

## 69964 MRS Agar (Lactobacillus Agar acc. to De Man, Rogosa and Sharpe)

For the enrichment, cultivation and isolation of all species of Lactobacillus from all types of material according to De Man, Rogosa and Sharpe.

### Composition:

Ingredients	Grams/Litre
Universal peptone	10.0
Meat extract	5.0
Yeast extract	5.0
D(+)-Glucose	20.0
Dipotassium hydrogen phosphate	2.0
Diammonium hydrogen citrate	2.0
Sodium acetate	5.0
Magnesium sulfate	0.1
Manganous sulfate	0.05
Agar	12.0
Final pH 6.5 +/- 0.2 at 37°C	

Store prepared media below 8°C, protected from direct light. Store dehydrated powder, in a dry place, in tightly-sealed containers at 2-25°C.

### Directions:

Dissolve 61 g in 1 litre distilled water and add 1 ml Tween 80 (Fluka No. 93780). Autoclave at 121°C for 15 minutes.

Incubate the culture up to 3 days at 35°C or up to 5 days at 30°C. If possible, incubate the plates in a CO<sub>2</sub>-enriched atmosphere in an anaerobic jar. Do not allow the surface of the plates to dry as this will cause acetate concentration increasing at the surface, which inhibits the growth of lactobacilli.

### Principle and Interpretation:

The MRS media formulation was developed by de Man, Rogosa and Sharpe to replace the tomato juice medium and the meat extract tomato juice medium. It is a medium supporting good growth of lactobacilli in general, even those strains which have shown poor growth in existing media, like strains of *L. brevis* and *L. fermenti*. The MRS culture media contain polysorbate (Tween 80), acetate, magnesium and manganese which are known to act as special growth factors for lactobacilli as well as a rich nutrient base. As these media show a very low degree of selectivity, *Pediococcus* and *Leuconostoc* species as well as other secondary bacteria may grow on them. Most of the accompanying microflora can be inhibited by thallium acetate (Fluka 88204), sorbic acid (Fluka 85510), acetic acid (Fluka 45740), sodium nitrite (Fluka 71759), cycloheximide (Fluka 01810) and polymyxin (Fluka 81334). These substances can be used at varying concentrations and combinations, but inevitably a compromise has to be reached between selectivity and productivity of the organism sought.

Cultural characteristics:

Organisms (ATCC)	Growth
<i>Lactobacillus acidophilus</i> (4356)	++
<i>Lactobacillus fermentum</i> (9338)	++
<i>Bifidobacterium bifidum</i> (11863)	++ (anaerobic)
<i>Escherichia coli</i> (25922)	-/+
<i>Pseudomonas aeruginosa</i> (27853)	none to poor

References:

1. J. C. de Man, M. Rogosa and M. Elisabeth Sharpe, *Appl. Bact.* 23. 130-135 (1960)
2. M. Briggs, *J. Dairy Res.* 20. 36-40 (1953)
3. G. Reuter, *Intern. J. Food Microbiol.* 2. 55-68 (1985)
4. ISO/TC 34/SC 6/WG 15, No.3 and 5, Enumeration of Lactobacteriaceae in meat and meat products (1984)
5. Lankaputhra W.E.V., Shah N.P. and Britz M.L. *Food Australia* 48. 113-118 (1996)
6. W. Hummel, et al., *Biocatalysis* 2, 293 (1989)
7. L.C. Laleye, et al., Involvement of heterofermentative lactobacilli in development of open texture in cheese, *J. Food Prot.* 50, 1009 (1987)
8. P. Laloi, et al., Cell-wall-associated proteinase of *L. delbrueckii* subsp. *bulgaricus* CNRZ 397, *Appl. Microbiol. Biotechnol.* 36, 196 (1991)

## 69966 MRS Broth (Lactobacillus Broth acc. to De Man, Rogosa and Sharpe)

For the enrichment, cultivation and isolation of all species of Lactobacillus from all types of material according to De Man, Rogosa and Sharpe.

### Composition:

Ingredients	Grams/Litre
Peptone	10.0
Meat extract	8.0
Yeast extract	4.0
D(+)-Glucose	20.0
Dipotassium hydrogen phosphate	2.0
Sodium acetate trihydrate	5.0
Triammonium citrate	2.0
Magnesium sulfate heptahydrate	0.2
Manganous sulfate tetrahydrate	0.05

Store prepared media below 8°C, protected from direct light. Store dehydrated powder, in a dry place, in tightly-sealed containers at 2-25°C.

### Directions:

Dissolve 51 g in 1 litre distilled water and add 1 ml Tween 80 (Fluka No. 93780). Fill into containers and sterilize by autoclaving at 121°C for 15 minutes.

Incubate the culture up to 3 days at 35°C or up to 5 days at 30°C. If possible, incubate the culture in a CO<sub>2</sub> enriched atmosphere in an anaerobic jar.

### Principle and Interpretation:

The MRS media formulation was developed by de Man, Rogosa and Sharpe to replace the tomato juice medium and the meat extract tomato juice medium. It is a medium supporting good growth of lactobacilli in general, even those strains which have shown poor growth in existing media, like strains of *L. brevis* and *L. fermenti*. The MRS culture media contain polysorbate (Tween 80), acetate, magnesium and manganese which are known to act as special growth factors for lactobacilli as well as a rich nutrient base. As these media show a very low degree of selectivity *Pediococcus* and *Leuconostoc* species as well as other secondary bacteria may grow on them. Most of the accompanying microflora can be inhibited by thallium acetate (Fluka 88204), sorbic acid (Fluka 85510), acetic acid (Fluka 45740), sodium nitrite (Fluka 71759), cycloheximide (Fluka 01810) and polymyxin (Fluka 81334). These substances can be used at varying concentrations and combinations, but inevitably a compromise has to be reached between selectivity and productivity of the organism sought.

Cultural characteristics:

Organisms (ATCC)	Growth
<i>Lactobacillus acidophilus</i> (4356)	++
<i>Lactobacillus fermentum</i> (9338)	++
<i>Bifidobacterium bifidum</i> (11863)	++ (anaerobic)
<i>Escherichia coli</i> (25922)	-/+
<i>Pseudomonas aeruginosa</i> (27853)	none to poor

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1. J. C. de Man, M. Rogosa and M. Elisabeth Sharpe, *Appl. Bact.* 23. 130-135 (1960)
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5. Lankaputhra W.E.V., Shah N.P. and Britz M.L. *Food Australia* 48. 113-118 (1996)
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7. L.C. Laleye, et al., Involvement of heterofermentative lactobacilli in development of open texture in cheese, *J. Food Prot.* 50, 1009 (1987)
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