



PRODUCT SPECIFICATION SHEET

Malonate Broth (DM155)

Intended Use

Malonate Broth (DM155) is recommended for differentiation of *Enterobacter* and *Escherichia* on the basis of malonate utilization.

Product Summary and Explanation

Malonate Broth, prepared according to the formula described by Leifson, is a liquid medium containing ammonium sulfate as the only source of nitrogen and malonate as the only source of carbon. Leifson during his experiments demonstrated that the *Enterobacter* group utilizes malonate, producing an alkaline reaction and changing the color of the medium from green to dark blue, whereas the *Escherichia* group is unable to utilize malonate (medium remained green) and failed to grow on the medium.⁽¹⁾ Malonate Broth is further described for differentiating *Enterobacteriaceae* in food and dairy products.⁽²⁻⁴⁾ More often, the medium referenced is the modified Edwards and Ewing formulation that contains yeast extract and dextrose.⁽⁵⁾ The modification permits growth of organisms that would otherwise fail on the un-supplemented Leifson medium.

Principles of the Procedure

Malonate Broth contains ammonium sulfate, which is the sole source of nitrogen in the medium; sodium malonate is the sole source of carbon. Dipotassium phosphate and monopotassium phosphate provides buffering action. Sodium chloride maintains the osmotic balance of the medium. 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. Increased alkalinity resulting from malonate utilization causes the indicator, bromothymol blue, to change color from green to dark blue. 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

Formula / Liter

Ingredients	Gms / Liter
Ammonium sulphate	2.00
Dipotassium phosphate	0.60
Monopotassium phosphate	0.40
Sodium chloride	2.00
Sodium malonate	3.00
Bromothymol blue	0.025
Final pH: 6.7 ± 0.2 at 25°C	
Formula may be adjusted and/or supplemented as required to meet performance specifications	

Precautions

1. For Laboratory Use only.
2. IRRITANT. Irritating to eyes, respiratory system, and skin.

Directions

1. Suspend 8.02 grams of the medium in one liter of distilled water.
2. Mix well and distribute into final containers.
3. Autoclave at 121°C, 15 psi pressure, for 15 minutes / validated cycle.
4. Avoid the addition of carbon and nitrogen from other sources.





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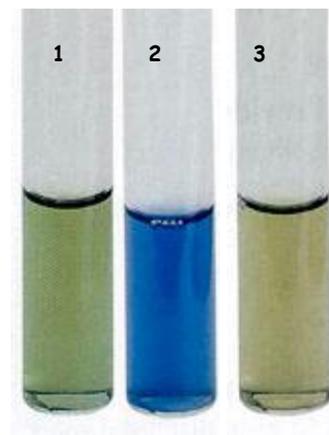
Quality Control Specifications

Dehydrated Appearance	Light yellow to light green homogeneous free flowing powder
Prepared Medium	Bluish green coloured clear solution without any precipitate
Reaction of 0.8% Solution	pH : 6.7 ± 0.2 at 25°C
Gel Strength	Not Applicable

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

Sr. No.	Organisms	Results to be achieved		
		Inoculum (CFU)	Growth	Malonate Utilization
1.	<i>Enterobacter aerogenes</i> ATCC 13048	50 -100	good-luxuriant	positive reaction, dark blue colour
2.	<i>Escherichia coli</i> ATCC 25922	50 -100	poor-fair	negative reaction
3.	<i>Klebsiella pneumoniae</i> ATCC 13883	50-100	good-luxuriant	positive reaction, dark blue colour
4.	<i>Salmonella arizonae</i> ATCC 13314	50-100	good-luxuriant	positive reaction, dark blue colour
5.	<i>Salmonella typhimurium</i> ATCC 14028	50 -100	fair-good	negative reaction

The organisms listed are the minimum that should be used for quality control testing.



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1. Uninoculated Tube
2. Enterobacter aerogenes
3. Echerichia coli

Test Procedure

1. Incubate at 35 - 37°C for 18-48 hours.
2. Inoculate tubes with a loopful of test organism.
3. Examine tubes for a change in the color of the medium from green to dark blue.

Results

Malonate utilization is indicated by a change in the color of the medium from green to dark blue,

Positive : Dark Blue

Negative : Green





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Storage

Store the sealed bottle containing the dehydrated medium at 10 - 30°C. Once opened and recapped, place container in a low humidity environment at the same storage temperature. Protect from moisture and light.

Expiration

Refer to the expiration date stamped on the container. The dehydrated medium should be discarded if not free flowing, or if the appearance has changed from the original color. Expiry applies to medium in its intact container when stored as directed.

Limitations of the Procedure

A slight bluing (blue-green) of the medium may occur after prolonged incubation. In such cases, care should be taken in interpreting results.

Packaging

Product Name : Malonate Broth

Product Code : DM155

Available Pack sizes : 100gm

References

1. Leifson, E. 1933. The fermentation of sodium malonate as a means of differentiating *Aerobacter* and *Escherichia*. J. Bacteriol. 26:329.
2. Bacteriological analytical manual. 1995. 8th ed. AOAC International, Arlington, VA.
3. Vanderzant, C., and D. F. Splittstoesser (eds.). 1992. Compendium of methods for the microbiological examination of foods, 3rd ed. American Public Health Association, Washington, D.C.
4. Marshall, R. T. (ed.). Standard methods for the examination of dairy products, 16th ed. American Public Health Association Washington, D.C.
5. Edwards, P. R., and W. H. Ewing. 1962. *Enterobacteriaceae*. U. S. Public Health Service Bulletin No. 734:19.

Further Information

For further information please contact your local MICROMASTER Representative.



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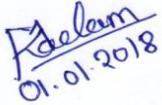
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