

Certificate of Analysis for NR-43808

Mycobacterium tuberculosis, Strain KT-0023

Catalog No. NR-43808

Product Description:

Mycobacterium tuberculosis (M. tuberculosis), strain KT-0023 was isolated from a human in South Korea. Strain KT-0023 was deposited as a multidrug-resistant (XDR) Beijing genotype strain, with resistance to isoniazid, moxifloxacin, moxifloxacin, ofloxacin, pyrazinamide, rifampin and streptomycin. NR-43808 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.

Lot: 70021306 Manufacturing Date: 17JAN2019

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TEST	SPECIFICATIONS	RESULTS
Phenotypic Analysis ¹		
Cellular morphology	Gram-positive rods	Gram-positive rods
20 days at 37°C in an aerobic atmosphere with		
5% CO ₂ on Middlebrook 7H10 agar with OADC		
enrichment		
Colony morphology	Report results	Irregular, slight peaked, undulate, rough and cream
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 ²	Positive	Positive
Nitrate reduction	Positive	Positive
Pyrazinamidase	Positive	Positive
Antibiotic Susceptibility Profile		
Sensititre™ System ^{3,4}		
Amikacin	Report results	4 μg/mL
Cycloserine	Report results	> 256 µg/mL ^{5,6}
Ethambutol	Report results	> 32 μg/mL ⁷
Ethionamide	Report results	> 40 μg/mL ⁷
Isoniazid	Report results	> 4 μg/mL
Kanamycin	Report results	10 μg/mL
Moxifloxacin	Report results	> 8 µg/mL ^{5,8}
Ofloxacin	Report results	16 μg/mL
Para-aminosalicylic acid	Report results	> 64 μg/mL ⁷
Rifabutin	Report results	> 16 μg/mL ⁷
Rifampin	Report results	> 16 µg/mL
Streptomycin	Report results	4 μg/mL ^{5,7,9}
Genotypic Analysis		
Sequencing of Heat Shock Protein 65 gene	≥ 99% sequence identity to	100% sequence identity to
(~ 1620 base pairs)	M. tuberculosis, strain KT-0023 (GenBank: JLSB01000003.1)	M. tuberculosis, strain KT-0023 (GenBank: JLSB01000003.1) ¹⁰

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TEST	SPECIFICATIONS	RESULTS
Purity (post-freeze) Middlebrook 7H10 agar with OADC enrichment 47 days at 37°C in an aerobic atmosphere with 5% CO ₂	Growth consistent with expected colony morphology	Growth consistent with expected colony morphology
Tryptic Soy agar 22 days at 37°C in an aerobic atmosphere with 5% CO ₂	Report results	Growth consistent with expected colony morphology
Viability (post-freeze) 20 days at 37°C in an aerobic atmosphere with 5% CO₂ on Middlebrook 7H10 agar with OADC enrichment	Growth	Growth

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, (<u>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." <u>Int. J. Syst.</u> Bacteriol. 42 (1992): 315-323. PubMed: 1581193.

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31 MAR 2023

Technical Manager or designee, ATCC Federal Solutions

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²All mycobacteria produce niacin but only *M. tuberculosis* accumulates it, resulting in a positive test for *M. tuberculosis*.

³Sensititre™ System *Mycobacterium tuberculosis* MIC Plate, Thermo Scientific™, Catalog Number MYCOTB

⁴Minimum Inhibitory Concentration (MIC); No Clinical & Laboratory Standards Institute (CLSI) interpretations of the Sensititre™ System data for *M. tuberculosis* are currently available.

⁵Two MICs were observed for cycloserine (256 μg/mL and > 256 μg/mL) under identical test conditions. The highest MIC is being reported as the test result.

⁶Variability 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.

⁷For 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.

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

 $^{^9}$ Two MICs were observed for streptomycin (2 μ g/mL and 4 μ g/mL) under identical test conditions. The highest MIC is being reported as the test result.

¹⁰Also consistent with *M. africanum*, *M. bovis*, *M. canettii*, *M. caprae* and *M. microti*