SUPPORTING INFECTIOUS DISEASE RESEARCH

Mycobacterium tuberculosis, Strain 98-2852

Catalog No. NR-30882

This reagent is the tangible property of the U.S. Government.

Product Description: *Mycobacterium tuberculosis (M. tuberculosis)*, strain 98-2852 was isolated between 1995 and 2000 from human sputum from an HIV-negative patient infected with pulmonary tuberculosis in North America.

Lot¹: 70003482

Manufacturing Date: 07JUL2017

TEST	SPECIFICATIONS	RESULTS
Phenotypic Analysis ²		
Cellular morphology	Gram-positive rods	Gram-positive rods
Colony morphology ³	Report results	Irregular, slight peaked, undulate, rough and cream (Figure 1)
Growth rate	≥ 7 days	22 days
Growth at 26°C	Negative	Negative
Growth at 37°C	Positive	Positive
Acid-fast stain	Positive (red colonies)	Positive (red colonies)
Pigmentation in the dark (Scotochromogen)	Negative (no pigment)	Negative (no pigment)
Photoinduction for 1 hour (Photochromogen)	Negative (no pigment)	Negative (no pigment)
Nonchromogen (no pigment)	Positive (no pigment)	Positive (no pigment)
Biochemical tests		
Niacin production ⁴	Positive	Positive
Nitrate reduction	Positive	Positive
Pyrazinamidase	Positive	Positive
Antibiotic Susceptibility Profile		
Sensititre [™] System ^{5,6}		
Amikacin	Report results	0.5 μg/mL
Cycloserine	Report results	16 µg/mL
Ethambutol	Report results	2 µg/mL ⁷
Ethionamide	Report results	2.5 μg/mL ^{7,8}
Isoniazid	Report results	≤ 0.03 µg/mL
Kanamycin	Report results	2.5 µg/mL
Moxifloxacin	Report results	0.25 µg/mL
Ofloxacin	Report results	0.5 μg/mL
Para-aminosalicylic acid	Report results	≤ 0.5 µg/mL ⁷
Rifabutin	Report results	≤ 0.12 µg/mL ⁷
Rifampin	Report results	≤ 0.12 µg/mL
Streptomycin	Report results	1 μg/mL ^{7,9}
Genotypic Analysis		
Sequencing of Heat Shock Protein 65 gene	≥ 99% sequence identity to	100% sequence identity to
(~ 420 base pairs)	M. tuberculosis type strain	M. tuberculosis type strain
	(GenBank: AL123456)	(GenBank: AL123456) ¹⁰
Purity (post-freeze)		
Middlebrook 7H10 agar with OADC enrichment ¹¹	Growth consistent with expected	Growth consistent with expected
-	colony morphology	colony morphology
Tryptic Soy agar ¹²	Report results	Growth consistent with expected colony morphology
Viability (post-freeze) ³	Growth	Growth

¹NR-30882 was produced by inoculation of the deposited material into Middlebrook 7H9 broth with ADC enrichment. Broth inoculum was added to Middlebrook 7H10 agar with OADC enrichment kolles, which were grown for 46 days at 37°C in an aerobic atmosphere with 5% CO₂. The resulting

BEI Resources www.beiresources.org E-mail: <u>contact@beiresources.org</u> Tel: 800-359-7370 Fax: 703-365-2898 DICIÍ RESOURCES

SUPPORTING INFECTIOUS DISEASE RESEARCH

growth was harvested in Middlebrook 7H9 broth with ADC enrichment supplemented with 10% glycerol and frozen. The frozen material was later thawed and aliquoted into cryovials and frozen to produce this lot.

²Information on Mycobacterium testing is available from Ribón, W. "Biochemical Isolation and Identification of Mycobacteria." <u>Biochemical Testing.</u> (2012) Jose C. Jimenez-Lopez (Ed.), InTech, <u>http://www.intechopen.com/books/biochemical-testing/biochemical-isolation-and-identification-of-mycobacteria</u> and Lévy-Frébault, V. V. and F. Portaels. "Proposed Minimal Standards for the Genus *Mycobacterium* and for Description of New Slowly Growing Mycobacterium Species." Int. J. Syst. Bacteriol. 42 (1992): 315-323. PubMed: 1581193.

 3 22 days at 37°C in an aerobic atmosphere with 5% CO₂ on Middlebrook 7H10 agar with OADC enrichment

⁴All mycobacteria produce niacin but only *M. tuberculosis* accumulates it, resulting in a positive test for *M. tuberculosis*.

⁵SensititreTM System Mycobacterium tuberculosis MIC Plate, Thermo ScientificTM, catalog number MYCOTB

⁶Minimum Inhibitory Concentration (MIC); No Clinical & Laboratory Standards Institute (CLSI) interpretations of the Sensititre™ System data for M. tuberculosis are currently available.

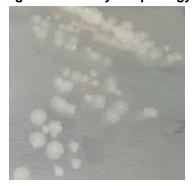
⁷For streptomycin, ethionamide, para-aminosalicylic acid, rifabutin and ethambutol, the endpoint for these drugs is determined by the well with approximately 80% inhibition of growth compared to the positive control well with no drug.

⁸Two MICs were observed for ethionamide (1.2 µg/mL and 2.5 µg/mL) under identical test conditions. The highest MIC is being reported as the test result. Variability in the MIC result by the Sensititre™ method has been demonstrated (Lee, J., et al. "Sensititre MYCOTB MIC Plate for Testing Drugs." Antimicrob. and Mycobacterium tuberculosis Susceptibility to First-Second-Line Agents Chemother. 58 (2014): 11-18. PubMed: 24100497.), with the results for a single antibiotic typically within one doubling dilution.

⁹Two MICs were observed for streptomycin (0.5 µg/mL and 1 µg/mL) under identical test conditions. The highest MIC is being reported as the test result. Variability in the MIC result by the Sensititre™ method has been demonstrated (Lee, J., et al. "Sensititre MYCOTB MIC Plate for Testing tuberculosis and Susceptibility to Second-Line Drugs." Antimicrob. Agents Chemother. Mvcobacterium First-58 (2014): 11-18. PubMed: 24100497.), with the results for a single antibiotic typically within one doubling dilution. ¹⁰Also consistent with *M. africanum, M. bovis, M. canettii, M. caprae* and *M. microti*

¹¹Purity of this lot was assessed for 32 days at 37°C in an aerobic atmosphere with 5% CO₂. ¹²Purity of this lot was assessed for 22 days at 37°C in an aerobic atmosphere with 5% CO₂.

Figure 1: Colony Morphology



/Heather Couch/

Heather Couch Program Manager or designee, ATCC Federal Solutions

ATCC®, on behalf of BEI Resources, hereby represents and warrants that the material provided under this certificate has been subjected to the tests and procedures specified and that the results described, along with any other data provided in this certificate, are true and accurate to the best of ATCC®'s knowledge.

ATCC[®] is a trademark of the American Type Culture Collection. You are authorized to use this product for research use only. It is not intended for human use.



24 AUG 2018