

# **TM 838 – R-3A AGAR**

#### **INTENDED USE**

For sub culturing of microorganisms recovered on less nutritive R-2A Agar from potable water.

#### PRODUCT SUMMARY AND EXPLANATION

R-3A Agar is slightly more nutritious than R-2A Agar and is used for sub culturing the isolates obtained on the less nutritive R-2A Agar.

R-2A Agar is recommended by APHA for estimating the heterotrophic plate count by the pour plate, spread plate or membrane filter procedure. R-2A Agar is formulated as per Reasoner and Geldreich. Stressed or injured organisms during water treatment are unable to grow on high nutrient media, since the faster growing organisms outgrow the former. Therefore, the use of a low nutrient medium like R-2A Agar incubated for longer incubation periods allows these stressed organisms to grow well.

#### **COMPOSITION**

Ingredients	Gms / Ltr	
Casein Acid Hydrolysate	1.000	
Yeast extract	1.000	
Biopeptone	1.000	
Dextrose	1.000	
Starch soluble	1.000	
Dipotassium phosphate	0.600	
Magnesium sulphate	0.048	
Sodium pyruvate	0.600	
Agar	15.000	

# **PRINCIPLE**

This medium consists of Biopeptone, casein acid hydrolysate and yeast extract which provide nitrogen, vitamins, amino acids, carbon and minerals. Dextrose serves as an energy source. Soluble starch aids in the recovery of injured organisms by absorbing toxic metabolic byproducts while sodium pyruvate increases the recovery of stressed cells. Magnesium sulphate is a source of divalent cations and sulphate. Dipotassium phosphate is used to balance the pH of the medium.

# **INSTRUCTION FOR USE**

- Dissolve 21.25 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 min. DO NOT OVERHEAT.
- Mix well and pour into sterile Petri plates.

## **QUALITY CONTROL SPECIFICATIONS**















**Appearance of Powder** : Cream to yellow homogeneous free flowing powder.

Appearance of prepared medium : Light yellow coloured clear to slightly opalescent gel forms in Petri plates.

pH (at 25°C) : 7.2 ± 0.2

#### **INTERPRETATION**

Cultural characteristics observed after incubation. (In case of water samples from fields it is suggested to incubate further for upto 7 days to ascertain the absence of organisms).

Microorganism	ATCC	Inoculum (CFU/ml)	Growth	Recovery	Incubation Temperature	Incubation Period
Candida albicans	10231	10-100	Good-luxuriant	>=50%	35-37°C	24-72 Hours
Escherichia coli	25922	50-100	Good-luxuriant	>=50%	35-37°C	24-72 Hours
Salmonella Enteritidis	13076	50-100	Good-luxuriant	>=50%	35-37°C	24-72 Hours
Enterococcus faecalis	29212	50-100	Good-luxuriant	>=50%	35-37°C	24-72 Hours
Salmonella Typhi	6539	50-100	Good-luxuriant	>=50%	35-37°C	24-72 Hours

### **PACKAGING:**

In pack size of 500 gm bottles.

## **STORAGE**

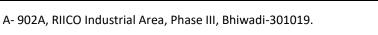
Dehydrated powder, hygroscopic in nature, store in a dry place, in tightly-sealed containers between 25-30°C and protect from direct sunlight. Under optimal conditions, the medium has a shelf life of 4 years. When the container is opened for the first time, note the time and date on the label space provided 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**





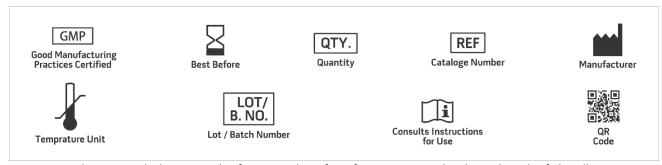








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- 2. Downes F. P. and Ito K., (Eds.), Compendium of Methods for the Microbiological Examination of Foods, 4th Ed., American Public Health Association, Washington, D.C.
- 3. Reasoner D. J. and Geldreich E. E., 1985, Appl. Environ. Microbiol., 49:1.
- 4. Collins V. J. and Willoughby J. G., 1962, Arch. Microbiol., 43:294.
- 5. Atlas R. M., 2004, Handbook of Microbiological Media, Lawrence C. Parks, (Ed.), 3rd Edition, CRC Press.



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

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