

# MRS BROTH w/ and w/o DURHAM TUBE

## INTENDED USE

Remel MRS Broth w/ and w/o Durham Tube (DT) are liquid media recommended for use in the cultivation of *Lactobacillus* species and in the differentiation of gram-positive cocci.

## SUMMARY AND EXPLANATION

In the 1950s, tomato juice agar was used to isolate *Lactobacillus* from food products. Rogosa et al. recommended a medium for isolation of lactobacilli from oral and fecal specimens, but it was found to be inadequate for recovery of *Lactobacillus* spp. from dairy products.<sup>1</sup> A modification of this formulation was developed by de Man, Rogosa, and Sharpe by eliminating tomato juice.<sup>2</sup> This medium became known as MRS Broth. It was found to be superior to previous formulations because it supported the growth of slower-growing lactobacilli. Recently, MRS Broth has been used in clinical laboratories to differentiate certain genera of gram-positive cocci on the basis of gas production.<sup>3-6</sup>

## PRINCIPLE

Gelatin peptone and beef extract provide essential nutrients and amino acids necessary for the growth of bacteria. Yeast extract is a source of B-complex vitamins and enhances bacterial growth. Dextrose provides a ready source of energy, and fermentation of dextrose is detected by gas production in the Durham tube. Dipotassium phosphate aids in the maintenance of osmotic equilibrium. Polysorbate 80 supplies fatty acids required for bacterial metabolism. Ammonium citrate and sodium acetate are selective agents which inhibit the growth of certain organisms, including gram-negative bacteria and molds.

## REAGENTS (CLASSICAL FORMULA)\*

Dextrose.....	20.0 g	Ammonium Citrate.....	2.0 g
Gelatin Peptone .....	10.0 g	Dipotassium Phosphate .....	2.0 g
Beef Extract.....	8.0 g	Polysorbate 80.....	1.0 g
Sodium Acetate.....	5.0 g	Magnesium Sulfate.....	0.2 g
Yeast Extract.....	4.0 g	Manganese Sulfate .....	0.05 g
		Demineralized Water .....	1000.0 ml

pH 6.2 ± 0.2 @ 25°C

\*Adjusted as required to meet performance standards.

## PROCEDURE

1. Inoculate MRS Broth with an isolated colony from an 18-24 hour culture. Alternatively, a 0.1 ml aliquot from an 18-24 hour pure culture in Todd Hewitt Broth may be used.
2. To determine gas production, inoculate MRS Broth w/ DT.
3. Incubate aerobically at 33-37°C for up to 7 days.
4. Observe for growth and/or gas production. Further biochemical testing may be required for definitive identification of *Lactobacillus* and certain gram-positive cocci. Consult appropriate references for further instructions.<sup>7-8</sup>

## INTERPRETATION OF THE TEST

### Gas Production:

Positive - Bubbles in Durham tube  
Negative - No bubbles

### Growth:

Positive - Turbidity in medium, growth on subculture  
Negative - No turbidity and no growth on subculture

## QUALITY CONTROL

All lot numbers of MRS Broth w/ and w/o DT have been tested using the following quality control organisms and found to be acceptable. Testing of control organisms should be performed in accordance with established laboratory quality control procedures. If aberrant quality control results are noted, patient results should not be reported.

## CONTROL

*Lactobacillus johnsonii* ATCC® 33200  
*Pediococcus acidilactici* ATCC® 33314  
*Weissella paramesenteroides* ATCC® 33313

## INCUBATION

Aerobic, up to 72 h @ 33-37°C  
Aerobic, up to 72 h @ 33-37°C  
Aerobic, up to 72 h @ 33-37°C

## RESULTS

Good growth, Gas (-)  
Good growth, Gas (-)  
Good growth, Gas (+)

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Refer to the front of Remel Technical Manual of Microbiological Media for General Information regarding precautions, product storage and deterioration, specimen collection, storage and transportation, materials required, quality control, and limitations.

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**remel**

12076 Santa Fe Drive, Lenexa, KS 66215, USA

General Information: (800) 255-6730 Website: [www.remel.com](http://www.remel.com) Email: [remel@remel.com](mailto:remel@remel.com)  
Local/International Phone: (913) 888-0939 International Fax: (913) 895-4128