

Columbia Agar Plate (RP004H)

Intended Use

Medium for selective isolation and subculture of Clostridium sporogenes in compliance with the harmonized method of USP/EP/BP/JP.

Product Summary and Explanation

Principles of the Procedure

Cetrimide Agar Base contains gelatin peptone which supplies the nutrients necessary to support growth. The production of pyocyanin is stimulated by the magnesium chloride and potassium sulfate in the medium. Cetrimide is a quaternary ammonium, cationic detergent compound, which is inhibitory to a wide variety of bacterial species including *Pseudomonas* species other than *P. aeruginosa*. Cetrimide Agar Base is supplemented with 1% glycerol as a source of carbon.

Formula / Liter

| Ingredients | Gms / Liter | | | |
|--|-------------|--|--|--|
| Pancreatic digest of gelatin | 20.00 | | | |
| Magnesium chloride | 1.40 | | | |
| Potassium sulphate | 10.00 | | | |
| Cetrimide | 0.30 | | | |
| Agar | 15.00 | | | |
| Final pH: 7.2 ± 0.2 at 25°C | | | | |
| Formula may be adjusted and/or supplemented as required to meet performance specifications | | | | |

Precautions

- 1. Prepared plated media are For in vitro Diagnostic Use or For Laboratory Use as labeled.
- 2. Directions for use should be read and followed carefully.
- 3. If excessive moisture is observed, invert the bottom over an off-set lid and allow to air dry in order to prevent formation of a seal between the top and bottom of the plate during incubation.
- 4. Observe aseptic techniques and established precautions against microbiological hazards throughout all procedures, since it must be assumed that all specimens/samples collected might contain infectious microorganisms.

Product Deterioration

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

Quality Control Specifications

| Appearance | pearance Sterile Cetrimide Agar in 90mm plates | |
|--|--|--|
| Colour Light amber coloured medium | | |
| Reaction pH: 7.2 ± 0.2 at 25°C | | |
| Quantity of medium 25ml of medium in 90mm plates | | |

Sterility Check: Passes release criteria.

| Cultural magnana | Cultural response was observed after an incubation at 30-35°C for specified time. Recovery rate is considered as 100% for bacteria growth on Soyabean Casein |
|-------------------|---|
| Cultural response | Digest Agar. |











Expected Cultural Response:

| Sr. | Organisms | Results to be achieved | | | | |
|-----|---|------------------------|----------------|--------------------------------|----------|-------------------------------------|
| No. | | Inoculum (CFU) | Growth | Observed Lot value (CFU) | Recovery | Incubation Temperature & Time |
| 1. | Pseudomonas aeruginosa ATCC 9027 | 50 - 100 | good-luxuriant | 25 -100 | >=50 % | 30-35°C <=18 hrs |
| 2. | Escherichia coli ATCC 8739 | >=10 ³ | inhibited | 0 | 0% | 30-35°C >=72 hrs |
| 3. | Pseudomonas aeruginosa ATCC 27853 | 50 - 100 | good-luxuriant | 25 - 100 | >=50 % | 30-35°C 18-24 hrs |
| 4. | Pseudomonas aeruginosa ATCC 25668 | 50 - 100 | good-luxuriant | 25 - 100 | >=50 % | 30-35° <i>C</i> 18-24 hrs |
| 5. | Stenotrophomonas maltophila ATCC 13637 | >=10 ³ | inhibited | 0 | 0% | 30-35°C >=72 hrs |
| 6. | Escherichia coli ATCC 25922 | >=10 ³ | inhibited | 0 | 0% | 30-35°C >=72 hrs |
| 7. | Escherichia coli NCTC 9002 | >=10 ³ | inhibited | 0 | 0% | 30-35° <i>C</i> >=72 hrs |
| 8. | Staphylococcus aureus ATCC 6538 | >=10 ³ | inhibited | 0 | 0% | 30-35°C >=72 hrs |
| 9. | Staphylococcus aureus ATCC 25923 | >=10 ³ | inhibited | 0 | 0% | 30-35° <i>C</i> >=72 hrs |
| 10. | Salmonella typhimurium ATCC 14028 | >=10 ³ | inhibited | 0 | 0% | 30-35°C >=72 hrs |
| 11. | Proteus mirabilis ATCC 29906 | >=10³ | inhibited | 0 | 0% | 30-35 ° <i>C</i> >=72 hrs |

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

Test Procedure

- 1. For the isolation of *P.aeruginosa*, plates of Cetrimide Agar should be inoculated from non-selective medium such as Brain Heart Infusion Broth (DM811) or Soyabean Casein Digest Medium (DM277).
- 2. If the count is high, the test sample can be directly inoculated onto Cetrimide Agar.
- 3. Refer to appropriate references for standard test procedures.

Results

- 1. Colonies that are surrounded by a blue-green pigment and fluoresce under short wavelength (254 nm) ultraviolet light may be presumptively identified as *Pseudomonas aeruginosa*. Note, however, that certain strains of P. aeruginosa may not produce pyocyanin.
- 2. Other species of *Pseudomonas* do not produce pyocyanin, but fluoresce under UV light.
- 3. Most non-Pseudomonas species are inhibited, and some species of *Pseudomonas* may also be inhibited.
- 4. Gram staining, biochemical tests and serological procedures should be performed to confirm findings.

Storage

On receipt, store plates at 20-25°C.

