

Technical Data

Malonate Broth M382

Malonate Broth is recommended for the differentiation of *Enterobacter* and *Escherichia* on the basis of malonate utilization.

Composition**

Ingredients	Gms / Litre
Ammonium sulphate	2.000
Dipotassium phosphate	0.600
Monopotassium phosphate	0.400
Sodium chloride	2.000
Sodium malonate	3.000
Bromothymol blue	0.025
Final pH (at 25°C)	6.7±0.2

^{**}Formula adjusted, standardized to suit performance parameters

Directions

Dissolve 8.02 grams in 1000 ml distilled water. Dispense and sterilize by autoclaving at 15 lbs pressure (121°C) for 15 minutes. Avoid the addition of carbon and nitrogen from other sources.

Principle And Interpretation

Leifson developed a synthetic liquid medium, which differentiated *Aerobacter* (now *Enterobacter*) from *Escherichia* species based on their ability to utilize malonate (1) where *Enterobacter* utilizes malonate and *Escherichia* does not.

An organism that can simultaneously utilize sodium malonate as its carbon source and ammonium sulfate as its nitrogen source produces alkalinity due to the formation of sodium hydroxide (2). The alkali changes the color of the bromothymol blue indicator in the medium to light blue and finally to prussian blue. The color of the medium remains unchanged in the presence of an organism that cannot utilize these substances. Also some malonate-positive organisms produce only a slight alkalinity that causes the results to be difficult to interpret. Therefore these tubes should be compared with an un-inoculated malonate tube (2).

Quality Control

Appearance

Light yellow to light green homogeneous free flowing powder

Colour and Clarity of prepared medium

Bluish green coloured clear solution without any precipitate

Reaction

Reaction of 0.8% w/v aqueous solution at 25°C. pH: 6.7±0.2

pН

6.50-6.90

Cultural Response

M382: Cultural characteristics observed after an incubation at 35-37°C for 18-48 hours .

Organism	Inoculum (CFU)	Growth	Malonate Utilization
Enterobacter aerogenes ATCC 13048	50-100	luxuriant	positive reaction, dark blue colour
Escherichia coli ATCC 25922	50-100	poor-fair	negative reaction
Klebsiella pneumoniae ATCC 13883	50-100	luxuriant	positive reaction, dark blue colour

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Salmonella Arizonae ATCC 50-100 luxuriant positive reaction, dark blue colour Salmonella Typhimurium 50-100 fair-good negative reaction

Storage and Shelf Life

Store below 30°C in tightly closed container and the prepared medium at 2 - 8°C. Use before expiry date on the label.

Reference

- 1. Leifson, 1933, J. Bact., 25:329.
- 2. MacFaddin J., 1985, Media for Isolation-Cultivation-Identification-Maintenance of Medical Bacteria, Vol. I, Williams and Wilkins, Baltimore

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