



R2A Agar (i23153)

For use in standard methods for pour plate, spread plate, and membrane filter analysis to enumerate heterotrophic bacteria from water.

Industry: Pharmaceutical / Veterinary / Water / Quality Control / Clinical

Principles & Uses

R2A Agar is a recommended medium for heterotrophic plate counts, particularly in drinking water analysis. This medium, developed by Reasoner and Geldreich, is designed to recover stressed and chlorine-tolerant bacteria, offering advantages over high-nutrient media like Tryptone Glucose Agar. It's ideal for estimating live bacteria in water, monitoring changes during water treatment, and is widely used in pour plate, spread plate, and membrane filter methods.

R2A Agar's formulation includes various components serving distinct roles. Yeast extract and peptones provide essential nutrients for bacterial growth. Glucose acts as a carbon source, while soluble starch helps recover injured organisms by absorbing toxic byproducts. Sodium pyruvate aids in the recovery of stressed cells, and magnesium sulfate contributes divalent cations and sulfate. Dipotassium phosphate helps balance the medium's pH. Agar serves as the solidifying agent.

This medium's low-nutrient content, combined with a lower incubation temperature and extended incubation time, promotes the recovery of slow-growing, stressed and chlorine-resistant bacteria in drinking water. Its usage aligns with standard methods for water examination, and it's recommended in pharmaceutical applications as well, especially for microbial monitoring during production and storage of medicines and various purified water types.

Composition (gr/L)

Peptones 1, Yeast Extract 0.5, Glucose 0.5, Soluble Starch 0.5, K_2HPO_4 0.3, Sodium Pyruvate 0.3, Magnesium Sulphate 0.05, Agar 12. Final pH at 25°C 7.2 ± 0.2

Preparation from dehydrated Powder

Suspend 15.2 g of the medium in 1 Liter of purified water. Mix thoroughly. Autoclave at 121°C for 15 min, cool to 45–50°C and pour into sterile Petri dishes. **NOTE:** To use R2A plates in streaking methods, add 3 g/L of additional agar to the powder.

Quality Control

Dehydrated Appearance: Light beige, free-flowing, homogeneous.

Prepared Appearance: Light to medium amber, clear to very slightly opalescent.

Reaction of 1.52% Solution at 25°C: pH 7.2 ± 0.2

Cultural Response

Cultural response was observed after 42-48 hours of incubation at $35 \pm 2^{\circ}$ C.

Organism (ATCC*)	Recovery
Enterococcus faecalis (29212)	Good
Escherichia coli (25922)	Good
Staphylococcus aureus (25923)	Good
Pseudomonas aeruginosa (27853)	Good

*ATCC is a registered trade mark of the American Type Culture Collection.





E. faecalis on R2A agar using the streaking method. The background has been darkened to enhance colony visibility.

Storage

Keep the container at 15-30 °C. Store prepared medium at 2-8 °C.