



MRS Agar w/low pH

M1927

Recommended for cultivation of all *Lactobacillus* species from all types of materials.

Composition**

Ingredients	Gms / Litre
Meat peptone	10.000
Meat extract	10.000
Yeast extract	5.000
Diammonium citrate	2.000
Dipotassium hydrogen phosphate	2.000
Glucose	20.000
Magnesium sulphate,heptahydrate	0.200
Manganous sulphate,tetrahydrate	0.050
Sodium acetate trihydrate	5.000
Agar	12.000
Final pH (at 25°C)	5.4±0.2

**Formula adjusted, standardized to suit performance parameters

Directions

Suspend 64.15(the equivalent weight of dehydrated medium per litre) grams in 1000 ml distilled water. Heat to boiling to dissolve the medium completely. Sterilize by autoclaving at 15lbs pressure (121°C) for 15 minutes.Cool to 45-50°C Mix well and pour into sterile Petri plates.

Principle And Interpretation

Lactobacilli MRS medium is based on the formulation of deMan, Rogosa and Sharpe (1) with slight modification. It supports luxuriant growth of all Lactobacilli from oral cavity (1), dairy products (2), foods (3), faeces (4) and other sources (5).

Proteose peptone and beef extract supply nitrogenous and carbonaceous compounds. Yeast extract provides vitamin B complex and dextrose is the fermentable carbohydrate and energy source. Polysorbate 80 supplies fatty acids required for the metabolism of Lactobacilli. Sodium acetate and ammonium 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 (5). Lactobacilli isolated on MRS Agar should be further confirmed biochemically.

Quality Control

Appearance

Cream to light yellow homogeneous free flowing powder

Gelling

Firm, comparable with 1.2% 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 6.42% w/v aqueous solution at 25°C. pH : 5.4±0.2

pH

5.20-5.60

Cultural Response

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

Cultural Response

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

Storage and Shelf Life

Store dehydrated and the prepared medium at 2-8°C in tightly closed container. Use before expiry date on the label.

Reference

1. deMan J., Rogosa M. and Sharpe M., 1960, J. Appl. Bacteriol., 23:130.
2. Marshall R.T. (Ed.), 1992, Standard Methods for the Examination of Dairy Products, 16th ed., APHA, Washington, D.C.
3. Downes F. P. and Ito K., (Eds.), 2001, Compendium of Methods For the Microbiological Examination of Foods, 4th Ed., APHA, Washington, D.C.
4. Sabine and Vaselekos, 1965, Nature, 206:960.
5. MacFaddin J., 1985, Media for Isolation-Cultivation-Identification -Maintenance of Medical Bacteria, Vol.1, Williams and Wilkins, Baltimore.

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