Expiration









Refer to the expiration date stamped on the pack. Prepared plates stored in their original sleeve wrapping at $20-25^{\circ}C$ until just prior to use may be inoculated up to the expiration date and incubated for recommended incubation times.

Product Disposal

After use, prepared plates, specimen/sample containers and other contaminated materials must be sterilized before discarding.

Limitations of the Procedure

- 1. The type of peptone used in the base may affect pigment production.
- 2. No single medium can be depended upon to exhibit all pigment-producing *P. aeruginosa* strains.
- 3. Occasionally some enterics will exhibit a slight yellowing of the medium; however, this coloration is easily distinguished from fluorescin production since this yellowing does not fluoresce.
- 4. Some nonfermenters and some aerobic sporeformers may exhibit a water-soluble tan to brown pigmentation on this medium. Serratia strains may exhibit a pink pigmentation.
- 5. Studies of Lowbury and Collins showed *P. aeruginosa* may lose its fluorescence under UV light if the cultures are left at room temperature for a short time. Fluorescence reappears when plates are reincubated.
- 6. Consult appropriate texts for detailed information and recommended procedures.

Packaging

Product Name: Cetrimide Agar Plate

Product Code: RP002

Available Pack sizes : Pack of 10 plates

References

- 1. Leavitt, J. M., I. J. Naidorf and P. Shugaevsky. 1955. The undetected anaerobe in endodontics: a sensitive medium for detection of both aerobes and anaerobes. The NY J. Dentist. 25:377-382.
- 2. United States Pharmacopeial Convention, Inc. 2008. The United States pharmacopeia 31/The national formulary 26, Supp. 1, 8-1-08, online, United States Pharmacopeial Convention, Inc., Rockville, Md.
- MacFaddin. 1985. Media for isolation-cultivation-identification-maintenance of medical bacteria, vol. 1. Williams & Wilkins, Baltimore, Md.
- 4. Forbes, Sahm and Weissfeld. 2007. Bailey & Scott's diagnostic microbiology, 12th ed. Mosby Inc., St. Louis, Mo.
- 5. Murray, Baron, Jorgensen, Landry and Pfaller (ed.). 2007. Manual of clinical microbiology, 9th ed.American Society for Microbiology, Washington, D.C.
- Isenberg and Garcia (ed.). 2004 (update, 2007). Clinical microbiology procedures handbook, 2nd ed.American Society for Microbiology, Washington, D.C.
- 7. Eaton, Rice and Baird (ed.). 2005. Standard methods for the examination of water and wastewater, 21st ed., online. American Public Health Association, Washington, D.C.
- 8. Downes and Ito (ed.). 2001. Compendium of methods for the microbiological examination of foods, 4th ed. American Public Health Association, Washington, D.C.
- 9. U.S. Food and Drug Administration. 2001. Bacteriological analytical manual, online. AOAC International, Gaithersburg, Md.
- 10. U.S. Department of Agriculture. Microbiology laboratory guidebook, online. Food Safety and Inspection Service, USDA, Washington,
- 11. Horwitz (ed.). 2007. Official methods of analysis of AOAC International, 18th ed., online. AOAC International, Gaithersburg, Md.
- 12. Health Canada. The compendium of analytical methods, online. Food Directorate, Health Products and food Branch, Health Canada, Ottawa, Ontario Canada.
- 13. International Organization for Standardization. 1994. Microbiology General guidance for the detection of presumptive pathogenic *Yersinia enterocolitica*. ISO 10273, 1st ed., 1994-12-15. International Organization for Standardization, Geneva, Switzerland.
- 14. Wehr and Frank (ed.). 2004. Standard methods for the examination of dairy products, 17th ed. American Public Health Association, Washington, D.C.
- 15. Curry, Joyce and McEwen. 1993. CTFA microbiology guidelines. The Cosmetic, Toiletry and Fragrance Association, Inc., Washington, D.C.
- 16. Indian Pharmacopoeia, 2007, Govt. of India, Ministry of Health and Family Welfare, New Delhi, India.
- 17. Gunn B. A., Ohashi D K., Gaydos C. A., Holt E. S., 1977, J. Clin. Microbiol., 5(6): 650.





Further Information

For further information please contact your local MICROMASTER Representative.



MICROMASTER LABORATORIES PRIVATE LIMITED

RP002PS5, Rev. 00, Ver. 00/01.02.2016

Unit 38/39, Kalpataru Industrial Estate,

Off G.B. Road, Near 'R-Mall' , Thane (W) - 400607. M.S. INDIA.

Ph: +91-22-25895505, 4760, 4681. Cell: 9320126789.

Email: micromaster@micromasterlab.com

| Prepared By | Checked By | Approved By | | |
|----------------|----------------------|------------------------|--|--|
| 01.01.2018 | Ausdak 01.01.2018 | 01012018 | | |
| Microbiologist | Head Quality Control | Head Quality Assurance | | |

Disclaimer:

All Products conform exclusively to the information contained in this and other related Micromaster Publications. Users must ensure that the product(s) is appropriate for their application, prior to use. The information published in this publication is based on research and development work carried out in our laboratory and is to the best of our knowledge true and accurate. Micromaster Laboratories Pvt Ltd reserves the right to make changes to specifications and information related to the products at any time. Products are intended for laboratory, diagnostic, research or further manufacturing use only and not for human or animal or therapeutic use, unless otherwise specified. Statements included herein should not be considered as a warranty of any kind, expressed or implied, and no liability is accepted for infringement of any patents.





