Product Insert

REINFORCED CLOSTRIDIAL AGAR (RCA)

Products

AS-6061 Reinforced Clostridial Agar (RCA) 4 plates / pkg

Intended Use

Reinforced Clostridial Agar (RCA) is an enriched non-selective medium intended for the cultivation of *Clostridia* and other anaerobic and facultative bacteria from various clinical and non-clinical specimens.

Summary

RCA agar is an enriched non-selective medium formulated by Hirsch and Grinstead to enhance the growth of clostridia from a small inoculum. Casein, beef extract, and yeast extract form the nutritive base that supplies a source of carbon, nitrogen, vitamins, and minerals. Dextrose is added as a source of carbohydrates. L-Cysteine is included as a reducing agent. Soluble starch is a detoxifier of metabolic by-products while sodium acetate acts as a buffer. This media is prepared, dispensed, and packaged under oxygen-free conditions to prevent the formation of oxidized products prior to use.

Formulation*

Pancreatic Digest of Casein	10.00	g
Beef Extract	10.00	g
Yeast Extract	3.00	g
Dextrose	5.00	g
Sodium Chloride	5.00	g
Soluble Starch	1.00	g
Sodium Acetate	3.00	g
Agar	15.00	g
L-Cysteine Hydrochloride (25.0% solution)	2.00	mL
DI Water	1.00	L

Final pH: 6.8 ± 0.3 at 25° C
Final volume: 16.0 g ± 1.6 g

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

Precautions

For *IN VITRO DIAGNOSTIC USE* only. Utilize approved biohazard precautions and aseptic technique when using this product. This product is for use only by properly-trained and qualified personnel. Sterilize all biohazard waste prior to disposal.

Storage and Shelf Life

Storage: Upon receipt, store at room temperature in original package until used. Avoid overheating or freezing. Do not use media if there are signs of deterioration (shrinking, cracking, or discoloration due to oxidation of media) or contamination. The expiration date applies to the product in its original packaging and stored as directed. Do not use product past the expiration date shown on the label.

Shelf Life: 90 days from date of manufacture.

Procedure

Specimen Collection: Specimens for anaerobic culture should be protected from oxygen during collection, transportation, and processing. Consult appropriate references for detailed instructions concerning collection and transportation of anaerobes.

Methods for Use: RCA agar should be inoculated directly with clinical specimen or from a broth that has been inoculated from a clinical specimen. Streak plates with inoculum to obtain isolated colonies and immediately place in an anaerobic atmosphere, incubating at 35-37°C for 18-48 hours. Extended periods of incubation may be required to recover some anaerobes. Detailed instructions for processing anaerobic cultures can be found in the listed references.

Materials Required, But Not Provided

Standard microbiological supplies and equipment such as loops, saline blanks, slides, staining supplies, microscope, incinerator / autoclave, incubators, anaerobic chamber / anaerobic jars, disinfectant, other culture media, and serological / biochemical reagents.

Interpretation of Results

If used properly, RCA agar will support the good growth of Clostridia and many other anaerobes isolated from clinical or non-clinical specimens.

Limitations

RCA agar will not provide complete information for identification of bacterial isolates. Additional test procedures and media are required for complete identification. It is recommended that a non-selective media, such as Brucella Blood Agar (BRU, catalog #: AS-111) also be inoculated from the same clinical specimen to assure recovery of all species present. Consult reference materials for additional information.

Quality Control

The following organisms are routinely used for quality control testing at Anaerobe Systems.

Organism Tested	ATCC #	Results	Time
Bacteroides fragilis	25285	Growth	24 hrs
Propionibacterium acnes	6919	Growth	24 – 48 hrs
Enterococcus faecalis	29212	Growth	24 hrs
Clostridium difficile	9689	Growth	24 hrs
Clostridium difficile	700057	Growth	24 hrs
Clostridium perfringens	13124	Growth	24 hrs
Clostridium innocuum	14501	Growth	24 hrs
Clostridium sporogenes	3584	Growth	24 hrs
Clostridium beijerinckii	8260	Growth	24 hrs
Clostridium sordellii	9714	Growth	24 hrs

User Quality Control: The final determination to the extent and quantity of user laboratory quality control must be determined by the end user.

If sterility testing is to be performed on this product, a representative sample of the lot(s) should be incubated anaerobically and aerobically for 48 – 96 hours.

If the nutritive capacity of this media is to be tested for performance, it is recommended that the following ATCC organisms be evaluated for growth.

Organism	ATCC #	Expected Growth
B. fragilis	25285	24 hrs
C. perfringens	13124	24 hrs
C. difficile	9689	24 hrs
C. innocuum	14501	24 hrs
E. faecalis	29212	24 hrs

Physical Appearance: RCA should appear opaque to translucent light-yellow in color.

References

1. Dowell, V. R., Jr., G. L. Lombard, F. S. Thompson and A. Y. Armfield. 1977. *Media for the Isolation, Characterization and Identification of Obligately Anaerobic Bacteria.* USDHHS, CDC. Atlanta, GA 30333.

2. Engelkirk, P. G., Duben-Engelkirk, J. and Dowell, V. R. 1992. *Principles and Practices of Clinical Anaerobic Bacteriology*. Star Publishing Co., Belmont, CA 94002.

3. Holdeman, L. V., F. P. Cato and W. E. C. Moore. 1987. *Anaerobe Laboratory Manual*. Virginia Polytechnic Institute and State University. Blacksburg, VA 24061

4. Jousimeis-Somer, H. R., Summanen, P., Citron, D. M., Baron, E. J., Wexler, H. M. and S. M. Finegold. 2002. *Wadsworth – KYL Anaerobic Bacteriology Manual*. Star Publishing Co., Belmont, CA 94002.

5. CLSI. *Quality Control for Commercially Prepared Microbiological Culture Media; Approved Standard- Third Edition.* (2004). CLSI document M22-A3. CLSI, 940 West Valley Road, Suite 1400, Wayne, PA 19087-1898.

6. Wilkins, T. D. and T. Theil. 1973. Modified Broth-Disc Method for Testing the Antibiotic Susceptibility of Anaerobic Bacteria. *Antimicrob. Agents Chemother*. 3: 350 – 356.

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