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TECHNICAL SHEET TS 610019 Rev.1 / 22.05.2012 Page 1 of 2

# E.M.B. LEVINE AGAR

Selective medium for gram-negative enteric bacteria isolation (harmonized US Pharmacopeia)

TYPICAL FORMULA	(g/l)
Peptone	10.0
Lactose	10.0
Dipotassium Phosphate	2.0
Eosin Y	0.4
Methylene Blue	0.065
Agar	15.0
Final pH 7.2 ± 0.2 at 25°C	

# DESCRIPTION

E.M.B. LEVINE AGAR is a selective medium for gram-negative enteric bacteria isolation conforms with specifications of the United States Pharmacopeia (USP). E.M.B. LEVINE AGAR is used for testing clinical materials, food and dairy products primary for the detection and confirmation of coliforms.

## PRINCIPLE

Peptone is the nitrogen source, lactose is the fermentable carbohydrate and dipotassium phosphate is the buffer. Eosin Y and methylene blue are the indicators. These dyes also play a role in differentiating between lactose fermenters and lactose non fermenters due to the presence or absence of dye uptake in the bacterial colonies. Methylene blue works also as selective agent inhibiting gram-positive bacteria to a limited degree.

#### PREPARATION

Suspend 37.5 g of powder in 1 liter of distilled water. Heat until completely dissolved. Autoclave at 121°C for 15 minutes. Cool to 45-50°C. Mix thoroughly. Dispense in petri dishes.

#### TECHNIQUE

Use standard procedures to obtain isolated colonies from specimens. A non selective medium should also be streaked to increase the chance of recovery when the population of gram-negative organisms is low and to provide and indication of other organisms present in the specimen. Incubate plates, protected from light, at 35±2 for 18-24 hours. If negative after 24 hours, reincubate an additional 24 hours.

## INTERPRETATION OF RESULTS

Lactose-fermenting microorganisms, such as coliforms, are visualized as blue-black colonies, whereas lactose non fermenters, such as Salmonella spp and Shigella spp, appear colorless, transparent or amber. Some gram-positive bacteria, such as fecal streptococci, staphylococci and yeast, will grow in this medium and usually form pinpoint colonies. A number of non pathogenic lactose non fermenting gram-negative bacteria will grow in this medium and must be distinguished from pathogenic strains by additional biochemical tests.

### STORAGE

The powder is very hygroscopic, store the powder at 10-30°C, in a dry environment, in its original container tightly closed and use it before the expiry date on the label or until sings of deterioration or contamination are evident. Store prepared plates at 2-8°C away from light.

### WARNING AND PRECAUTIONS

The product does not contain hazardous substances in concentrations exceeding the limits set by current legislation and therefore is not classified as dangerous. il is nevertheless recommended to consult the safety data sheet for its correct use. The product is intended for in vitro diagnostic use only and must be used by properly trained operators.

#### **DISPOSAL OF WASTE**

Disposal of waste must be carried out according to national and local regulations in force.

### REFERENCES

- Holf-Harris and Tongue (1916) J. infect. Dis. 18:596. 1.
- Levine (1918) J. Infect. Dis. 23:43. 2
- Marshall ed. (1993) Standard methods for the examination of dairy products, 16th ed. American Public Health Association , 3. Washington, D.C.
- 4 Downes and Ito ed. (2001) Compendium of methods for the microbiological examination of foods, 4th ed. American Public Health Association, Washington, D.C.
- United States Pharmacopeial Convention, Inc. (2001) The United States Pharmacopeia 25/The National formulary 20 2002. The 5. United States Pharmacopeial Convention, Rockville, Md.

# **PRODUCT SPECIFICATIONS**

# NAME

E.M.B. LEVINE AGAR

#### PRESENTATION

Dehydrated medium

### STORAGE

10-30°C

# PACKAGE

Ref.	Content	Packaging
610019	500 g	500 g of powder in plastic bottle
620019	100 g	100 g of powder in plastic bottle

# pH OF THE MEDIUM

 $7.2 \pm 0.2$ 

## USE

E.M.B. LEVINE AGAR is a selective medium for gram-negative enteric bacteria isolation conforms with specifications of the United States Pharmacopeia (USP)

#### TECHNIQUE

Refer to technical sheet of the product

# APPEARANCE OF THE MEDIUM

Dehydrated medium Appearance: homogeneous, free-flowing light red-purple Colour: light red-purple <u>Prepared medium</u> Appearance: slightly hazy Colour: dark red to blue-purple

### SHELFLIFE

4 years

# QUALITY CONTROL

- 1. Control of general characteristics, label and print
- Microbiological control Inoculum for productivity: 10-100 CFU/ml Inoculum for selectivity: 10<sup>4</sup>-10<sup>5</sup> CFU/ml Inoculum for specificity: ≤10<sup>4</sup> CFU/ml Incubation conditions: 18-24 h at 36 ± 1°C

Microorganism	ATCC	Growth	Features
Escherichia coli	25922	Good	Green metallic sheen
Klebsiella pneumoniae	13883	Good	Pink
Proteus mirabilis	25933	Good	Colorless
Pseudomonas aeruginosa	27853	Good	Colorless
Salmonella typhimurium	14028	Good	Colorless
Streptococcus faecalis	19433	Inhibition	



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