CETRIMIDE AGAR (AGAR MEDIUM N) (7688)

Intended Use

Cetrimide Agar (Agar Medium N) is used in the isolation and identification of *Pseudomonas aeruginosa*. Conforms to Harmonized USP/EP/JP Requirements.[1,2,3]

Product Summary and Explanation

Pseudomonas aeruginosa is one of the most commonly isolated pathogens, especially in clinical specimens.[4] This organism is a significant cause of burn and nosocomial infections.[5] The ability of *Pseudomonas aeruginosa* to destroy tissue may be related to the production of various extracellular enzymes.[4] *Pseudomonas aeruginosa* produces a number of water-soluble pigments, including the yellow-green or yellow-brown fluorescent pigment pyoverdin (fluorescein).[5] When pyoverdin combines with the blue water-soluble pigment pyocyanin, the bright green color characteristic of *Pseudomonas aeruginosa* is created.[5] Agar containing Cetrimide have successfully isolated *Pseudomononas aeruginosa* from contaminated specimens.[6]

King, Ward, and Raney developed Medium A to enhance the production of pyocyanin in *Pseudomonas* spp.[7] Cetrimide Agar is prepared according to this formula with the addition of Cetrimide.[7] Cetrimide Agar is recommended in the examination of food and in United States Pharmacopeia (USP XXIII) for use in Microbial Limit Test.[1,8] Cetrimide Agar is also recommended in the European Pharmacopeia and Japanese Pharmacopoeia (JP) for use in the examination of non-sterile products for specific microorganisms.[2,3]

Principles of the Procedure

Enzymatic Digest of Gelatin provides the nitrogen, vitamins, and carbon in Cetrimide Agar. Magnesium Chloride and Potassium Sulfate enhance the production of pyocyanin and fluorescein.[7] Cetrimide (cetyltrimethylammonium bromide) is the selective agent. Cetrimide acts as a quaternary ammonium cationic detergent causing nitrogen and phosphorous to be released from bacterial cells other than *Pseudomonas aeruginosa*. Agar is the solidifying agent. Glycerol is supplemented as a source of carbon.

Formula / Liter	
Enzymatic Digest of Gelatin	20 g
Magnesium Chloride	1.4 g
Potassium Sulfate	10 g
Cetrimide (Cetyltrimethylammonium Bromide)	0.3 g
Agar	13.6 g

Final pH 7.2 ± 0.2 at 25°C

Formula may be adjusted and/or supplemented as required to meet performance specifications.

Supplement /Liter
Glycerol.....10 mL

Precautions

1. For Laboratory Use.
2. Irritant. Irritating to eyes, skin, and respiratory tract. May be harmful if swallowed.

Directions

1. Suspend 45.3 g of the medium and 10 mL of glycerol in one liter of purified water.
2. Heat with frequent agitation and boil for one minute to completely dissolve the medium.
3. Autoclave at 121°C for 15 minutes.

Quality Control Specifications

Dehydrated Appearance: Powder is homogeneous, free flowing, and light beige.

Prepared Appearance: Prepared medium is light to moderately hazy and grey-white w/precipitate.

Expected Cultural Response and USP/EP/JP Growth Promotion Testing: Cultural response on Cetrimide Agar were tested at Harmonized USP/EP/JP specified temperatures and incubation times.[1,2,3]

Microorganism	Approx. Inoculum (CFU/mL)	Response	Reactions
Escherichia coli ATCC® 25922	10^3	Inhibited	---
Escherichia coli ATCC® 8739	10^3	Inhibited	---
Pseudomonas aeruginosa ATCC® 9027	10 - 100	Growth	Yellow – green to blue - green colonies
Pseudomonas aeruginosa ATCC® 10145	10 - 100	Growth	Yellow - green to blue - green colonies
Pseudomonas aeruginosa ATCC® 27853	10 - 100	Growth	Yellow - green to green colonies
Staphylococcus aureus ATCC® 25923	10^3	Inhibited	---

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

Test Procedure

Inoculate *Pseudomonas aeruginosa* colonies directly on Cetrimide Agar by the streak method from nonselective medium or the clinical specimen. When plating directly from the specimen, the inoculum level will vary.

Results

Examine plates or tubes for the presence of characteristic blue, blue-green, or yellow-green pigment. *Pseudomonas aeruginosa* typically produces both pyocyanin and fluorescein.

Storage

Store sealed bottle containing the dehydrated medium at 2 - 30°C. Once opened and recapped, place the container in a low humidity environment at the same storage temperature. Protect from moisture and light by keeping container tightly closed.

Expiration

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

Limitations of the Procedure

1. Due to nutritional variation, some strains may grow poorly or fail to grow on this medium.
2. Occasionally some enterics will exhibit a slight yellowing of the medium; however, this coloration is easily distinguished from fluorescein production because this yellowing does not fluoresce.[7]
3. Some nonfermenters and some aerobic spores formers may exhibit a water-soluble tan to brown pigmentation on this medium. *Serratia* strains may exhibit a pink pigmentation.[4]
4. Studies of Lowbury and Collins[8] showed *Ps. aeruginosa* can lose its fluorescence under UV if the cultures are left at room temperature for a short time. Fluorescence reappears when plates are reincubated.
5. Further tests are necessary for confirmation of *Ps. aeruginosa*.

Packaging

Cetrimide Agar (Agar Medium N) Code No. 7688A 500 g
7688B 2 kg
7688C 10 kg

References

1. **United States Pharmacopeial Convention.** 2007. The United States pharmacopeia, 31st ed., Amended Chapters 61, 62, 111. The United States Pharmacopeial Convention, Rockville, MD.
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3. **Japanese Pharmacopoeia.** 2007. Society of Japanese Pharmacopoeia. Amended Chapters 35.1, 35.2, 7. The Minister of Health, Labor, and Welfare.
4. **Baron, E. J., L. R. Peterson, and S. M. Finegold.** 1994. Nonfermentative gram-negative bacilli and coccobacilli, p. 386-405. Bailey & Scott's diagnostic microbiology, 9th ed. Mosby-Year Book, Inc. St. Louis, MO.
5. **Gilligan, P. H.** 1995. *Pseudomonas* and *Burkholderia*, p. 509-519. *In* P. R. Murray, E. J. Baron, M. A. Pfaller, F. C. Tenover, and R. H. Yolken (eds.)., Manual of clinical microbiology, 6th ed. American Society of Microbiology, Washington, D.C.
6. **Robin, T., and J. M. Janda. 1984.** Enhanced recovery of *Pseudomonas aeruginosa* from diverse clinical specimens on a new selective agar. Diag. Microbiol. Infect. Dis. **2**:207.
7. **King, E. O., M. K. Ward, and E. E. Raney.** 1954. Two simple media for the demonstration of pyocyanin and fluorescein. J. Lab. Clin. Med. **44**:301.
8. **Association of Official Analytical Chemists.** 1995. Bacteriological analytical manual, 8th ed. AOAC International, Gaithersburg, MD.
9. **Lowbury, E. J. L., and A. G. Collins.** 1955. The use of a new cetrimide product in a selective medium for *Pseudomonas aeruginosa*. J. Clin. Pathol. **8**:47.

Technical Information

Contact Acumedia Manufacturers, Inc. at TEL (800)327-1619 in the US/Canada or (517)371-2235 and FAX (800)329-9386 in the US/ Canada or (517)371-1106 for Technical Service on questions involving dehydrated culture media preparation or performance.