowing, homogeneous powder
Reaction of 3.6% solution	7.2 ±0.2 at 25 °C
pH	7.00- 7.40
Gelling	Firm comparable with 1.35% agar gel
Color and clarity of ready medium	Purple with light greenish sheen colored opalescent gel
Growth Promotion properties	Best at ≤ 100 CFU at 32-37 °C for 18-72 h
Indicative properties	Optimum at ≤ 100 CFU at 32-37 °C for 18-48 h
Negative control	Performed using sterile distilled water

Different Microbial Response

Organism	Inoculum	Growth	Recovery	Colony Color	Incubation Temperature	Incubation period
<i>Escherichia coli</i> (ATCC 8739)	50-100	Luxurious	70-80%	Black color with Shiny Metallic Green Sheen	33-37°C	18-48 h
<i>Salmonella typhi</i> (ATCC14028)	50-100	Poor	50-60%	Translucent Amber Colored Colonies	33-37°C	18-48 h
<i>Bacillus subtilis</i> (ATCC 6633)	50-100	Inhibited	-	-	33-37°C	18-48 h
<i>Staphylococcus aureus</i> (ATCC 6538)	50-100	Inhibited	-	-	33-37°C	18-48 h

Storage and Shelf Life

Hygroscopic; keep container tightly closed. Store in cool dry place.

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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