



Acetamide Agar (Twin Pack)

M1033

Acetamide Agar is recommended for confirmation *Pseudomonas aeruginosa* in water samples.

Composition**

Ingredients	Gms / Litre
Part A	-
Acetamide	10.000
Part B	-
Sodium chloride	5.000
Dipotassium hydrogen phosphate	1.390
Potassium dihydrogen phosphate	0.730
Phenol red	0.012
Magnesium sulphate	0.500
Agar	15.000
Final pH (at 25°C)	7.0±0.2

**Formula adjusted, standardized to suit performance parameters

Directions

Suspend 22.63 grams of part B in 1000 ml distilled water. Add 10.0 grams of Part A. Heat to boiling to dissolve the medium completely. Dispense in tubes or as desired. Sterilize by autoclaving at 15 lbs pressure (121°C) for 15 minutes. Cool the tubes in a slanted position.

Principle And Interpretation

Acetamide Agar is formulated as per the recommendation of Standard Methods for the Examination of Water and Wastewater (1). Gilardi and others showed that a wide variety of non-fermenting organisms were capable of utilizing acetamide by using basal mineral media (2, 3). However very few organisms growing in the medium metabolize acetamide by the process of deamination (acrylamidase activity) (4, 5). This unique ability is useful in identification of various non-fermenting gram-negative organisms (6, 7, 8). This ability is shown by *Pseudomonas aeruginosa*, *Pseudomonas aciovorans* Group III (*Achromobacter xylosoxidans*) and *Alcaligenes odorans* (9). Acetamide deamination leads to the liberation of ammonia, which thereby increases the pH of the medium, leading to a subsequent colour change of the phenol red indicator from yellow orange to purplish red. Some strains require upto seven days to exhibit a positive reaction as they deaminate acrylamide slowly. However, only about 40% of apyocyanogenic strains of *Pseudomonas aeruginosa* exhibit a positive reaction. It is therefore, not advisable to rely on this test as the only criterion for identification.

The medium contains inorganic salts and acetamide a sole carbon and nitrogen source. Sodium chloride maintains the osmotic equilibrium. Phenol red is the pH indicator.

Quality Control

Appearance

Part A :Colourless deliquescent crystals Part B : Light yellow to brick red homogeneous free flowing powder

Gelling

Firm, comparable with 1.5% Agar gel

Colour and Clarity of prepared medium

Orange coloured clear to slightly opalescent gel forms in tubes as slants.

Reaction

Reaction of the medium (Mixture of 1% w/v Part A and 2.263% Part B)at 25°C. pH : 7.0±0.2

pH

6.80-7.20

Cultural Response

M1033: Cultural characteristics observed after an incubation at 35-37°C for 4-7 days.

Organism	Inoculum (CFU)	Growth	Deamination
Cultural Response <i>Stenotrophomonas maltophilia</i> ATCC 13637	50-100	good-luxuriant	negative reaction ,no purplish red colour within 7 days
<i>Pseudomonas aeruginosa</i> ATCC 27853	50-100	good-luxuriant	positive reaction, purplish red colour within 7 days

Storage and Shelf Life

Store below 30°C in tightly closed container and the prepared medium at 2-8°C. Use before expiry date on the label.

Reference

1. Eaton A. D., Clesceri L. S., and Greenberg A. W., (Eds.), 1995, Standard Methods for the Examination of Water and Wastewater, 21st Ed., APHA, Washington, D.C.
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3. Stainier, Palleroni and Doudoroff, 1966, J. Gen Microbiol., 43:159.
4. Pickett M. J. and Pedersen M.M., 1970, Can. J. Microbiol., 16:351.
5. Pickett M. J. and Pedersen M.M., 1970, Can. J. Microbiol., 16:401.
6. Hedberg, 1969, Appl. Microbiol., 17: 481
7. Smith and Dayton, 1972, Appl. Microbiol., 24: 143
8. Buhlmann, Vischer and Bruhin, 1961, J. Bacteriol., 82:787
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