



Technical Data

MRS Agar w/pH 5.5

M1923

Intended Use:

Recommended for the enrichment, cultivation and isolation of *Lactobacillus* species.

Composition**

Ingredients	Gms / Litre
Casitose ▲	10.000
HM extract#	10.000
Yeast extract	4.000
Dextrose (Glucose)	20.000
Dipotassium hydrogen phosphate	2.000
Polysorbate 80 (Tween 80)	1.000
Diammonium hydrogen citrate	2.000
Sodium acetate	5.000
Magnesium sulfate	0.200
Manganese sulphate	0.040
Agar	14.000
Final pH (at 25°C)	5.5±0.2

**Formula adjusted, standardized to suit performance parameters

▲- Equivalent to Casein peptone

- Equivalent to Meat extract

Directions

Suspend 68.24 grams in 1000 ml purified / distilled water. Heat to boiling to dissolve the medium completely. Sterilize by autoclaving at 15 lbs pressure (121°C) for 15 minutes. Mix well and pour into sterile Petri plates.

Principle And Interpretation

MRS Agar w/pH 5.5 is based on the formulation of deMan, Rogosa and Sharpe (1) with slight modification. It supports growth of all Lactobacilli from all types of materials for eg Brewery industry.

Lactobacilli can cause spoilage in foods due to improper storage conditions, temperature, improper hygiene, cleaning. *Lactobacillus* is acidophilic and mostly grows on acidic foods. The pH of foods can be effective against activity of pathogens.

Lactobacilli commonly cause spoilage of meats and fermented foods.

Casitose and HM extract supply nitrogenous and carbonaceous compounds. Yeast extract provides vitamin B complex and glucose is the fermentable carbohydrate and energy source. Tween 80 supplies fatty acids required for the metabolism of Lactobacilli. Sodium acetate and Diammonium hydrogen citrate inhibit Streptococci, moulds and many other microorganisms. Magnesium sulphate and manganese sulphate provide essential ions for multiplication of lactobacilli. Phosphates provide good buffering action in the media.

Lactobacilli are microaerophilic and generally require layer plates for aerobic cultivation on solid media. When the medium is set, another layer of un-inoculated MRS Agar is poured over the surface to produce a layer plate (2). Lactobacilli isolated on MRS Agar w/pH 5.5 should be further confirmed biochemically.

Type of specimen

Food and dairy samples - Meat, Fermented foods, Brewery, Milk and milk products, etc.

Specimen Collection and Handling

For food and dairy samples, follow appropriate techniques for sample collection and processing as per guidelines (1,6,7). After use, contaminated materials must be sterilized by autoclaving before discarding.

Warning and Precautions

Read the label before opening the container. Wear protective gloves/protective clothing/eye protection/ face protection. Follow good microbiological lab practices while handling specimens and culture. Standard precautions as per established guidelines should be followed while handling specimens. Safety guidelines may be referred in individual safety data sheets.

Limitations

1. Further biochemical and serological test must be carried out for further identification.
2. Some species may show poor growth due to nutritional variations.

Performance and Evaluation

Performance of the medium is expected when used as per the direction on the label within the expiry period when stored at recommended temperature.

Quality Control

Appearance

Cream to light yellow homogeneous free flowing powder

Gelling

Firm, comparable with 1.4% Agar gel.

Colour and Clarity of prepared medium

Medium to dark amber coloured, clear to slightly opalescent gel forms in Petri plates

Reaction

Reaction of 6.71% w/v aqueous solution at 25°C. pH : 5.5±0.2

pH

5.30-5.70

Cultural Response

Cultural characteristics observed after an incubation at 35-37°C for 18-24 hours or longer.(with 5% CO₂)

Organism	Growth	Inoculum (CFU)	Recovery
<i>Lactobacillus fermentum</i> ATCC 9338	luxuriant	50-100	≥50%
<i>Lactobacillus leichmannii</i> ATCC 7830	luxuriant	50-100	≥50%
<i>Lactobacillus plantarum</i> ATCC 8014	luxuriant	50-100	≥50%
<i>Lactobacillus casei</i> ATCC 9595	luxuriant	50-100	≥50%

Storage and Shelf Life

Store dehydrated and the prepared medium at 2-8°C. Use before expiry date on the label. On opening, product should be properly stored dry, after tightly capping the bottle in order to prevent lump formation due to the hygroscopic nature of the product. Improper storage of the product may lead to lump formation. Store in dry ventilated area protected from extremes of temperature and sources of ignition Seal the container tightly after use. Product performance is best if used within stated expiry period.

Disposal

User must ensure safe disposal by autoclaving and/or incineration of used or unusable preparations of this product. Follow established laboratory procedures in disposing of infectious materials and material that comes into contact with sample must be decontaminated and disposed of in accordance with current laboratory techniques (3,4).

Reference

1. American Public Health Association, Standard Methods for the Examination of Dairy Products, 1978, 14th Ed., Washington D.C.
2. deMan J., Rogosa M. and Sharpe M., 1960, J. Appl. Bacteriol., 23:130.
3. Isenberg, H.D. Clinical Microbiology Procedures Handbook 2nd Edition.
4. Jorgensen, J.H., Pfaller, M.A., Carroll, K.C., Funke, G., Landry, M.L., Richter, S.S and Warnock, D.W. (2015) Manual of Clinical Microbiology, 11th Edition. Vol. 1.
5. MacFaddin J., 1985, Media for Isolation-Cultivation-Identification-Maintenance of Medical Bacteria, Vol.1, Williams and Wilkins, Baltimore.
6. 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.
7. Wehr H. M. and Frank J. H., 2004, Standard Methods for the Microbiological Examination of Dairy Products, 17th Ed., APHA Inc., Washington, D.C.

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