

TM 2299 – R2A AGAR, MODIFIED

INTENDED USE

For the enumeration and cultivation of bacteria from potable water.

PRODUCT SUMMARY AND EXPLANATION

This medium is recommended in standard methods for pour plate, spread plate and membrane filter methods for heterotrophic plate count. It was developed by Reasoner and Geldreich for bacterial plate counts of treated potable water. The HPC, heterotrophic plate count formerly known as the standard plate count is a procedure for estimating the number of live bacteria in water and measuring changes during water treatment in distribution systems or in swimming pools. The use of low nutrient media favours growth of injured or stressed organisms at longer incubation periods as compared to the use of high nutrient media. As compared to Tryptone Glucose Agar or Standard methods agar, R2A agar has been reported to give improved recovery of stress and chlorine tolerant bacteria from drinking water systems.

COMPOSITION

Ingredients	Gms / Ltr
Casein Enzymic hydrolysate	0.250
Peptic digest of animal tissue	0.250
Casein Acid Hydrolysate	0.500
Yeast extract	0.500
Glucose	0.500
Starch soluble	0.500
Dipotassium phosphate	0.030
Magnesium sulphate, heptahydrate	0.500
Sodium pyruvate	0.030
Agar	15.000

PRINCIPLE

This medium consists of Enzymic digest of casein, enzymatic digest of animal tissue, casein acid hydrolysate and yeast extract which provides necessary nitrogen sources, carbohydrates, vitamins, minerals and growth factors to growing organisms. Dextrose serves as carbon source, Soluble starch aids in recovery of injured organisms toxic metabolic byproducts while sodium pyruvate increases recovery of stressed cells. Magnesium sulphate is a source of divalent cations and sulphate. Dipotassium phosphate is used to balance the pH of medium. Agar acts as a solidifying agent.

INSTRUCTION FOR USE

- Dissolve 18.12 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.

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
<i>Candida albicans</i>	10231	10-100	Good-luxuriant	≥50%	35-37°C	24-72 Hours
<i>Escherichia coli</i>	25922	50-100	Good-luxuriant	≥50%	35-37°C	24-72 Hours
<i>Salmonella</i> Enteritidis	13076	50-100	Good-luxuriant	≥50%	35-37°C	24-72 Hours
<i>Staphylococcus aureus</i>	25923	50-100	Good-luxuriant	≥50%	35-37°C	24-72 Hours
<i>Enterococcus faecalis</i>	29212	50-100	Good-luxuriant	≥50%	35-37°C	24-72 Hours
<i>Salmonella</i> Typhi	6539	50-100	Good-luxuriant	≥50%	35-37°C	24-72 Hours
<i>Escherichia coli</i>	8739	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

1. Eaton, A.D., L.S.Clesceri, and A.E. Greenberg (eds.), 1995, Standard Methods for the examination of water and wastewater, 19th ed. American Public Health Association, Washington, D.C.
2. Reasoner, D. J and Geldreich, E.E ,1979, A new medium for the enumeration and subculture of bacteria form potable water. Abstracts of the Annual meeting of the American Society for microbiology 79th Meeting, Paper No. N7.
3. Fiksdal, L., E.A. Vik, A. Mills, and T. Staley, 1982, Non-standard methods for enumerating bacteria in drinking water. Journal AWWA, 74: 313-318.
4. Kelly, A.J., C.A. Justice, and L.A. Nagy, 1983, Predominance of chlorine tolerant bacteria in drinking water systems. Abstracts of the Annual meeting of the American Society for Microbiology 79th Meeting paper No. Q122.
5. Means E.G., L. Hanami, H.F. Ridgway, and B.H. Olson, 1981, Evaluating mediums and plating techniques for enumerating bacteria in water distribution systems. Journal AWWA 53: 585-590.
6. VanSoestberger, A.A., and C.H. Lee. 1969 Appl. Microbiol. 18: 1092.
7. Klein D.A. and S. Wu. (1974). Appl. Microbiol. 27: 429.

 Good Manufacturing Practices Certified	 Best Before	 Quantity	 Cataloge Number	 Manufacturer
 Temprature Unit	 Lot / Batch Number	 Consults Instructions for Use	 QR Code	

NOTE: Please consult the Material Safety Data Sheet for information regarding hazards and safe handling Practices.

***For Lab Use Only**
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