

Specification

Solid culture medium for detection, isolation and cultivation of lactobacilli and other lactic acid bacteria from food and beverages according to de Man, Rogosa and Sharpe.

Presentation

20 Prepared Plates
90 mm
with: 21 ± 2 ml

Packaging Details

1 box with 2 packs of 10 plates/pack. Single cellophane.

Shelf Life

3 months

Storage

2-14 °C

Composition

Composition (g/l):

Proteose peptone.....	10.0
Meat extract.....	8.00
Yeast extract.....	4.00
D-(+)-Glucose.....	20.0
Sodium acetate.....	5.00
Magnesium sulfate.....	0.20
Manganese sulfate.....	0.05
Dipotassium phosphate.....	2.00
Triammonium citrate.....	2.00
Polysorbate 80.....	1.00
Agar.....	14.0

Description /Technique

Description:

MRS Agar is a medium used for the cultivation of lactobacilli. It is a modification of a medium based on the highly nutritious properties of tomato juice. The addition of magnesium, manganese and acetate, together with polysorbate, provides an improved medium for the growth of lactobacilli, including very fastidious species such as *Lactobacillus brevis* and *Lactobacillus fermentum*.

The quality of the peptones in addition to the meat and yeast extracts, combine all the necessary growth factors that make MRS medium one of the best media for the cultivation of lactobacilli. As the selectivity of this medium is low and contaminants tend to grow subculturing in a (double layer) solid medium, and then in broth is recommended to increase selectivity. In many cases, growth is encouraged by incubation in a CO₂ enriched atmosphere.

MRS medium is particularly recommended for the enumeration and maintenance of lactobacilli either by the MPN technique in broth, or by inoculation on a plate, overlaying it with a second layer of molten medium. This technique overcomes the need for a CO₂ enriched atmosphere.

Technique:

Collect, dilute and prepare samples and volumes as required according to specifications, directives, official standard regulations and/or expected results.

Spread the plate by streaking methodology or by spiral method. Incubate the plates right side up in a CO₂ atmosphere at $30 \pm 1^\circ\text{C}$ for $72 \pm 3\text{h}$.

(Incubation times longer than those mentioned above or different incubation temperatures may be required depending on the sample, on the specifications,...

This medium can be inoculated directly or after enrichment broth like MRS broth) Incubated under microaerophilic conditions to promote lactobacilli enrichment.

After incubation, enumerate all the colonies that have appeared onto the surface of the agar.

Each laboratory must evaluate the results according to their specifications.

Calculate total microbial count per ml of sample by multiplying the average number of colonies per plate by inverse dilution factor if streaked a diluted sample. Report results as Colony

Forming Unit (CFU's) per ml or g along with incubation time and temperature.

Quality control**Physical/Chemical control**

Color : Yellowish-brown

pH: 6.2 ± 0.2 at 25°C**Microbiological control**Inoculate: Practical range 100 ± 20 CFU. min. 50 CFU (productivity)/ 10^3 - 10^4 (qualitative selectivity).

Analytical methodology according to ISO 11133:2014/A1:2018; A2:2020.

Anaerobiosi. Incubation at 30 ± 1 °C for 72 ± 3 h

Microbiological control according to ISO 11133:2014/A1:2018.

Microorganism*Escherichia coli* ATCC® 25922, WDCM 00013*Lactobacillus sakei* ATCC® 15521*Lactococcus lactis* ATCC® 19435*Pediococcus pentosaceus* ATCC® 33316**Growth**

Poor to good

Good ($\geq 70\%$)Good ($\geq 70\%$)Good ($\geq 70\%$)**Sterility Control**

Incubation 48 h at 30-35 °C and 48 h at 20-25 °C: NO GROWTH.

Check at 7 days after incubation in same conditions.

Bibliography

- ATLAS, R.M. & L.C. PARKS (1993) Handbook of Microbiological Culture Media. CRC Press. BocaRaton, Fla. USA
- CORRY, J.E.L., G.D.W. CURTIS & R.M. BAIRD, Eds. (2003) Handbook of Culture Media for Food Microbiology. Elsevier Science B.V. Amsterdam
- DOWNES, F.P. & K. ITO (2001) Compendium of Methods for the Microbiological Examination of Foods. 4th ed. APHA. Washington DC., USA
- LAWRENCE, D.R. & P.A. LEEDHAM (1979). The detection of acid lactic bacteria. J. Int. Brew. 85:119-121
- ISO Standard 11133:2014 Microbiology of food, animal feed and water. Preparation, production, storage, and performance testing of culture media.
- McFADDIN, J. (1985) Media for the isolation-cultivation-identification-maintenance of medical bacteria. Vol. I. William & Wilkins. Baltimore. USA
- MAN, J.C. de, ROGOSA, M. y SHARPE, M. Elisabeth (1960) A medium for the cultivation of lactobacilli. J. Appl. Bact.; 23:130.
- SMITH, C.E., G.P. CASEY & W.M. INGLEDEW (1987). The use and understanding of media used in Brewing Microbiology. - Update 1987 - Brewer's Digest 62(10)12-16, 43.
- VAN KEER, C., L. van MELKEBEKE, W. VERTRIST, G. HOOZEE & E. Van SCHOONENBERGHE (1983) Growth of Lactobacillus species on different media. J. Inst. Brew. 89:361-363.