# MRS Broth Art. No. 02-135

#### Specification

Liquid culture medium for the isolation of lactobacilli, according to de Man, Rogosa and Sharpe and ISO standards.

## Formula\* in g/L

Proteose peptone	
Meat extract	
Yeast extract	4,00
D(+)-Glucose	
Sodium acetate	5,00
Triammonium citrate	
Magnesium sulfate	0,20
Manganese sulfate	
Dipotassium phosphate	2,00
Polysorbate 80	1,00
Final pH 6,2 ± 0,2 at 25°C	

\* Adjusted and /or supplemented as required to meet performance criteria

#### Directions

Suspend 52 g of powder in 1 L of distilled water. Heat to dissolve completely and dispense into suitable containers. Sterilize in the autoclave at 121°C for 15 minutes. **Do not overheat**.

#### Description

MRS Agar and Broth are media for the cultivation of lactobacilli, they are a modification of a medium based on the highly nutritious properties of tomato juice. The addition of magnesium, manganese and acetate, together with Polysorbate, has provided an improved medium for the growth of lactobacilli, including that of 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 the medium is low and contaminants tend to grow subculturing in a (double layer) solid medium and then in broth is recommended to improve selectivity. In many cases, growth is encouraged by incubation in a  $CO_2$  enriched atmosphere.

MRS media is particularly recommended for the enumeration and maintenance of lactobacilli either by the MPN technique (in broth) or on a plate by inoculation on a plate, overlaying it with a second layer of molten medium. This technique overcomes the need for a  $CO_2$  enriched atmosphere.

## References

- DOWNES, F.P. & K. ITO (2001) Compendium of Methods for the Microbiological Examination of Foods. 4<sup>th</sup> ed. APHA. Washington. DC. USA.
- FIL-IDF Standard 146 (2003) Yoghurt. Identification of characteristic microorganisms.
- IFU Method No. 5 (1996) Lactic Acid Bacteria Count Procedure. Schweizerischer Obstverband. CH-6302 Zug.
- IFU Method No. 7 (1998) Sterility testing of aseptic filled products, commercial sterile products and preserved products. Schweizerischer Obstverband. CH-6302 Zug.
- IFU Method No. 9 (1998) Microbiological examination of potential spoilage microorganisms of tomato products. Schweizerischer Obstverband. CH-6302 Zug.
- ISO Standard 9232 (2003) Yoghurt Identification of characteristic microorganisms (*Lactobacillus delbrueckii* subsp *bulgaricus* and *Streptococcus thermophilus*).
- ISO/TS 11133-1: 2009. Microbiology of food and animal feeding stuffs.-Guidelines on preparation and production of culture media. Part 1: General guidelines on quality assurance for the preparation of culture media in the laboratory.
- ISO/TS 11133-2: 2003 Corr. 2004. Microbiology of food and animal feeding stuffs.- Guidelines on preparation and production of culture media. Part 2: Practical guidelines on performance testing of culture media.
- ISO Standard 15214 (1998) Horizontal method for the enumeration of mesophilic lactic acid bacteria - Colony count technique at 30°C.
- MAN, J.C. de, ROGOSA, M. & SHARPE, M. Elisabeth (1960) A medium for the cultivation of lactobacilli. J. Appl. Bact.; 23:130.

#### Storage

For laboratory use only. Keep tightly closed, away from bright light, in a cool dry place (+4 $^{\circ}$ C to 30 $^{\circ}$ C and <60 $^{\circ}$ RH).

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## **Quality control**

Incubation temperature: 30°C ± 2,0 Incubation time: 48 h - 3 days Inoculum: 10-100 CFU (Productivity) // 1.000-10.000 CFU (Selectivity). (ISO/TS 11133-1/2)

Microorganism	Growth	Remarks
Escherichia coli ATCC 25922	Partial inhibition	Incubate in a 5% $CO_2$ atmosphere
Lactobacillus sakei ATCC 15521	Good - very good	Incubate in a 5% $CO_2$ atmosphere
Lactobacillus lactis ATCC 19435	Good - very good	Incubate in a 5% CO <sub>2</sub> atmosphere
Lactobacillus fermentum ATCC 9338	Good - very good	Incubate in a 5% CO <sub>2</sub> atmosphere
Lactobacillus acidophilus ATCC 4536	Good - very good	Incubate in a 5% $CO_2$ atmosphere



Left: Lactobacillus fermentum ATCC 9338 Centre: Escherichia coli ATCC 25922 Right: Uninoculated tube (Control)