

## **Certificate of Analysis for NR-48757**

## Mycobacterium tuberculosis, Strain 11949-0

## Catalog No. NR-48757

**Product Description:** *Mycobacterium tuberculosis* (*M. tuberculosis*), strain 11949-0 was isolated in October 2012 from a subculture of a strain originally isolated from a patient with pulmonary tuberculosis in the Republic of South Africa. *M. tuberculosis*, strain 11949-0 was deposited as a multidrug-resistant (MDR) strain with resistance to amikacin, capreomycin, ethambutol, ethionamide, isoniazid, kanamycin, pyrazinamide, rifampin and streptomycin.

Lot<sup>1</sup>: 63950994 Manufacturing Date: 15JAN2016

TEST	SPECIFICATIONS	RESULTS
Phenotypic Analysis <sup>2</sup>		
Cellular morphology	Gram-positive rods	Gram-positive rods
Colony morphology <sup>3</sup>	Report results	Irregular, slight peaked, undulate,
5 y gy		rough and cream
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	1 ositive (no pigment)	1 ositive (no pigment)
Niacin production <sup>4</sup>	Positive	Positive
Nitrate reduction	Positive	Positive
Pyrazinamidase	Positive	Negative <sup>5</sup>
i yrazinamidase	1 Ositive	Negative
Antibiotic Susceptibility Profile		
Sensititre <sup>™</sup> System <sup>6,7</sup>		
Amikacin	Report results	> 16 µg/mL
Cycloserine	Report results	32 μg/mL
Ethambutol	Report results	8 μg/mL <sup>8</sup>
Ethionamide	Report results	5 μg/mL <sup>8</sup>
Isoniazid	Report results	> 4 µg/mL
Kanamycin	Report results	> 40 µg/mL
Moxifloxacin	Report results	0.5 μg/mL
Ofloxacin	Report results	1 μg/mL
Para-aminosalicylic acid	Report results	≤ 0.5 μg/mL <sup>8</sup>
Rifabutin	Report results	0.5 μg/mL <sup>8</sup>
Rifampin	Report results	> 16 µg/mL
Streptomycin	Report results	32 μg/mL <sup>8</sup>
	<u> </u>	
Genotypic Analysis		4000
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)9
Purity (post-freeze)		
Middlebrook 7H10 agar with OADC enrichment <sup>10</sup>	Growth consistent with expected	Growth consistent with expected
middioblook /1110 agai willi OADO GillollillGill	colony morphology	colony morphology
Tryptic Soy agar <sup>11</sup>	Report results	Growth consistent with expected
Trypho ooy agai	Report results	colony morphology
		Colony morphology
Viability (post-freeze) <sup>3</sup>	Growth	Growth
·-		

<sup>&</sup>lt;sup>1</sup>NR-48757 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 29 days at 37°C in an aerobic atmosphere with 5% CO₂ to produce this lot.

**BEI Resources** 

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<sup>2</sup>Information 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, <a href="http://www.intechopen.com/books/biochemical-testing/biochemical-isolation-and-identification-of-mycobacteria">http://www.intechopen.com/books/biochemical-testing/biochemical-isolation-and-identification-of-mycobacteria</a> 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://example.com/normalized/linearing/l

<sup>3</sup>21 days at 37°C in an aerobic atmosphere with 5% CO₂ on Middlebrook 7H10 agar with OADC enrichment

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

<sup>5</sup>A 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.).

<sup>6</sup>Sensititre<sup>™</sup> System *Mycobacterium tuberculosis* MIC Plate, Thermo Scientific<sup>™</sup>, catalog number MYCOTB

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

<sup>8</sup>For 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.

<sup>9</sup>Also consistent with M. africanum, M. bovis, M. canettii, M. caprae and M. microti

<sup>10</sup>Purity of this lot was assessed for 50 days at 37°C in an aerobic atmosphere with 5% CO<sub>2</sub>.

<sup>11</sup>Purity of this lot was assessed for 21 days at 37°C in an aerobic atmosphere with 5% CO<sub>2</sub>.

Date: 08 AUG 2017

Signature:

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