

TECHNICAL DATA SHEET

R-2A Broth

Principle

R2A broth was developed by Reasoner and Geldreich (1979) for bacteriological plate counts of treated potable water. It is a low nutrient medium, in combination with a lower incubation temperature and longer incubation time stimulates the growth of stressed and chlorine-tolerant bacteria. Media is composed of yeast extract, proteose peptone, casamino acids, dextrose, starch, sodium pyruvate, dipotassium phosphate, and magnesium sulfate. Yeast extract provides nitrogen, carbon and vitamins. Proteose peptone and casamino acids provide nitrogen, amino acids, carbon and minerals. Dextrose is a source of carbon. Starch absorbs toxic metabolites and help in the recovery of injured organisms. Sodium Pyruvate increases the recovery of stressed cells. Potassium phosphate is the source of phosphate and balance the pH. Magnesium sulphate is a source of sulphate and divalent cations.

Use: For cultivation and maintenance of heterotrophic bacteria from potable waters.

Contents*

Ingredients	Gram/Liter
Yeast Extract	0.500
Proteose Peptone	0.500
Casamino Acids	0.500
Dextrose	0.500
Starch	0.500
Sodium Pyruvate	0.300
Dipotassium Phosphate	0.300
Magnesium Sulfate	0.024
pH at 25°C	7.2 ±0.2

* Formula adjusted for optimum performance and parameters

Directions: Dissolve 3.12 grams in 1000 ml distilled water. Boil to dissolve the medium completely and sterilize by autoclaving at 15 lbs pressure (121°C) for 15 min, cool it to 42-45 °C and inoculate test sample aseptically.

Specimens types analyzed

Portable water samples etc.

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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	Light beige colored free flowing, homogeneous powder
Reaction of 0.31% solution	7.2 ±0.2 at 25 °C
pH	7.00- 7.40
Color and clarity of ready medium	Light amber colored, opalescent solution
Growth Promotion properties	Best at 50-100 CFU at 32-37°C for 18-72 hours for bacteria and 23-27°C for 3-5 days for yeast and molds.
Indicative properties	Optimum at 50-100 CFU at 32-37 °C for 18-48 hours for bacteria and 23-27°C for 3-5 days for yeast and molds.
Negative control	Performed using sterile distilled water

Different Microbial Response: Growth observed after incubation at 33-37°C for 24-72 hours for bacteria and 23-27°C for 3-5 days for yeast and molds.

Organism	ATCC	Inoculum (CFU)	Growth
<i>Pseudomonas aeruginosa</i>	27853	50-100	Luxuriant
<i>Escherichia coli</i>	8739	50-100	Luxuriant
<i>Salmonella typhimurium</i>	14028	50-100	Luxuriant
<i>Staphylococcus aureus</i>	25923	50-100	Luxuriant
<i>Bacillus spizizenii</i>	6633	50-100	Luxuriant
<i>Enterococcus faecalis</i>	14506	50-100	Luxuriant
<i>Candida albicans</i>	10231	50-100	Luxuriant
<i>Aspergillus brasiliensis</i>	16404	50-100	Luxuriant

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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. Atlas, R. M. (2005). Handbook of media for environmental microbiology. CRC press.
2. Difco Manual (1998). 11th Edition. Difco Laboratories., Division of Becton Dickinson and Company, Sparks, Maryland, USA.
3. Rand, M. C., Arnold E. Greenberg, and Michael J. Taras, (1976), Standard methods for the examination of water and wastewater. Prepared and published jointly by American Public Health Association, American Water Works Association, and Water Pollution Control Federation.
4. Salfinger Y., and Tortorello M.L. Fifth (Ed.), (2015), Compendium of Methods for the Microbiological Examination of Foods, 5th Ed., American Public Health Association, Washington, D.C.

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