M.R.S. Agar

(de Man, Rogosa and Sharpe Agar)

LAB 93

Description
This medium was developed for the cultivation and enumeration of *Lactobacillus* spp. from various sources and is intended as a substitute for Tomato Juice Agar. The medium is suitable for most lactic acid bacteria. When acidified to pH 5.4 M.R.S. Agar can be used to enumerate *Lactobacillus bulgaricus* in yoghurts. Tween® 80, sodium acetate, magnesium and manganese sulphates act as growth stimulants.

Formula	g/litre
Mixed Peptones	10.0
Yeast Extract	5.0
Beef Extract	10.0
Glucose	20.0
Potassium phosphate	2.0
Sodium acetate	5.0
Ammonium citrate	2.0
Magnesium sulphate	0.2
Manganese sulphate	0.05
Tween® 80	1.08
Agar No. 1	15.0

Method for reconstitution
Weigh 70 grams of powder and add 1 litre of deionised water. Allow to soak for 10 minutes, swirl to mix. Heat to dissolve and sterilise by autoclaving at 121°C for 15 minutes. Cool to 47°C and adjust pH if the acidified medium is required.

Appearance: Light amber, clear.

pH: 6.4 ± 0.2

Minimum Q.C. organisms: *Lactobacillus acidophilus*

Storage of Prepared Medium: Plates – up to 7 days at 2-8°C in the dark. Capped containers – up to 1 month at 15-20°C in the dark.

Inoculation: Surface, spread to cover surface, or use pour plate technique.

Incubation: 25°C microaerobically for 2-5 days.

Interpretation: Count all colonies exhibiting typical morphology.

Growth Characteristics			
organism	**colony size (mm)**	**shape & surface**	**colour**
Lactobacillus acidophilus	0.5-2.5	F.E.G.	White Rough
Lactobacillus sake	0.5-1.0	F.E.G.	White
Streptococcus lactis	0.5-1.5	CV.E.G.	White
Lactobacillus bulgaricus	1.0-1.3	CV.E. G.	White

References
de Man, J.C.,. Rogosa, M and Sharpe, M.E. (1960). Amedium for the cultivation of lactobacilli. J. Appl. Bacteriol. 23: 130-135.