

Malonate Phenylalanine Broth

Cat. 1275

For the differentiation of gram-negative bacteria on the basis of the use of malonate and the pyruvic acid formation from phenylalanine.

Practical information

Applications	Categories
Detection	Gram-negative enteric bacilli

Industry: Clinical

Principles and uses

Malonate Phenylalanine Broth is used to detect the utilization of malonate and phenylalanine deamination acc. to Shaw and Clarke.

This medium is prepared according to the formulation developed by Shaw and Clarke for differentiating gram negative enteric bacteria on the basis of their ability to utilize malonate and produce pyruvic acid from phenylalanine which changes the color of the medium from green to blue due to the pH indicator Bromothymol blue. Yeast extract supplies nutrients to the organisms while two phosphates buffer the medium. Bromothymol blue is the pH indicator. Sodium chloride maintains osmotic balance. Potassium phosphates act as a buffer system. Sodium chloride supplies essential electrolytes for transport and osmotic balance.

Organisms like *Klebsiella* and *Salmonella arizonae* being capable of utilizing malonate, produce an alkaline reaction and thus change the colour of the medium from light green to dark blue. The colour of the medium remains light green if the organisms are malonate negative. Members of the group *Proteus* and *Providencia* are capable of forming pyruvic acid from phenylalanine. This can be determined by the addition of few drops of 10% iron(II) chloride (Fluka 44944) dissolved in acidified distilled water to a freshly grown culture. Deep green colour is formed due to production of pyruvic acid from phenylalanine.

Formula in g/L

Bromthymol blue	0,025	Ammonium sulfate	2
Dipotassium phosphate	0,6	DL-Phenylalanine	2
Monopotassium phosphate	0,4	Sodium chloride	2
Yeast extract	1	Sodium Malonate	3

Preparation

Suspend 11 grams of the medium in one liter of distilled water. Mix well and dissolve by heating with frequent agitation. Boil for one minute until complete dissolution. Dispense into appropriate containers and sterilize in autoclave at 121 °C for 15 minutes.

Instructions for use

Inoculate and incubate at 35±2 °C for 18-24 hours.

Quality control

Solubility	Appearance	Color of the dehydrated medium	Color of the prepared medium	Final pH (25°C)
w/o rests	Fine powder	Beige	Green	6,3±0,2

Microbiological test

Incubation conditions: (35±2 °C for 18-24 h).

Microorganisms	Specification	Characteristic reaction
<i>Klebsiella pneumoniae</i> ATCC 13883	Good growth	Malonate (+), Phenylalanine (-)

Salmonella typhimurium ATCC 14028
Escherichia coli ATCC 25922
Proteus mirabilis ATCC 25933

Good growth
Good growth
Good growth

Malonate (-), Phenylalanine (-)
Malonate (-), Phenylalanine (-)
Malonate (-), Phenylalanine (+)

Storage

Temp. Min.:2 °C
Temp. Max.:25 °C

Bibliography

Ewing, W:H, Davis, B.R. and Reavis, R.W. 1957, Phenylalanine and Malonate Media and Their Use in Enteric Bacteriology, The Public Health Laboratory, 15:153.