

00185 Acetamide Nutrient Broth

Synthetic medium to detect microbial utilization of acetamide. Corresponds to the suggestions of the "Deutsches Institut für Normung (DIN)" and the test procedures set out in §35 LMBG for the examination of water for *Pseudomonas aeruginosa*.

Composition:

Ingredients	Grams/Litre
Acetamide	2.0
Magnesium sulfate	0.158
Sodium chloride	0.2
Sodium molybdate	0.005
Ferrous sulfate	0.0005
Dipotassium hydrogen phosphate	0.2
Final pH 7.0 +/- 0.2 at 25°C	

Store prepared media below 8°C, protected from direct light. Store dehydrated powder, in a dry place, in tightly-sealed containers at 2-25°C.

Appearance: White coloured, homogeneous, fine crystals (may with lumps). It is possible that the medium

contains grey to black crystals which comes from acetamide and ferrous sulfate. But these

has no influence on the performance of the media.

Colour and Clarity: Colourless, clear to slightly turbid solution with some precipitate.

Directions:

Dissolve 2.56 g in 950 ml distilled water. Check the pH and if necessary adjust. Make up to 1 litre and pour into tubes. Before pouring mix well to evenly distribute precipitate formed. Autoclave at 121°C for 15 minutes.

Technique:

Medium is inoculated with a couple off loops from the culture to be assayed and it is incubated at 32-35°C for 24-48 hours before going to the isolation medium.

To confirm *Ps. aeruginosa*, inoculate a loop of culture in Asparagine Proline Broth (Cat. No. 17129) and incubate at 35-37°C for 24 hours. After this period, pour 1-2 drops of Nessler's Reagent (Cat. No. 72190) per 5 ml culture and verify ammonia production: a change to yellow will mean ammonia production positive and therefore, *Ps. aeruginosa* presence.

Principle and Interpretation:

Acetamide Nutrient Broth is a basal mineral media for a wide variety of nonfermenting organisms. The acetamide is the substrate providing nitrogen and carbon. The potassium phosphate is the buffering agent and the other inorganic salts are essential for the growth of microorganisms.

Organisms growing in this medium metabolize acetamide by deamination with acylamidase activity. This ability was detected by *Alcaligens ordorans*, *Ps. aeruginosa* and *Ps. acidovorans group III (Achrombacter xylosoxidans*). Some strains deaminate acetamide slowly and may require upto 7 days. Only about 40% of pycocyanogenic strains of *Ps. aeruginosa* will show a positive reaction. It is therefore important to have further tests for the identification if the results are negative.



Cultural characteristics after 4-7 days at 35-37°C.

Organisms (ATCC)	Growth	Deamination
Pseudomonas aeruginosa (27853)	+++	+
Pseudomonas maltophilia (13637)	+++	-

References:

- 1. Deutsches Einheitsverfahren zu Wasser, Abwasser und Schlammuntersuchung, Mikrobiologische Verfahren: Nachweis von Pseudomonas aeruginosa (K8), DIN Standard 3841
- R.Y. Stainer, N.J. Palleroni, M. Doudoroff, The aerobic Pseudomonas, a taxonomic study, J. Gen. Microbiol., 42, 159 (1966)
- 3. M.J. Pickett, M.M Pedersen, Can. J. Microbiol., 16, 351 (1970)
- 4. M.J. Pickett, M.M Pedersen, Can. J. Microbiol., 16, 401 (1970)
- 5. Oberhofer, Rowen, Appl. Microbiol. 28, 720 (1974)
- 6. Gilardi, A. Van Leeuwenhoek, J. Microbiology Scrol., 39, 229 (1974)
- 7. N.M. Kelly, C.T. Keanz, Acetamide Broth for Isolation of Pseudomonas aeruginosa from patients with cystic fibrosis, J. Clin. Microbiol. 17, 159 (1983)
- 8. A.D. Eaton, L.S. Clesceri, A.E. Greenberg (ed.), Standard methods for the examination of water and wastewater, 19th ed. American Public Health Association, Washington, D.C. (1995)

Precautions and Disclaimer

This product is for R&D use only, not for drug, household, or other uses. Please consult the Material Safety Data Sheet for information regarding hazards and safe handling practices.

