



Asparagine Proline Broth

RDM-APB-01

Principle

Asparagine proline broth is an enrichment broth for *Pseudomonas aeruginosa*. The composition is strictly mineral base with enantiomeric forms of asparagine and proline, magnesium sulphate, dipotassium sulphate and potassium sulphate. DL-asparagine and proline serve as the sole source of nitrogen. The dipotassium and potassium salts act as a buffer system and magnesium sulfate perform multiple functions, magnesium ion required in a large variety of enzymatic reactions, reproduction and also acts as a buffer. *Pseudomonas aeruginosa* hydrolyze asparagine to aspartic acid and reproduce vigorously, the appearance of growth with or without fluorescent pigmentation is considered a presumptive test for *P. aeruginosa*. The microbial count can be determined by using the MPN test and confirmation need to be done by subculturing on cetrimide agar.

Use: Recommended for the cultivation of *Pseudomonas aeruginosa* using membrane filter technique.

Contents*

Ingredients	Gram/Litre
DL-Asparagine	2.00
L-Proline	1.00
Dipotassium phosphate, anhydrous	1.00
Magnesium Sulphate	0.50
Potassium Sulphate	10.00
pH at 25°C	7.2 ±0.2

* Formula adjusted for optimum performance and parameters

Directions: Dissolve 14.50 grams in 900 ml distilled water, boil to dissolve the medium completely allow to cool at room temperature and add 25ml of ethanol and dilute up to 1000 ml with distilled water. Distribute as desired in screw-capped bottles. Do not used polypropylene caps with seat. Close the caps so that the seal in the lid just touches the lip of the bottle. Sterilize by autoclaving at 15 lbs pressure (121 °C) for 15 min, cool it to 42-45 °C and inoculate test sample aseptically. Alternatively, sterilize the ethanol by filtration through the cellulose acetate or nitrate membrane of 0.22 µm and then aseptically to the medium after autoclaving and cooling.

Specimens types analyzed

Drinking water and packing drinks and carbonated beverages etc.

Precautions to be taken

These microbial media are intended for the in-vitro use only. All the handling, experiments, storage, and discarding should be performed with the help of skilled and knowledgeable technicians and as per the established guidelines. The material should be disposed only after proper sterilization by autoclaving. Please go through the MSDS of the media to avoid any accidents or in emergency.

Performance and Evaluation

The expected performance of the medium is liable to use as per the direction on the label when stored at optimum conditions and within expiry date.

Quality Control

Appearance	Off white colored free flowing, homogeneous powder
Reaction of 1.45% solution	7.2 ±0.2 at 25 °C
pH	7.00- 7.40
Color and clarity of ready medium	Colorless clear solution, without any precipitate
Growth Promotion properties	Best at ≤ 100 CFU at 32-37 °C for 18-72 h
Indicative properties	Optimum at ≤ 100 CFU at 32-37 °C for 18-48 h
Negative control	Performed using sterile distilled water

Different Microbial Response

Organism	Inoculum	Growth	Color	Incubation Temperature	Incubation period
<i>Pseudomonas aeruginosa</i> (ATCC 27853)	50-100	Luxurious	Greenish yellow pigment	33-37 °C	18-48 h

Storage and Shelf Life

Hygroscopic; keep container tightly closed. Store in cool dry place.

Disposal: To avoid the contamination or propagation of any hazardous microbes the used, unusable or modified preparation of this product must be disposed after autoclaving after completion of task.

Reference

1. Atlas, R. M. (2005). *Handbook of media for environmental microbiology*. CRC press.
2. Baird R.B., Eaton A.D., and Rice E.W., (Eds.), (2015), *Standard Methods for the Examination of Water and Wastewater*, 23rd Ed., APHA, Washington, D.C.
3. Bureau of Indian Standards (BIS),(2005), *Draft IS 13428:2005*

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