

TECHNICAL DATA SHEET

Azospirillum Medium w/o Agar (Twin Pack)

Principle

These mediums contain malic acid which serves as the prime carbon source and favors the growth of *Azospirillum* and prevent the overgrowth of other nitrogen fixers. Dipotassium phosphates help to maintain the pH of medium. Necessary trace elements like ferrous Sulphate, manganese Sulphate, sodium molybdate support growth of *Azospirillum* species. Divalent salts like calcium and magnesium helps metabolism of cells. Sodium chloride helps to maintain osmotic balance of cell. Due to the nitrogen fixation by diazotroph bacteria ammonia is produced, the color of the medium changed from light yellow color to greenish blue or blue (Bromothymol blue pH indicator).

Use: For the cultivation of *Azospirillum* species.

Contents*

Ingredients

Gram/Litre

Part A

Malic acid 5.000

Dipotassium hydrogen phosphate 0.500

Ferrous Sulphate 0.500

Manganese Sulphate 0.010

Magnesium Sulphate 0.200

Sodium Chloride 0.100

Bromothymol blue 0.002

Sodium molybdate 0.002

Calcium chloride 0.020

Part B# 4.000

Potassium hydroxide 6.8 ±0.2

pH at 25°C

* Formula adjusted for optimum performance and parameters # The concentration of Part B may vary to maintain pH 6.8 ± 0.2.

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Directions: Part A: Dissolve 6.33 grams in 950 ml distilled water, boil to dissolve the medium completely. Sterilize by autoclaving at 15 lbs pressure (121 °C) for 15 min, cool it to 42-45 °C. Part B: Dissolve 4 grams of part B in 50 ml of sterile distilled water. Mix 950 ml of part A with 50 ml of part B in 1-liter sterile container, mix well and dispense into sterile tubes or flasks as desired.

Specimens types analyzed
Soil samples 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	Part A: Cream to yellow homogeneous free flowing powder Part B: White to cream pellets.
Reaction of 0.63%-part A and 0.4%-part B solution	6.8 ±0.2 at 25 °C
pH	6.60- 7.00
Color and clarity of ready medium	Light yellow colored solution
Growth Promotion properties	Best at ≤ 100 CFU at 30°C for 5-8 days
Indicative properties	Optimum at ≤ 100 CFU at 30°C for 5-8 days
Negative control	Performed using sterile distilled water

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Different Microbial Response

Cultural characteristics observed after incubation at $30 \pm 2^{\circ}\text{C}$ for 5-8 days

Organism	ATCC	Inoculum (CFU)	Growth	Media color
<i>Azospirillum brasiliensis</i>	29710	50-100	Luxuriant	Blue color

Storage and Shelf Life: The product is highly hygroscopic; keep the container tightly closed at all times and store it properly as per the conditions mentioned on the label. The declared expiry is valid only when stored as per the conditions mentioned on the label. Note: Sterilize media immediately after reconstitution.

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. *Bergey's Manual of Systematic Bacteriology* (1984), Vol. 1, Pg. 100-103. Kreig NR and Holt JG (eds.), Williams and Wilkins, London.
2. *Difco Manual* (1998). 11th Edition. Difco Laboratories., Division of Becton Dickinson and Company, Sparks, Maryland, USA.
3. Jorgensen, J.H., Pfaller, M.A., Carroll, K.C., Funke, G., Landry, M.L., Richter, S.S and Warnock., D.W. (2015) *Manual of Clinical Microbiology*, 11th Edition. Vol. 1.
4. Subba Rao N. S., (1977), *Soil Microorganisms and Plant Growth*, Oxford and IBH Publishing Co., New Delhi.

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