

TECHNICAL DATA SHEET

MRS BROTH

CULTURE OF *LACTOBACILLUS* AND OTHER LACTIC ACID BACTERIA

1 INTENDED USE

MRS Broth is used for the growth and enumeration of lactobacilli in food products. By transferring isolated colonies, luxuriant subcultures can be obtained. The medium can also be used to culture slowly-growing lactobacilli such as *Lactobacillus brevis* and *Lactobacillus fermentum*.

2 HISTORY

For the culture of lactobacillus, de Man, Rogosa & Sharpe developed in 1960, the composition of a media susceptible to grow lactobacilli used in dairy products without the need to add tomato juice.

3 PRINCIPLES

The different peptones, glucose, manganese and magnesium salts supply the nutritive elements required for the growth of lactobacilli.

Tween 80 is composed of a mixture of oleic esters and is a source of fatty acids essential for the growth of these bacteria.

Dipotassium phosphate stabilizes the pH during bacterial growth.

Ammonium citrate and sodium acetate inhibit the development of most contaminants, including streptococci and molds

4 TYPICAL COMPOSITION

The composition can be adjusted in order to obtain optimal performance.

For 1 liter of media :

- Peptones	20,00 g
- Yeast Extract	5,00 g
- Glucose	20,00 g
- Tween 80 (*)	1,08 g
- Dipotassium phosphate	2,00 g
- Sodium acetate	5,00 g
- Ammonium citrate	2,00 g
- Magnesium sulfate	0,20 g
- Manganese sulfate	0,05 g

pH of the ready-to-use media at 25 °C (BK070) : 6,4 ± 0,2.

5 PREPARATION

- Dissolve 55,3 g of the dehydrated media in 1 liter of distilled or demineralized water.
- Stir slowly until complete dissolution, heating if necessary.
- Dispense in tubes or vials.
- Sterilize in an autoclave at 121 °C for 15 minutes..
- Cool to room temperature.

✓ **Reconstitution :**
55,3 g/L

✓ **Sterilization :**
15 min at 121 °C

6 INSTRUCTIONS FOR USE

- Transfer 1 mL of the product to analyze and its serial dilutions to one or several tubes of medium.
- For industrial fermentation, inoculate the broth culture using the appropriate concentration of starter culture for the strain in question.
- Incubate at 30 °C or at 37 °C, from 48 hours to 5 days according to the microorganisms being studied or the pertinent industrial production protocol.

✓ **Inoculation :**
1 mL

✓ **Incubation :**
48 h to 5 days at
30 or 37 °C

7 RESULTS

Examine the tubes containing characteristic cloudiness of microbial growth. In addition to lactobacilli, *Leuconostoc* and *Pediococcus* may also develop. It is recommended to prepare subcultures on an appropriate media. Depending on the qualitative results obtained, use the most probable number method for enumeration.

Growth in a fermentative context will depend on strain, conditions, material and the specific protocols used for harvesting biomass or yield-dependent criteria.

8 QUALITY CONTROL

Dehydrated media : cream powder, slightly clumped.

Prepared media : amber solution, may contain a slight precipitate.

Typical culture response after 48 hours of incubation at 37 °C, inoculum ≤ 100 microorganisms

Microorganisms		Growth
<i>Lactobacillus delbrueckii</i> subsp. <i>lactis</i>	ATCC® 4797	Positive
<i>Lactobacillus casei</i> subsp. <i>rhamnosus</i>	WDCM 00101	Positive
<i>Lactobacillus plantarum</i>	ATCC 8014	Positive
<i>Lactobacillus fermentum</i>	ATCC 9338	Positive

9 STORAGE / SHELF LIFE

Dehydrated media : 2-20 °C.

The expiration dates are indicated on the labels..

Prepared media in tubes or vials (*) : 180 days at 2-8 °C.

(*) Benchmark value determined under standard preparation conditions, following manufacturer's instructions. .

10 PACKAGING

Dehydrated media :

500 g bottle BK070HA

11 BIBLIOGRAPHY

de Man, J.C., Rogosa, M., and Sharpe, M.E.. 1960. A medium for the cultivation of lactobacilli. *Journal of Applied Bacteriology*, **23** : 130-135.

MacFaddin, J.F.. 1985. *Media for isolation-cultivation-identification-maintenance of medical bacteria*. Williams & Wilkins, Baltimore, volume 1: 543-545.

ISO 9232 / IDF 146. Février 2003. Yaourt. Identification des micro-organismes caractéristiques (*Lactobacillus delbrueckii* subsp. *bulgaricus* et *Streptococcus thermophilus*).

12 ADDITIONAL INFORMATION

The information provided on the labels take precedence over the formulations or instructions described in this document and are susceptible to modification at any time, without warning.

Document code : MRS BROTHS_ENv7
Creation date : 06-2004
Updated : 08-2016
Origin of revision : General update.