Cetrimide Agar (Harmonized)

Intended Use
Cetrimide Agar is used for the isolation and cultivation of *Pseudomonas aeruginosa* from pharmaceutical products in accordance with microbial limit testing by harmonized methodology of USP/EP/BP/JP/IP.

Summary
Cetrimide Agar is based on the formulation described by King *et al.,* and is widely recommended for use in the examination of cosmetics, pharmaceuticals and clinical specimens for the presence of *P. aeruginosa*, as well as for evaluating the efficacy of disinfectants against this organism. Strains of *P. aeruginosa* are identified from specimens by the production of pyocyanin, a blue, water-soluble, nonfluorescent, phenazine pigment in addition to their colonial morphology and the characteristic grape like odour of aminoacetophenone. *P. aeruginosa* is the only species of *Pseudomonas* or Gram-negative rod known to excrete pyocyanin. Cetrimide Agar Base is therefore, a valuable culture medium in the identification of this organism. It is also included in the Bacteriological Analytical Manual for cosmetics testing and recommended by the USP, BP and IP in Microbial Limit Tests.

Principle
Cetrimide (Cetyltrimethylammonium bromide) is a quaternary ammonium compound, cationic detergent, which is inhibitory to a wide variety of bacteria including *Pseudomonas* species other than *P. aeruginosa*. It causes nitrogen and phosphorous to be released from bacterial cells other than *Pseudomonas aeruginosa*. The magnesium chloride and Dipotassium sulphate in the medium stimulates the production of pyocyanin. Presence of magnesium ions can also neutralize EDTA, if present in the sample. Pancreatic digest of gelatin provides nitrogenous compounds. Sodium chloride maintains osmotic equilibrium. *Pseudomonas aeruginosa* colonies may appear pigmented greenish (under UV light also). Addition of antibiotic Nalidixic acid can aid in inhibiting the growth of accompanying flora.

Formula*

Ingredients	**g/L**
Pancreatic Digest of Gelatin	20.0
Magnesium Chloride	1.4
Dipotassium Sulphate	10.0
Cetrimide	0.3
Agar	13.6
Final pH (at 25°C)	7.2 ± 0.2

*Adjusted to suit performance parameters

Storage and Stability
Store between 10-30°C in a tightly closed container and the prepared medium at 20-30°C. Avoid freezing and overheating. Use before expiry date on the label. Once opened keep powdered medium closed to avoid hydration.

Type of Specimen
Pharmaceutical sample

Specimen Collection and Handling
Ensure that all samples are properly labelled.
Follow appropriate techniques for handling samples as per established guidelines.
Some samples may require special handling, such as immediate refrigeration or protection from light, follow the standard procedure.
The samples must be stored and tested within the permissible time duration.
After use, contaminated materials must be sterilized by autoclaving before discarding.

Directions
1. Suspend 45.30 g of the powder in 1000 mL purified water containing 10 mL glycerol.
2. Boil with frequent agitation to dissolve the powder completely.
3. Sterilize by autoclaving at 121°C (15 psi) for 15 minutes as per validated cycle.
4. If desired, Nalidixic Selective Supplement (204140370005) may be added aseptically to 1000 mL medium.

Quality Control

Dehydrated Appearance: Light yellow coloured, homogenous, and free flowing powder.
Prepared Appearance: Light amber coloured, very slightly opalescent gel with slight precipitate forms in petridishes.
Growth Promotion Test: Growth promotion is carried out in accordance with the harmonized method of USP/EP/JP/IP and growth is observed after an incubation at 30°C-35°C for 18 to 72 hours.
Growth Promoting Properties: The test results observed are within the specified temperature and shortest period of time specified in the test, inoculating ≤ 100 cfu of appropriate microorganism at 30°C-35°C for 18 hours.
Indicative Properties: The test results observed are within the specified temperature and time, inoculating ≤100 cfu of appropriate microorganism.
Inhibitory Properties: No growth of the test microorganism occurs for the specified temperature and not less than the longest period of the time specified, inoculating >100 cfu of the appropriate microorganism at 30°C-35°C for 72 hours.

Organism (ATCC)	**Growth**	**Colour of Colony**
Growth Promoting		
Pseudomonas aeruginosa (9027)	Good	Greenish
Inhibitory		
Escherichia coli (8739)	Inhibited	-

Note: For good growth - Growth observed on test media should be comparable to the growth observed on control media.
- For inhibition no growth of test microorganism should occur.

Performance and Evaluation

Performance of the product is dependent on following parameters as per product label claim:

1. Directions
2. Storage
3. Expiry

Warranty

This product is designed to perform as described on the label and package insert. The manufacturer disclaims any implied warranty of use and sale for any other purpose.

Reference

1. King, Ward and Raney, 1954, J. Lab. Clin. Med., 44:301.
2. The United States Pharmacopoeia, 2023. United States Pharmacopoeial Convention. Rockville, MD.
3. British Pharmacopoeia, 2023, The Stationery office British Pharmacopoeia.
4. European Pharmacopoeia, 2011 European Dept. for the quality of Medicines.
5. Japanese Pharmacopoeia, 2008.
6. Indian Pharmacopoeia, 2010, Govt. of India, Ministry of Health and Family Welfare, New Delhi.
7. Data on file: Microxpress®, A Division of Tulip Diagnostics (P) Ltd.

Product Presentation:

Cat No.	**Product description**	**Pack Size**
201030050500	Dehydrated Culture Media	500 g
201030052500	Dehydrated Culture Media	2.5 k
201030055000	Dehydrated Culture Media	5 k
203030280100	Bottle Media	100 mL
203030280250	Bottle Media	6 x 250 mL
205030390100	Ready Prepared Plate (90mm)	100 Plates