SUPPORTING INFECTIOUS DISEASE RESEARCH

Mycobacterium tuberculosis, Strain XTB13-111

Catalog No. NR-49366

Product Description: *Mycobacterium tuberculosis* (*M. tuberculosis*), strain XTB13-111 was isolated in 2012 from the sputum of an HIV positive patient with tuberculosis in the Republic of Belarus. Strain XTB13-111 was deposited as resistant to ethambutol, isoniazid, rifampin and streptomycin.

Lot¹: 64064219

Manufacturing Date: 11MAY2016

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	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	16 μg/mL
Ethambutol	Report results	2 µg/mL ⁸
Ethionamide	Report results	1.2 μg/mL ⁸
Isoniazid	Report results	4 µg/mL
Kanamycin	Report results	2.5 µg/mL
Moxifloxacin	Report results	0.25 µg/mL
Ofloxacin	Report results	1 μg/mL
Para-aminosalicylic acid	Report results	≤ 0.5 μg/mL ⁸
Rifabutin	Report results	4 μg/mL ⁸
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	99.7% sequence identity to
(~ 300 base pairs)	M. tuberculosis, strain XTB13-111	M. tuberculosis, strain XTB13-111
	(GenBank: JLLM01000002.1)	(GenBank: JLLM01000002.1) ⁹
Purity (post-freeze)		
Middlebrook 7H10 agar with OADC enrichment ¹⁰	Growth consistent with expected	Growth consistent with expected
	colony morphology	colony morphology
Tryptic Soy agar ¹¹	Report results	No growth
Viability (post-freeze) ³	Growth	Growth

¹NR-49366 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 62 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>

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Certificate of Analysis for NR-49366

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(2012) Jose C. Jimenez-Lopez (Ed.), InTech, <u>http://www.intechopen.com/books/biochemical-testing/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." Int. J. Syst. Bacteriol. 42 (1992): 315-323. PubMed: 1581193.

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 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.

⁹Also consistent with M. africanum, M. bovis, M. canettii, M. caprae and M. microti

¹⁰Purity of this lot was assessed for 93 days at 37°C in an aerobic atmosphere with 5% CO₂.

¹¹Purity of this lot was assessed for 21 days at 37°C in an aerobic atmosphere with 5% CO₂.

Figure 1: Colony Morphology

Date: 25 OCT 2017

Signature:

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