SUPPORTING INFECTIOUS DISEASE RESEARCH

Mycobacterium tuberculosis, Strain 97-2972

Catalog No. NR-30819

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

Product Description: *Mycobacterium tuberculosis (M. tuberculosis)*, strain 97-2972 was isolated between 1995 and 2000 from human sputum from an HIV-negative patient infected with pulmonary tuberculosis in North America.

Lot¹: 70001589

Manufacturing Date: 21MAR2017

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	1 µg/mL
Cycloserine	Report results	16 µg/mL ^{7,8}
Ethambutol	Report results	$4 \mu g/m L^{8,9,10}$
Ethionamide	Report results	2.5 µg/mL ¹⁰
Isoniazid	Report results	0.12 µg/mL
Kanamycin	Report results	2.5 μg/mL
Moxifloxacin	Report results	4 μg/mL ^{8,11}
Ofloxacin	Report results	2 µg/mL
Para-aminosalicylic acid	Report results	> 64 µg/mL ¹⁰
Rifabutin	Report results	0.25 μg/mL ¹⁰
Rifampin	Report results	1 μg/mL ^{8,12}
Streptomycin	Report results	1 μg/mL ¹⁰
Genotypic Analysis		
Sequencing of Heat Shock Protein 65 gene (~ 440 base pairs)	≥ 99% sequence identity to M. tuberculosis type strain (GenBank: AL123456)	100% sequence identity to <i>M. tuberculosis</i> type strain (GenBank: AL123456) ¹³
Purity (post-freeze)		
Middlebrook 7H10 agar with OADC enrichment ¹⁴	Growth consistent with expected colony morphology	Growth consistent with expected colony morphology
Tryptic Soy agar ¹⁵	Report results	No growth
Viability (post-freeze) ³	Growth	Growth

¹NR-30819 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 22 days at 37°C in an aerobic atmosphere with 5% CO₂ 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-</u>

BEI Resources www.beiresources.org E-mail: <u>contact@beiresources.org</u> Tel: 800-359-7370 Fax: 703-365-2898 **D**|**e**|**i** resources

SUPPORTING INFECTIOUS DISEASE RESEARCH

mycobacteria 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.

³22 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*.

⁵Sensititre[™] System *Mycobacterium tuberculosis* MIC Plate, Thermo Scientific[™], 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.

⁷Two MICs were observed for cycloserine (8 µg/mL and 16 µg/mL) under identical test conditions. The highest MIC is being reported as the test result.

⁸Variability in the MIC result by the Sensititre method has been demonstrated (Lee, J., et al. "Sensititre MYCOTB MIC Plate for Testing *Mycobacterium tuberculosis* Susceptibility to First- and Second-Line Drugs." <u>Antimicrob. Agents Chemother.</u> 58 (214): 11-18. PubMed: 24100497.), with the results for a single antibiotic typically within one doubling dilution.

⁹Two MICs were observed for ethambutol (2 μg/mL and 4 μg/mL) under identical test conditions. The highest MIC is being reported as the test result. ¹⁰For 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.

¹¹Two MICs were observed for moxifloxacin (2 µg/mL and 4 µg/mL) under identical test conditions. The highest MIC is being reported as the test result.

¹²Two MICs were observed for rifampin (0.5 μg/mL and 1 μg/mL) under identical test conditions. The highest MIC is being reported as the test result. ¹³Also consistent with *M. africanum*, *M. bovis*, *M. canettii*, *M. caprae* and *M. microti*

¹⁴Purity of this lot was assessed for 41 days at 37°C in an aerobic atmosphere with 5% CO₂.

¹⁵Purity of this lot was assessed for 22 days at 37°C in an aerobic atmosphere with 5% CO₂.



Figure 1: Colony Morphology

26 MAR 2018

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.

