# **MIDDLEBROOK 7H10 AGAR BASE**

# INTENDED USE

Remel Middlebrook 7H10 Agar Base is a solid medium recommended for use in qualitative procedures for the isolation and cultivation of *Mycobacterium* spp.

## SUMMARY AND EXPLANATION

Dubos and Middlebrook developed media formulations containing oleic acid and albumin which enhanced the growth of tubercle bacilli and protected the organisms against a variety of toxic agents.<sup>1</sup> In 1958, Middlebrook and Cohn improved the previous formulation of oleic acid-albumin agar to obtain 7H10 Agar which allowed faster, more luxuriant growth of *Mycobacterium* species.<sup>2</sup>

## PRINCIPLE

This medium contains inorganic salts which are essential for the growth of mycobacteria. Sodium citrate is converted to citric acid which holds inorganic cations in solution. Malachite-green dye is a selective agent which partially inhibits bacteria other than mycobacteria.

## **REAGENTS (CLASSICAL FORMULAE)\***

Dipotassium Phosphate	1.5	g
Monopotassium Phosphate	1.5	g
Ammonium Sulfate	0.5	g
Monosodium Glutamate	0.5	g
Sodium Citrate	0.4	g
Ferric Ammonium Citrate	0.04	g
Magnesium Sulfate	25.0 r	ng

Copper Sulfate1.0	
Pyridoxine Hydrochloride1.0	mg
Zinc Sulfate1.0	mg
Biotin	mg
Calcium Chloride0.5	mg
Malachite Green0.25	mg
Agar 15.0	g
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pH 6.6 ± 0.2 @ 25°C

\*Adjusted as required to meet performance standards.

#### PREPARATION OF DEHYDRATED CULTURE MEDIUM

- 1. Suspend 19 g of medium in 900 ml of demineralized water containing 5 ml of glycerol.
- 2. Heat to boiling with agitation to completely dissolve.
- 3. Sterilize by autoclaving at 121°C for 15 minutes or following established laboratory procedures.
- 4. Cool to 45-50°C and aseptically add 100 ml of OADC Enrichment (REF R450603).
- 5. Mix thoroughly and dispense into appropriate containers.

#### PROCEDURE

1. Consult current editions of appropriate references for the recommended procedure for sample preparation, inoculation, testing, and interpretation.<sup>3,4</sup>

## **QUALITY CONTROL**

Each lot number of Middlebrook 7H10 Agar Base has been manufactured, packaged, and processed in accordance with current Good Manufacturing Practice regulations. All lot numbers have been tested using the following quality control organisms and have been 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, sample results should not be reported.

CONTROL	INCUBATION	RESULTS
Mycobacterium fortuitum ATCC <sup>®</sup> 6841	CO <sub>2</sub> , up to 21 days @ 33-37°C	Growth
Mycobacterium intracellulare ATCC <sup>®</sup> 13950	CO <sub>2</sub> , up to 21 days @ 33-37°C	Growth
Mycobacterium kansasii ATCC <sup>®</sup> 12478	CO <sub>2</sub> , up to 21 days @ 33-37°C	Growth
Mycobacterium scrofulaceum ATCC <sup>®</sup> 19981	CO <sub>2</sub> , up to 21 days @ 33-37°C	Growth
$M$ ycobacterium tuberculosis ATCC $^{\circ}$ 25177	CO <sub>2</sub> , up to 21 days @ 33-37°C	Growth

#### **BIBLIOGRAPHY**

- 1. Dubos, R.J. and G. Middlebrook. 1947. Am. Rev. Tuberc. 56:334-345.
- 2. Middlebrook, G. and M.L. Cohn. 1958. Am. J. Public Health. 48:844-853.
- 3. Kent, P.T. and G.P. Kubica. 1985. Public Health Mycobacteriology, A Guide for the Level III Laboratory. U.S. Dept. of H.H.S. and CDC, Atlanta, GA.

4. Isenberg, H.D. 2004. Clinical Microbiology Procedures Handbook. 2<sup>nd</sup> ed. ASM Press, Washington, D.C.

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

 $\mathsf{ATCC}^{\oplus}$  is a registered trademark of American Type Culture Collection. IFU 453981, Revised August 23, 2010

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