ESTABLISHED 1960

EWING MALONATE BROTH MODIFIED

CAT Nº: 1212

For the differentiation of coliforms and other enteric organisms

FORMULA IN g/l

Sodium Malonate	3.00	Dipotassium Phosphate	0.60
Ammonium Sulphate	2.00	Monopotassium Phosphate	0.40
Sodium Chloride	2.00	Dextrose	0.25
Yeast Extract	1.00	Bromothymol Blue	0.025

Final pH 6.7 \pm 0.2 at 25°C

PREPARATION

Suspend 9.3 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. The prepared medium should be stored at 2-8°C. The color is green.

The dehydrated medium should be homogeneous, free-flowing and clear beige in color. If there are any physical changes, discard the medium.

USES

EWING MALONATE BROTH MODIFIED is prepared following Leifson's formula and modified with the addition of Yeast extract and Dextrose, for the differentiation of coliforms and other enteric organisms. It is widely used for the differentiation of *Enterobacter* and *Escherichia coli* based on the use of malonate.

Examples of microorganisms with positive malonate activity are *Enterobacter, Klebsiella* and strains of *Arizona*. Some examples of those not able to use malonate are *Escherichia, Salmonella* and *Serratia,* amongst others.

Malonate utilization as a carbon source, in conjunction with Ammonium sulphate as a nitrogen source during growth, produces Sodium hydroxide and thereby increased alkalinity, which changes the color of the medium from green to blue due to the pH indicator Bromothymol blue. Yeast extract is a source of vitamins, particularly of the B-group essential for bacterial growth. Dextrose is the fermentable carbohydrate providing carbon and energy. Potassium phosphates act as a buffer system. Sodium chloride supplies essential electrolytes for transport and osmotic balance.

Inoculate the broth with the suspect culture and incubate at 35±2°C for 18-48 hours in an aerobic atmosphere.

The organisms that do not utilize malonate do not produce a color change and the medium remains the original green color. Some malonate-negative strains produce a yellow color due to the fermentation of Dextrose, which increases acidity, and the medium turns yellow at a pH of 6.0.

MICROBIOLOGICAL TEST

The following results were obtained in the performance of the medium from type cultures after incubation at a temperature of $35\pm 2^{\circ}$ C and observed after 18-48 hours.

Microorganisms	Growth	Medium Color
Enterobacter aerogenes ATCC 13048	Good	Blue
Escherichia coli ATCC 25922	Good	Green
Klebsiella pneumoniae ATCC 13833	Good	Blue
Salmonella typhimurium ATCC 14028	Good	Green



Salmonella arizonae ATCC 13314 Good Blue

BIBLIOGRAPHY

Leifson, E. J. 26:329, 1993 Ewing. W. H. Identification of *Enterobacteriaceae*, Burgess Publishing Co., Minneapolis, Minn., 1972.



STORAGE

Once opened keep powdered medium closed to avoid hydration.