



## Rogosa SL Agar

M130

Rogosa SL Agar is used as a selective medium for cultivation of oral, vaginal and faecal *Lactobacilli*.

### Composition\*\*

Ingredients	Gms / Litre
Tryptose	10.000
Yeast extract	5.000
Dextrose	10.000
Arabinose	5.000
Saccharose	5.000
Sodium acetate	15.000
Ammonium citrate	2.000
Monopotassium phosphate	6.000
Magnesium sulphate	0.570
Manganese sulphate	0.120
Ferrous sulphate	0.030
Polysorbate 80	1.000
Agar	15.000
Final pH ( at 25°C)	5.4±0.2

\*\*Formula adjusted, standardized to suit performance parameters

### Directions

Suspend 74.72 grams in 1000 ml distilled water. Heat to boiling to dissolve the medium completely. Add 1.32 ml glacial acetic acid. Mix thoroughly, distribute into culture tubes or flasks. Heat to 90 - 100°C for 2-3 minutes. Cool to 45-50°C for direct inoculation. DO NOT AUTOCLAVE.

### Principle And Interpretation

Rogosa SL Agar also known as RMW Agar, is a modification of the media formulated by Rogosa, Mitchell and Wiseman (3, 4). This media is used for isolation, enumeration and identification of *Lactobacilli* from foodstuffs and clinical specimens (1, 2). Accompanying bacterial flora is suppressed due to the low pH of the medium and also because of the high sodium acetate concentration.

Tryptose and yeast extract provide nitrogenous compounds, sulphur, trace elements and vitamin B complex, essential for growth of *Lactobacilli*. Dextrose, arabinose and saccharose are the fermentable carbohydrates. Polysorbate 80 is the source of fatty acids. Ammonium citrate and Sodium acetate inhibit moulds, *Streptococci* and many other organisms. Monopotassium phosphate provides buffering capability. Magnesium sulphate, manganese sulphate and ferrous sulphate are sources of inorganic ions. Low pH of the medium and addition of acetic acid makes the medium selective for *Lactobacilli* inhibiting other bacterial flora (2).

It is recommended that the plates should be incubated at 30°C for 5 days or at 37°C for 3 days in an atmosphere of 95% hydrogen and 5% carbon dioxide (5). If this is not possible, overlay the inoculated plates with a second layer of the agar before incubation. High acetate concentration and acidic pH suppress many strains of other lactic acid bacteria. All colonies should be checked by gram staining and by catalase test before further identification. The salt in the formulation makes the medium unsuitable for isolation of dairy *Lactobacilli*. e.g. *L.lactis*, *L.bulgaricus* and *L.helveticus* (2, 4).

### Quality Control

#### Appearance

Cream to yellow homogeneous soft lumps which can be easily broken down to powder form.

#### Gelling

Firm, comparable with 1.5% Agar gel

**Colour and Clarity of prepared medium**

Light yellow coloured opalescent gel forms in Petri plates

**Reaction**

Reaction of 7.5% w/v aqueous solution with 0.132% v/v acetic acid at 25°C. pH : 5.4±0.2

**pH**

5.20-5.60

**Cultural Response**

M130: Cultural characteristics observed in presence of 5% Carbon dioxide (CO<sub>2</sub>) and 95% H<sub>2</sub> after an incubation at 35-37°C for 40-48 hours.

Organism	Inoculum (CFU)	Growth	Recovery
<b>Cultural Response</b>			
<i>Lactobacillus casei</i> ATCC 9595	50-100	good - luxuriant	≥50%
<i>Lactobacillus fermentum</i> ATCC 9338	50-100	good to luxuriant	≥50%
<i>Lactobacillus leichmanni</i> ATCC 4797	50-100	good to luxuriant	≥50%
<i>Lactobacillus plantarum</i> ATCC 8014	50-100	good-luxuriant	≥50%
<i>Staphylococcus aureus</i> ATCC 25923	≥10 <sup>3</sup>	inhibited	0%

**Storage and Shelf Life**

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

**Reference**

- Downes F. P. and Ito K., (Eds.), Compendium of Methods for the Microbiological Examination of Foods, 4th Ed., American Public Health Association, Washington, D.C.
- MacFaddin J. F., 1985, Media for Isolation-Cultivation-Identification- Maintenance of Medical Bacteria, Vol. I, Williams and Wilkins, Baltimore. Md.
- Rogosa M., Mitchell J. A. and Wiseman R. F., 1951, J. Bacteriol., 62, 132-133.
- Rogosa M., Mitchell J. A. and Wiseman R. F., 1951, J. Dental Res. 30:682.
- Sharpe M. L. (Ed.), 1960, Lab-Practice, 9(4): 223.

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