

Certificate of Analysis for NR-43809

Mycobacterium tuberculosis, Strain KT-0024

Catalog No. NR-43809

Product Description:

Mycobacterium tuberculosis (M. tuberculosis), strain KT-0024 is a human isolate from South Korea. Strain KT-0024 was deposited as an extensively drug-resistant (XDR) Beijing genotype strain with resistance to ethambutol, isoniazid, moxifloxacin, ofloxacin, pyrazinamide, rifampin and streptomycin.

Lot: 70021308¹ Manufacturing Date: 01MAR2019

TEST	SPECIFICATIONS	RESULTS
Phenotypic Analysis ²		
Cellular morphology	Gram-positive rods	Gram-positive rods
Colony morphology ³	Report results	Irregular, low convex, 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)
Biochemical tests	, , ,	, , ,
Niacin production ⁴	Positive	Positive
Nitrate reduction	Positive	Positive
Pyrazinamidase	Positive	Negative ⁵
Antibiotic Susceptibility Profile		
Sensititre™ System ^{6,7}		
Amikacin	Report results	0.25 μg/mL
Cycloserine	Report results	32 μg/mL
Ethambutol	Report results	8 µg/mL ⁸⁻¹⁰
Ethionamide	Report results	5 µg/mL ⁸
Isoniazid	Report results	1 µg/mL
Kanamycin	Report results	2.5 µg/mL ^{10,11}
Moxifloxacin	Report results	4 µg/mL
Ofloxacin	Report results	16 μg/mL
Para-aminosalicylic acid	Report results	2 µg/mL ^{8,10,12}
Rifabutin	Report results	4 µg/mL ^{8,10,13}
Rifampin	Report results	> 16 µg/mL
Streptomycin	Report results	> 32 μg/mL ⁸
Genotypic Analysis		
Sequencing of Heat Shock Protein 65 gene	≥ 99% sequence identity to	100% sequence identity to
(~ 1540 base pairs)	M. tuberculosis, strain KT-0024	M. tuberculosis, strain KT-0024
	(GenBank: JLSA01000069.1)	(GenBank: JLSA01000069.1) ¹⁴
Purity (post-freeze)		
Middlebrook 7H10 agar with OADC enrichment ¹⁵	Growth consistent with expected	Growth consistent with expected
-	colony morphology	colony morphology
Tryptic Soy agar ¹⁶	Report results	Growth consistent with expected
	·	colony morphology
Viability (post-freeze) ³	Growth	Growth
Trability (post-freeze)	Glowaii	Ciowuii

¹NR-43809 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.

BEI Resources

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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, 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." <a href="https://link.growing.nc.nlm.nlm.growing.nlm.grow

³21 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*.

⁵A negative result may indicate a low expression of pyrazinamidase activity or a mutation to the pyrazinamidase/nicotinamidase (*pncA*) gene conferring resistance to pyrazinamidase (Sheen, P., et al. "Effect of Pyrazinamidase Activity on Pyrazinamide Resistance in *Mycobacterium tuberculosis*." <u>Tuberculosis (Edinb).</u> 89 (2009): 109-113. PubMed: 19249243.).

⁶Sensititre™ System *Mycobacterium tuberculosis* MIC Plate, Thermo Scientific™, catalog number MYCOTB

⁷Minimum Inhibitory Concentration (MIC); No Clinical & Laboratory Standards Institute (CLSI) interpretations of the Sensititre™ System data for *M. tuberculosis* are currently available.

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 ethambutol (4 μg/mL and 8 μg/mL) under identical test conditions. The highest MIC is being reported as the test result. ¹⁰Variability 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.

¹¹Two MICs were observed for kanamycin (1.2 μg/mL and 2.5 μg/mL) under identical test conditions. The highest MIC is being reported as the test result.

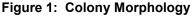
¹²Two MICs were observed for para-aminosalicylic acid (1 μg/mL and 2 μg/mL) under identical test conditions. The highest MIC is being reported as the test result.

13Two MICs were observed for rifabutin (2 µg/mL and 4 µg/mL) under identical test conditions. The highest MIC is being reported as the test result.

¹⁴Also consistent with M. africanum, M. bovis, M. canettii, M. caprae and M. microti

¹⁵Purity of this lot was assessed for 32 days at 37°C in an aerobic atmosphere with 5% CO₂.

¹⁶Purity of this lot was assessed for 21 days at 37°C in an aerobic atmosphere with 5% CO₂.





/Heather Couch/ Heather Couch

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Program Manager or designee, ATCC Federal Solutions

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