

LACTOBACILLI MRS AGAR (7543)

Intended Use

Lactobacilli MRS Agar is used for the cultivation of lactobacilli.

Product Summary and Explanation

Lactobacilli MRS Agar is based on the formulations of deMan, Rogosa and Sharpe (MRS).¹ This medium supports luxuriant growth of lactobacilli from oral, fecal, dairy, and other sources.

Principles of the Procedure

Enzymatic Digest of Animal Tissue, Beef Extract, and Yeast Extract are the carbon, nitrogen, and vitamin sources used to satisfy general growth requirements in Lactobacilli MRS Broth. Dextrose is the fermentable carbohydrate. Sodium Acetate is an inhibitory agent. Sodium Acetate and Ammonium Citrate act as selective agents as well as energy sources. Potassium Phosphate is the buffering agent. Magnesium Sulfate and Manganese Sulfate provide cations used in metabolism. Polysorbate 80 is a surfactant, facilitating uptake of nutrients by lactobacilli. Agar is the solidifying agent.

Formula / Liter

| | |
|--|--------|
| Enzymatic Digest of Animal Tissue..... | 10 g |
| Beef Extract | 10 g |
| Yeast Extract..... | 5 g |
| Dextrose..... | 20 g |
| Sodium Acetate..... | 5 g |
| Polysorbate 80 | 1 g |
| Potassium Phosphate | 2 g |
| Ammonium Citrate | 2 g |
| Magnesium Sulfate | 0.1 g |
| Manganese Sulfate | 0.05 g |
| Agar | 15 g |

Final pH: 6.5 ± 0.2 at 25°C

Formula may be adjusted and/or supplemented as required to meet performance specifications.

Precaution

1. For Laboratory Use.

Directions

1. Suspend 70 g of the medium in one liter of purified water.
2. Heat with frequent agitation and boil for one minute to completely dissolve the medium.
3. Autoclave at 121°C for 15 minutes.

Quality Control Specifications

Dehydrated Appearance: Powder is homogeneous with soft lumps and yellow-tan.

Prepared Appearance: Prepared medium is light beige.

Expected Cultural Response: Cultural response on Lactobacillus MRS Agar at 35°C after 24 - 96 hours incubation.

| Microorganism | Response |
|---|----------|
| <i>Lactobacillus casei</i> ATCC® 393 | growth |
| <i>Lactobacillus fermentum</i> ATCC® 9338 | growth |
| <i>Lactobacillus plantarum</i> ATCC® 8014 | growth |

The organisms listed are the minimum that should be used for quality control testing.

Test Procedure

1. To obtain direct counts of lactobacilli, pour 15 - 20 mL sterile, molten (45 - 50°C) Lactobacilli MRS Agar into sterile petri dishes containing 1 mL volumes of diluted test sample.
2. Distribute inoculum throughout medium by rotating the plate in one direction, then in the reverse direction.
3. Allow medium to solidify on a flat surface for 5 - 10 minutes.
4. Alternatively, plates of Lactobacilli MRS Agar can be used for direct recovery of organisms using the streak inoculation technique.
5. Incubate agar plates at 35°C for 3 days, or at 30°C for 5 days in an aerobic atmosphere supplemented with carbon dioxide.

Results

Lactobacilli appear as large, white colonies embedded in or on Lactobacilli MRS Agar. Growth can be subcultured onto appropriate media for use in additional procedures. Refer to appropriate references for recommendation on the identification of *Lactobacillus* spp.²⁻⁴

Storage

Store sealed bottle containing the dehydrated medium at 2 - 8°C. Once opened and recapped, place container in a low humidity environment at the same storage temperature. Protect from moisture and light by keeping container tightly closed.

Expiration

Refer to expiration date stamped on the container. The dehydrated medium should be discarded if not free flowing, or if appearance has changed from the original color. Expiry applies to medium in its intact container when stored as directed.

Limitations of the Procedure

1. Due to varying nutritional requirements, some strains may be encountered that grow poorly or fail to grow on this medium.
2. Organisms other than lactobacilli may grow in this medium. Isolates must be confirmed as lactobacilli by appropriate biochemical testing.

Packaging

| | | | |
|------------------------------|-----------------|--------------|--------------|
| Lactobacilli MRS Agar | Code No. | 7543A | 500 g |
| | | 7543B | 2 kg |
| | | 7543C | 10 kg |

References

1. **deMan, J. C., M. Rogosa, and M. E. Sharpe.** 1960. A medium for the cultivation of lactobacilli. *J. Bacteriol.* **23**:130.
2. **MacFaddin, J. F.** 1985. Media for the isolation-cultivation-identification-maintenance of medical bacteria, vol. 1 Williams & Wilkins, Baltimore, MD.
3. **Vanderzant, C. and D. F. Splittstoesser (eds.)**. Compendium of methods for the microbiological examination of foods, 3rd ed. American Public Health Association, Washington, D.C.
4. **Murray, P. R., E. J. Baron, M. A. Pfaller, F. C. Tenover, and R. H. Tenover (eds.)**. 1995. Manual of clinical microbiology, 6th ed. American Society for Microbiology, Washington, D.C.

Technical Information

Contact Acumedia Manufacturers, Inc. for Technical Service or questions involving dehydrated culture media preparation or performance at (410)780-5120 or fax us at (410)780-5470.