# MIDDLEBROOK 7H11 AGAR BASE

#### **INTENDED USE**

Remel Middlebrook 7H11 Agar Base is a solid medium recommended for use in qualitative procedures for the isolation and cultivation of Mycobacterium species.

## **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. In 1958, Middlebrook and Cohn improved the previous formulation of oleic acidalbumin agar to obtain 7H10 Agar which allowed faster, more luxuriant growth of Mycobacterium spp. In 1968, Cohn et al. demonstrated that the addition of casein hydrolysate to 7H10 Agar stimulated the growth of mycobacteria which would otherwise not grow on 7H10 media.3 formulation is designated as Middlebrook 7H11 Agar. Middlebrook 7H11 Agar Base requires the addition of glycerol and OADC Enrichment.

## **PRINCIPLE**

This medium contains inorganic salts, which are essential for the growth of mycobacteria. Casein hydrolysate serves as a growth stimulant for drug-resistant strains of Mycobacterium tuberculosis. Sodium citrate is converted to citric acid which holds inorganic cations in solution. Malachite-green dye is a selective agent, which inhibits bacteria other than mycobacteria. Biotin helps to stimulate the growth of damaged mycobacteria. Glycerol is added to Middlebrook 7H11 Agar Base as a source of carbon and energy, and OADC Enrichment provides nutrients required for the growth of mycobacteria: albumin, oleic acid, sodium chloride, catalase, and dextrose.

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

Dipotassium Phosphate1.5	g	Magnesium Sulfate	0.05	g
Monopotassium Phosphate1.5		Ferric Ammonium Citrate	0.04	g
Casein Hydrolysate	g	Malachite Green	1.0 m	ng
Ammonium Sulfate	g	Pyridoxine Hydrochloride	1.0 m	ng
Monosodium Glutamate	g	Biotin	0.5 m	ng
Sodium Citrate	g	Agar	.15.0	g

pH 6.6 ± 0.2 @ 25°C

## **PRECAUTIONS**

This product is For Laboratory Use only. It is not intended for use in the diagnosis of disease or other conditions.

## PREPARATION OF DEHYDRATED CULTURE MEDIUM

- Suspend 20 g of medium in 900 ml of demineralized water containing 5 ml of glycerol.
- Heat to boiling with agitation to completely dissolve.
- Sterilize by autoclaving at 121°C for 15 minutes or following established laboratory procedures. 3
- Cool to 45-50°C and aseptically add 100 ml of OADC Enrichment (REF R450603).
- Dispense into appropriate containers.

#### **PROCEDURE**

Consult current editions of appropriate references for the recommended procedure for sample preparation, inoculation, testing, and interpretation.5,6

#### **QUALITY CONTROL**

Each lot number of Middlebrook 7H11 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® 6841	CO <sub>2</sub> , up to 21 days @ 33-37°C	Growth	
Mycobacterium intracellulare ATCC® 13950	CO <sub>2</sub> , up to 21 days @ 33-37°C	Growth	
Mycobacterium kansasii ATCC® 12478	CO <sub>2</sub> , up to 21 days @ 33-37°C	Growth	
Mycobacterium scrofulaceum ATCC® 19981	CO <sub>2</sub> , up to 21 days @ 33-37°C	Growth	
Mycobacterium tuberculosis ATCC® 25177	CO <sub>2</sub> , up to 21 days @ 33-37°C	Growth	

## **BIBLIOGRAPHY**

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- Cohn, M.L., G.F. Waggoner, and J.K. McClatchy. 1968. Am. Rev. Respir. Dis. 98:295.
- MacFaddin, J.F. 1985. Media for Isolation-Cultivation-Identification-Maintenance of Medical Bacteria. Vol. 1. Williams & Wilkins, Baltimore, MD.
- 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. 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.

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<sup>\*</sup>Adjusted as required to meet performance standards.