

Instructions for Use

CRITERION™ SIMMONS CITRATE AGAR

Cat. no. C6950	CRITERION™ Simmons Citrate Agar	47.4gm
Cat. no. C6951	CRITERION™ Simmons Citrate Agar	500gm
Cat. no. C6952	CRITERION™ Simmons Citrate Agar	2kg
Cat. no. C6953	CRITERION™ Simmons Citrate Agar	10kg
Cat. no. C6954	CRITERION™ Simmons Citrate Agar	50kg

INTENDED USE

Hardy Diagnostics CRITERIONTM Simmons Citrate Agar is recommended for use in the differentiation of gramnegative enteric bacilli based on citrate utilization.

This dehydrated culture medium is a raw material intended to be used in the making of prepared media products, which will require further processing, additional ingredients, or supplements.

SUMMARY

In the early 1920s, Koser developed a liquid medium formulation for the differentiation of fecal coliforms from the coliform group. (5) Simmons later modified this formulation to produce a solid medium that eliminated potential errors when interpreting growth. (9)

CRITERIONTM Simmons Citrate Agar contains ammonium dihydrogen phosphate, which supplies the only source of nitrogen, and sodium citrate, which serves as the sole source of carbon. Organisms capable of utilizing ammonium dihydrogen phosphate and citrate will grow unrestricted on this medium. Bromothymol blue acts as a pH indicator, causing the medium to change from green (neutral) to blue (alkaline) with increasing pH. Citrate utilization produces an alkaline carbonate, resulting in a deep blue color change within the agar. The medium will remain green if organisms are not able to metabolize sodium citrate.

FORMULA*

Gram weight per liter:	23.7gm/L
Sodium Chloride	5.0gm
Sodium Citrate	2.0gm
Ammonium Dihydrogen Phosphate	2.0gm
Dipotassium Phosphate	1.0gm
Magnesium Sulfate	0.2gm

Bromothymol Blue	0.08gm
Agar	15.0gm

Final pH 6.9 +/- 0.2 at 25°C.

* Adjusted and/or supplemented as required to meet performance criteria.

STORAGE AND SHELF LIFE

Store the sealed bottle(s) containing dehydrated culture medium at 2-30°C. Dehydrated culture medium is very hygroscopic. Keep lid tightly sealed. Protect dehydrated culture media from moisture and light. The dehydrated culture media should be discarded if it is not free-flowing or if the color has changed from its original yellow with a light green tinge.

Store the prepared culture media at 2-8°C.

The expiration date on the product label applies to the product in its intact packaging when stored as directed. The product may be used and tested up to the expiration date on the product label and incubated for the recommended incubation times as stated below.

Refer to the document "Storage" for more information.

PRECAUTIONS

This product may contain components of animal origin. Certified knowledge of the origin and/or sanitary state of the animals does not guarantee the absence of transmissible pathogenic agents. Therefore, it is recommended that these products be treated as potentially infectious, and handle observing the usual universal blood precautions. Do not ingest, inhale, or allow to come into contact with skin.

This product is for laboratory use only. It is to be used only by adequately trained and qualified laboratory personnel. Observe approved biohazard precautions and aseptic techniques. All laboratory specimens should be considered infectious and handled according to "standard precautions." Refer to the document "Guidelines for Isolation Precautions" from the Centers for Disease Control and Prevention.

For additional information regarding specific precautions for the prevention of the transmission of all infectious agents from laboratory instruments and materials, and for recommendations for the management of exposure to infectious disease, refer to CLSI document M29: *Protection of Laboratory Workers from Occupationally Acquired Infections*.

Sterilize all biohazard waste before disposal.

Refer to the document "Precautions When Using Media" for more information.

METHOD OF PREPARATION FOR DEHYDRATED CULTURE MEDIA

- 1. Suspend 23.7gm of the dehydrated culture media in one liter of distilled or deionized water. Stir to mix thoroughly.
- 2. Heat to boiling to dissolve completely.
- 3. Sterilize in the autoclave at 121°C. for 15 minutes.
- 4. Cool to 45-50°C.

PROCEDURE AND INTERPRETATION OF RESULTS

For information on procedures and interpretation of results, consult listed references or refer to the prepared media Instructions for Use (IFU) for Cat. No. L80.

LIMITATIONS

It is recommended that biochemical, immunological, molecular, or mass spectrometry testing be performed on colonies from pure culture for complete identification.

