PRODUCT SPECIFICATION SHEET

Simmons Citrate Agar Slants (SB031)

Intended Use

Simmons Citrate Agar (DM244) is used for differentiating members of Enterobacteriaceae on the basis of citrate utilization.

Product Summary and Explanation

Simmons Citrate Agar is recommended by Ewing and Edwards[1] for the differentiation of the family Enterobacteriaceae based on whether or not citrate is utilized as the sole source of carbon. Koser[2] first developed a liquid medium for differentiating coliforms. Fecal coliforms were unable to use citrate as the sole source of carbon and inorganic ammonium salt as a sole source of nitrogen. Non-fecal coliform, such as *Enterobacter aerogenes* or *Salmonella enteritidis* could use citrate in such a medium with resultant alkalinity. Simmons Citrate Agar is a modification of Koser's medium to which bromothymol blue and 1.5% agar have been added. Organisms able to metabolize the citrate grow luxuriantly. The medium is alkalinized and changes from its initial green to deep blue in 24-48 hours. E. coli either do not grow at all on this medium, or grow so sparsely that no change in reaction is apparent. Simmons citrate agar is recommended for differentiation of enteric gram-negative bacilli from clinical specimens',[3,4] water samples',[5] and food samples.[6,7,8,9]

Principles of the Procedure

Ammoniun Dihydrogen Phosphate is the sole source of nitrogen in Simmons Citrate Agar. Magnesium is a cofactor for a variety of metabolic reactions. Phosphate acts as a buffer. Sodium citrate is the sole source of carbon in this medium. Sodium chloride maintains the osmotic balance of the medium. Agar is the solidifying agent. Bromothymol blue is the pH indicator. Organisms that can utilize ammonium dihydrogen phosphate and sodium citrate as their sole sources of nitrogen and carbon will grow on this medium and produce a color change from green (neutral) to blue (alkaline).

Formula / Liter

Ingredients	Gms / Liter
Magnesium sulphate	0.20
Ammonium dihydrogen phosphate	1.00
Dipotassium phosphate	1.00
Sodium citrate	2.00
Sodium chloride	5.00
Bromothymol blue	0.08
Agar	15.00
Final pH (at 25°C) 6.8 ± 0.2	
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.

Product Deterioration

Do not use slants if they show evidence of microbial contamination, discoloration, cracking or other signs of deterioration.

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Quality Control Specifications

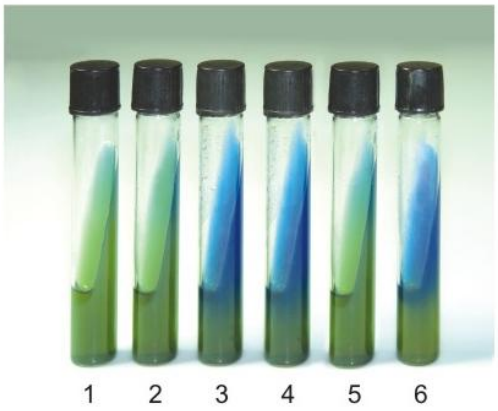
Color of medium	Forest green coloured
Clarity of medium (After sterilization)	slightly opalescent gel forms in tubes as slants
Reaction of 2.43%Solution	pH 6.8 ± 0.2 at 25°C
Gel Strength	Firm, comparable with 1.5% Agar gel
Quantity of medium	10 ml of medium in glass bottle

Sterility Check: Passes release criteria.

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	Citrate utilisation
1.	*Enterobacter aerogenes ATCC 13048*	50-100	Good-luxuriant	Positive reaction, blue colour
2.	*Escherichia coli ATCC 25922*	>=10^3	Inhibited	--
3.	*Salmonella choleraesuis ATCC 12011*	50-100	Good-luxuriant	Positive reaction, blue colour
4.	*Salmonella enteritidis ATCC 13076*	50-100	Good-luxuriant	Positive reaction, blue colour
5.	*Salmonella typhi ATCC 6539*	50-100	Fair-good	Negative reaction, green colour
6.	*Salmonella typhimurium ATCC 14028*	50-100	Good-luxuriant	Positive reaction, blue colour
7.	*Shigella dysenteriae ATCC 13313*	>=10^3	Inhibited	--

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



Simmons Citrate Agar (DM244)
1. Control
2. *Escherichia coli* ATCC 25922
3. *Enterobacter aerogenes* ATCC 13048
4. *Salmonella typhimurium* ATCC 14028
5. *Salmonella typhi* ATCC 6539
6. *Salmonella enteritidis* ATCC 13076

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Test Procedure

1. Obtain a pure culture of the organism to be tested.
2. With an inoculating needle or loop, pick the center of a well-isolated colonies obtained from solid culture media.
3. Streak only the surface of the slant with a light inoculums.
4. Incubate at 35± 2°C for 18-48 hours.

Results

A Positive reaction is indicated by growth on the slant with an intense blue color (alkaline reaction)

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

1. Due to nutritional variation, some strains may be encountered that grow poorly or fail to grow on this medium.
2. Some citrate positive organisms require a 48 hour incubation or longer for a pH change to occur.[10]

Packaging

Product Name: Simmons Citrate Agar
Product Code: DM244
Available Pack sizes: 100gm / 500gm

References

1. Edwing W. H. and Edwards P.R. (1960) Bull. Bact. Nomen. and Taxon. 10. 1-12.
2. Koser, S.A. 1923. Utilization of the salts of organic acids by the colon-aerogenes group. J. Bacteriol 8:493.
3. Pezzlo, M. (ed). 1992. Aerobic bacteriology, p. 1.0.0-1.20.47. In Isenberg, H.D. (ed), Clinical microbiology procedures handbook, vol. 1. American Society for Microbiology, Washington,D.C.
4. Baron, E. J., L. R. Peterson, S.M. Finegold. 1994. Bailey &Scott's diagnostic microbiology, 9th ed. Mosby-Year Book, Inc., St. Louis, MO.
5. Eaton, A.D., L.S. Clesceri, and A.E. Greenberg (ed.). 1995. Standard methods for the examinarion of water and wastewater, 19th ed. American Public Health Association, Washington, D.C.
6. Vanderzant, C., and D.F. Splittstoesser (ed.). 1992. Compendium of methods for the microbiological examination of foods, 3rd ed. American Public Health Association, Washington, D.C.
7. FDA Bacteriological Analytical Manual, 8th ed. AOAC International, Gaithersburg, MD.
8. Association of Official Analytical Chemists. 1995. Official methods of analysis of AOAC International, 16th ed. AOAC International, Arlington, VA.
9. Federal Register. 1996. Pathogen reduction; hazard analysis and critical point (HACCP) systems; final rule. Fed. Regis. 61:38917-38925.
10. MacFaddin, J.D. 1985. Edia for isolation –cultivation-identification-maintenance of medical bacteria, vol. 1. Williams & Wilkins, Baltimore. MD.

Further Information

For further information please contact your local MICROMASTER Representative.

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