TM 2061 – EMB AGAR, LEVINE (LEVIN EOSIN- METHYLENE BLUE AGAR) (as per IP)

INTENDED USE

For the isolation, enumeration and differentiation of members of *Enterobacteriaceae* in accordance with Indian Pharmacopoeia.

PRODUCT SUMMARY AND EXPLANATION

Levin Eosin Methylene Blue Agar was developed by Levine and is used for the differentiation of *Escherichia coli* and *Enterobacter aerogenes* and also for the rapid identification of *Candida albicans*. This medium is recommended for the detection, enumeration and differentiation of members of the coliform group by American Public Health Association and Indian Pharmacopoeia.

Eosin-Y and methylene blue make the medium slightly selective and inhibit certain gram-positive bacteria. Both dyes act as indicator and inhibiting agent. These dyes differentiate between lactose fermenters and non-fermenters. Eosin Y and methylene blue forms a complex at acidic pH which acts as inhibiting agent. Some gram-positive bacteria such as faecal Streptococci, yeasts grow on this medium and form pinpoint colonies.

Weld proposed the use of Levine EMB Agar, with added Chlortetracycline hydrochloride, for the rapid identification of *Candida albicans* in clinical specimens.

COMPOSITION

Ingredients	Gms / Ltr
Pancreatic digest of gelatin	10.000
Dibasic hydrogen phosphate	2.000
Lactose	10.000
Eosin - Y	0.400
Methylene blue	0.065
Agar	15.000

PRINCIPLE

The medium consists of pancreatic digest of gelatin which provides essential nutrients and growth factors. Eosin-Y and methylene blue serve as differential indicators. Phosphate buffers the medium.

INSTRUCTION FOR USE

- Dissolve 37.46 grams in 1000 ml purified / distilled water.
- Heat to boiling to dissolve the medium completely.
- Sterilize by autoclaving at 15 psi pressure (121°C) for 15 minutes. AVOID OVERHEATING.
- Cool to 50°C and shake the medium in order to oxidize the methylene blue (i.e. restore its blue colour) and to suspend the precipitate, which is an essential part of the medium.
- Mix well and pour into sterile Petri plates.
  Precaution: Store the medium away from light to avoid photo oxidation.

QUALITY CONTROL SPECIFICATIONS

Appearance of Powder : Light pink to purple homogeneous free flowing powder.
Appearance of prepared medium : Reddish purple with greenish cast coloured opalescent gel with finely dispersed precipitate forms in Petri plates.
pH (at 25°C) : 7.1 ± 0.2

INTERPRETATION

Cultural characteristics observed after incubation. Recovery rate is considered as 100% for bacteria growth on Soyabean Casein Digest Agar.

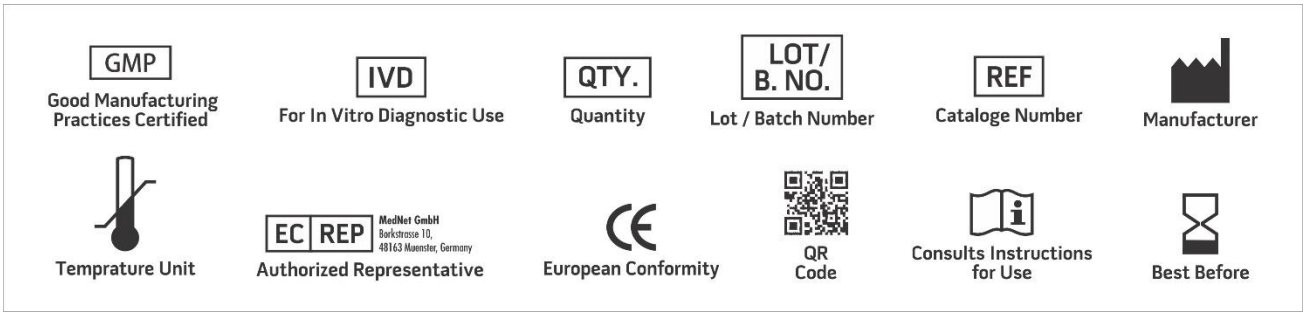
Microorganism	ATCC	Inoculum (CFU/ml)	Growth	Recovery	Colour of colony	Incubation Temperature	Incubation Period
Escherichia coli	8739	50-100	Good-luxuriant	>=50%	Blue-black with green metallic sheen	36-38°C	18-24 Hours
Escherichia coli	25922	50-100	Good-luxuriant	>=50%	Pink, red	36-38°C	18-24 Hours
Enterobacter aerogenes	13048	50-100	Good-luxuriant	>=50%	Colourless	36-38°C	18-24 Hours
Salmonella Typhimurium	14028	50-100	Good-luxuriant	>=50%	Colourless	36-38°C	18-24 Hours
Pseudomonas aeruginosa	9027	50-100	Good-luxuriant	>=50%	Colourless	36-38°C	18-24 Hours
Enterococcus faecalis	29212	>=10^3	Inhibited	0%	-	36-38°C	18-72 Hours
Staphylococcus aureus	6538	>=10^3	Inhibited	0%	-	36-38°C	18-72 Hours
Candida albicans	10231	10-100	Good-luxuriant	>=50%	Colourless	36-38°C	18-24 Hours
Saccharomyces cerevisiae	9763	10-100	None-poor	0-10%	Cream	36-38°C	18-24 Hours

PACKAGING:
In pack size of 100 gm and 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. Levine M., 1918, J. Infect. Dis., 23:43.
2. Levine M., 1921, Bull. 62, Iowa State College Engr. Exp. Station.
3. Eaton A. D., Clesceri L. S. and Greenberg A W.,(Eds.), 2005, Standard Methods for the Examination of Water and Wastewater, 21st ed., APHA, Washington, D.C.
4. Wehr H M and Frank J H., 2004, Standard Methods for the Examination of Dairy Products, 17th ed., APHA Inc., Washington, D.C.
5. Downes F P and Ito K. (Eds.), 2001, Compendium of Methods for the Microbiological Examination of Foods, 4th ed., APHA, Washington, D.C.
6. Indian Pharmacopoeia, 2007, Govt. of India, Ministry of Health and Family Welfare, New Delhi.
7. Weld J. T., 1952, Arch. Dermat. Syph., 66:691.
8. Weld J. T., 1953, Arch. Dermat. Syph., 67(5):433.



NOTE: Please consult the Material Safety Data Sheet for information regarding hazards and safe handling Practices.
***For Lab Use Only**
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