

# TM 1649 - AZOBACTER MEDIUM (MANNITOL)

### **INTENDED USE**

For isolation, cultivation and enrichment of Azotobacter species.

### **PRODUCT SUMMARY AND EXPLANATION**

Bacteria of the family Azotobacteraceae constitute the majority of heterotrophic free-living nitrogen fixing bacteria. *Azotobacter* is a genus of free-living diazotrophic bacteria which have the highest metabolic rate compared to any other microorganisms. *Azotobacter* have generated a good deal of interest in the scientific community because of their unique mode of metabolism, by which they can fix nitrogen aerobically.

Azotobacter Medium (Mannitol) is used for isolation and cultivation of mannitol positive *Azotobacter* species from soil. It is also useful for maintenance of *Azotobacter* species by adding extra 1% Mannitol to the medium as specified by the American Type Culture Collection.

### COMPOSITION

Ingredients	Gms / Ltr		
Mono Potassium phosphate	0.1		
Calcium chloride.2H2O	0.1		
Magnesium sulphate.7H2O	0.1		
Calcium Carbonate	5.000		
Mannitol	5.000		
Agar	15.000		
Sodium Molybdate .2H2O	0.005		
Di Potassium Phosphate	0.9		
Ferrous sulphate.7H2O	0.01		
Glucose	5.000		

#### PRINCIPLE

Azotobacter medium (mannitol) is composed of mannitol and glucose, a carbon source for *Azotobacter*. Dipotassium phosphate, Magnesium sulphate and ferrous sulphate provide essential ions for bacterial growth while sodium chloride maintains osmotic equilibrium of media. Calcium carbonate acts as buffer to neutralize the acid produced by the strain. Sodium Molybdate .2H2O role is to fix nitrogen which is important for microorganisms. The actual physiological function of calcium is unknown Nitrogen as the sole nitrogen source in the air. Phosphates are used as buffering agents. Magnesium presumably functions as an activator of phosphorylation, and may therefore be active at several different stages in the respiration. Equal concentrations of calcium are necessary for growth with free nitrogen, nitrate, ammonia, and asparagine.

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### **INSTRUCTION FOR USE**

- Dissolve 26.4 grams in 1000 ml distilled water.
- Heat to boiling to dissolve the medium completely.
- Sterilize by autoclaving at 15 psi pressure (121°C) for 15 minutes.
- Cool to 45-50°C



If slight precipitate occurs after autoclaving, distribute it evenly before pouring into sterile Petri plates.

QUALITY CONTROL SPECIFICATIONS						
Appearance of Powder	: Off white to beige color, homogenous free flowing powder					
pearance of prepared medium	: Yellow coloured, clearly to slightly opalescent gel with precipitate forms in Petri- dishes					
pH (at 25°C)	: 7.3					

# **INTERPRETATION**

Cultural characteristics observed after incubation.

Microorganism	ATCC	Inoculum (CFU/ml)	Growth	Recovery	Incubation Temperature	Incubation Period
Azotobacter beijerinckii	12981	50-100	Good - luxuriant	>=50 %	25-30°C	24-48 Hours
Azotobacter nigricans	35009	50-100	Good - luxuriant	>=50 %	25-30°C	24-48 Hours

### PACKAGING:

In pack size of 500 gm bottles.

## **STORAGE**

Dehydrated powder, hygroscopic in nature, store in a dry place, in tightly-sealed containers below 25°C and protect from direct sunlight. Under optimal conditions, the medium has a shelf life of 4 years. When the container is opened for the first time, note the time and date on the label space provided on the container. After the desired amount of medium has been taken out replace the cap tightly to protect from hydration.

Product Deterioration: Do not use if they show evidence of microbial contamination, discoloration, drying or any other signs of deterioration.

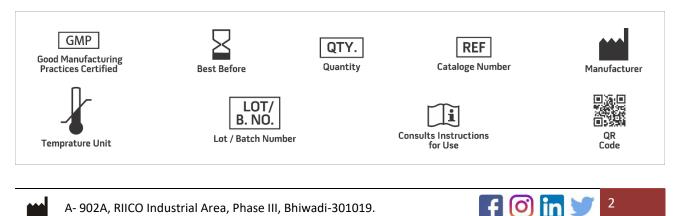
### DISPOSAL

After use, prepared plates, specimen/sample containers and other contaminated materials must be sterilized before discarding.

### REFERENCES

1. ATCC Catalogue of Bacteria and Bacteriophages, 1992, 18th Ed, American Type Culture Collection, Rockville, MD.

- 2. Pelczar M. Jr., 1957, Manual of Microbiological Methods.
- 3. Subba Rao N. S., 1977, Soil Microorganisms and Plant Growth, Oxford and IBH Publishing Co., New Delhi



A- 902A, RIICO Industrial Area, Phase III, Bhiwadi-301019.

# **PRODUCT DATA SHEET**



NOTE: Please consult the Material Safety Data Sheet for information regarding hazards and safe handling Practices. \*For Lab Use Only Revision: 08 Nov., 2019

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