

Certificate of Analysis for NR-43823

Mycobacterium tuberculosis, Strain KT-0038

Catalog No. NR-43823

Product Description:

Mycobacterium tuberculosis (M. tuberculosis), strain KT-0038 was isolated from a human in South Korea. Strain KT-0038 was deposited as an extensively drug-resistant (XDR) Beijing genotype strain with resistance to isoniazid, capreomycin, kanamycin, moxifloxacin, pyrazinamide and rifampin. NR-43823 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 21 days at 37°C in an aerobic atmosphere with 5% CO₂ to produce this lot.

Lot: 70023006 Manufacturing Date: 06MAY2019

TEST	SPECIFICATIONS	RESULTS
Phenotypic Analysis ¹		
Cellular morphology ²	Gram-positive rods	Gram-positive rods
Colony morphology	Report results	Irregular, peaked, undulate, rough and cream (Figure 1)
Growth rate	≥ 7 days	21 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)
Nonchromogen (no pigment)	Positive (no pigment)	Positive (no pigment)
Biochemical tests		
Niacin production ³	Positive	Positive
Nitrate reduction	Positive	Positive
Antibiotic Susceptibility Profile		
Sensititre™ System ^{4,5}		
Amikacin	Report results	0.5 μg/mL ^{6,7}
Cycloserine	Report results	4 μg/mL
Ethambutol	Report results	4 μg/mL ⁸
Ethionamide	Report results	1.2 μg/mL ^{7,8,9}
Isoniazid	Report results	> 4 μg/mL
Kanamycin	Report results	40 μg/mL
Moxifloxacin	Report results	0.12 µg/mL
Ofloxacin	Report results	2 μg/mL
Para-aminosalicylic acid	Report results	≤ 0.5 μg/mL ⁸
Rifabutin	Report results	0.25 μg/mL ^{7,8,10}
Rifampin	Report results	2 μg/mL ^{7,11}
Streptomycin	Report results	≤ 0.25 µg/mL ⁸
Genotypic Analysis		
Sequencing of Heat Shock Protein 65 gene	≥ 99% sequence identity to	100% sequence identity to
(~ 1620 base pairs)	<i>M. tuberculosi</i> s type strain (GenBank: AL123456.3)	<i>M. tuberculosi</i> s type strain (GenBank: AL123456.3) ¹²
Purity (post-freeze)	,	,
Middlebrook 7H10 agar with OADC enrichment	Growth consistent with expected	Growth consistent with expected
36 days at 37°C in an aerobic atmosphere with 5% CO ₂	colony morphology	colony morphology
Tryptic Soy agar	Report results	Growth consistent with expected
21 days at 37°C in an aerobic atmosphere with 5% CO ₂		colony morphology
Viability (post-freeze) ²	Growth	Growth
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¹Information 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, http://www.intechopen.com/books/biochemical-testing/biochemical-isolation-and-identification-of-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." <u>Int. J. Syst. Bacteriol.</u> 42 (1992): 315-323. PubMed: 1581193.

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

³All mycobacteria produce niacin but only *M. tuberculosis* accumulates it, resulting in a positive test for *M. tuberculosis*.

⁴Sensititre™ System *Mycobacterium tuberculosis* MIC Plate, Thermo Scientific™, catalog number MYCOTB

⁵Minimum Inhibitory Concentration (MIC); No Clinical & Laboratory Standards Institute (CLSI) interpretations of the Sensititre™ System data for *M. tuberculosis* are currently available.

⁶Two MICs were observed for amikacin (0.25 μg/mL and 0.5 μg/mL) under identical test conditions. The highest MIC is being reported as the test result.

⁷Variability 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 (2014): 11-18. PubMed: 24100497.], with the results for a single antibiotic typically within one doubling dilution.

⁸For ethambutol, ethionamide, para-aminosalicylic acid, rifabutin and streptomycin, 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.

⁹Two MICs were observed for ethionamide (0.6 μg/mL and 1.2 μg/mL) under identical test conditions. The highest MIC is being reported as the test result.

¹⁰Two MICs were observed for rifabutin (≤ 0.12 μg/mL and 0.25 μg/mL) under identical test conditions. The highest MIC is being reported as the test result.

11Two MICs were observed for rifampin (1 μg/mL and 2 μg/mL) under identical test conditions. The highest MIC is being reported as the test result.

¹²Also consistent with M. africanum, M. bovis, M. canettii, M. caprae and M. microti

Figure 1: Colony Morphology



/Heather Couch/ Heather Couch

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Program Manager or designee, ATCC Federal Solutions

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