Some formulations may require a settling period before pH testing of the prepared medium. If the pH is tested immediately after preparation and is out of specification, retest the medium after 24 hours to obtain final pH results. Always take pH reading at room temperature.

Failure to incubate tubes with loose caps may result in false-negative results.

Use a light inoculum to streak the slant; a heavy inoculum may result in false-positive results.

When inoculating multiple biochemicals from the same culture, inoculate this medium first, or flame inoculating needle prior to streaking this medium. Carryover of glucose or other nutrients onto this medium may result in false-positive results.

Refer to the document "Limitations of Procedures and Warranty" for more information.

MATERIALS REQUIRED BUT NOT PROVIDED

Standard microbiological supplies and equipment such as autoclaves, incinerators, and incubators, etc., are not provided.

QUALITY CONTROL

Hardy Diagnostics tests each lot of commercially manufactured media using appropriate quality control microorganisms and quality specifications as outlined on the Certificate of Analysis (CofA) and the CLSI document M22-A3 *Quality Assurance for Commercially Prepared Microbiological Culture Media*. The following microorganisms are routinely used for testing at Hardy Diagnostics:

Test Organisms	Inoculation Method*	Incubation			Results
		Time	Temperature	Atmosphere	Results
Salmonella enterica ATCC® 14028	Е	24-48hr	35°C	Aerobic	Growth; deep blue color in medium
Klebsiella pneumoniae ATCC® 13883	E	24-48hr	35°C	Aerobic	Growth; deep blue color in medium
Escherichia coli ATCC [®] 25922	Е	24-48hr	35°C	Aerobic	Inhibited; no color change to blue

^{*} Refer to the document "Inoculation Procedures for Media OC" for more information.

USER QUALITY CONTROL

Users of dehydrated culture media should perform QC testing in accordance with applicable government regulatory agencies, and in compliance with accreditation requirements. Hardy Diagnostics recommends end users check for signs of contamination and deterioration and, if dictated by laboratory quality control procedures or regulation, perform quality control testing to demonstrate growth or a positive reaction and to demonstrate inhibition or a negative reaction, if applicable. Hardy Diagnostics quality control testing is documented on the certificate of analysis (CofA) available from Hardy Diagnostics Certificate of Analysis website. In addition, refer to the following document "Finished Product Quality Control Procedures," for more information on QC or see the reference(s) for more specific information.

PHYSICAL APPEARANCE

CRITERIONTM Simmons Citrate Agar powder should appear homogeneous, free-flowing, and yellow with a light green tinge in color. The prepared medium should appear slightly opalescent, and dark green in color.

REFERENCES

- 1. Anderson, N.L., et al. *Cumitech 3B; Quality Systems in the Clinical Microbiology Laboratory*, Coordinating ed., A.S. Weissfeld. American Society for Microbiology, Washington, D.C.
- 2. Tille, P., et al. Bailey and Scott's Diagnostic Microbiology, C.V. Mosby Company, St. Louis, MO.
- 3. Isenberg, H.D. *Clinical Microbiology Procedures Handbook*, Vol. I, II & III. American Society for Microbiology, Washington, D.C.
- 4. Koneman, E.W., et al. *Color Atlas and Textbook of Diagnostic Microbiology*, J.B. Lippincott Company, Philadelphia, PA.
- 5. Koser, S.A. 1923. Utilization of the salts of organic acids by the colon-aerogenes group. J. Bacteriol., 8:493.
- 6. MacFaddin, J.F. 1985. *Media for Isolation, Cultivation, Identification, Maintenance of Bacteria*, Vol. I. Williams & Wilkins, Baltimore, MD.
- 7. Jorgensen., et al. Manual of Clinical Microbiology, American Society for Microbiology, Washington, D.C.
- 8. *Quality Assurance for Commercially Prepared Microbiological Culture Media*, M22. Clinical and Laboratory Standards Institute (CLSI formerly NCCLS), Wayne, PA.

ATCC is a registered trademark of the American Type Culture Collection.



1430 West McCoy Lane, Santa Maria, CA 93455, USA Phone: (805) 346-2766 ext. 5658

Fax: (805) 346-2760 Website: <u>HardyDiagnostics.com</u>

Email: TechnicalServices@HardyDiagnostics.com

Ordering Information

Distribution Centers:

California · Washington · Utah · Arizona · Texas · Ohio · New York · Florida · North Carolina

The Hardy Diagnostics manufacturing facility and quality management system is certified to ISO 13485.

Copyright© 2020 by Hardy Diagnostics. All rights reserved.

IFU-10258[A]

HDQA 2207B [D]