

Certificate of Analysis for NR-30716

Mycobacterium tuberculosis, Strain 96-3160

Catalog No. NR-30716

This reagent is the tangible property of the U.S. Government.

Product Description: *Mycobacterium tuberculosis (M. tuberculosis)*, strain 96-3160 was isolated between 1995 and 2000 from human sputum from an HIV-negative patient infected with pulmonary tuberculosis in North America.

Lot¹: 64121866 Manufacturing Date: 03MAY2016

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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	Positive
Genotypic Analysis		
Sequencing of Heat Shock Protein 65 gene (~ 400 base pairs)	≥ 99% sequence identity to M. tuberculosis type strain (GenBank: AL123456)	100% sequence identity to M. tuberculosis type strain (GenBank: AL123456) ⁵
Purity (post-freeze)		
Middlebrook 7H10 agar with OADC enrichment ⁶	Growth consistent with expected colony morphology	Growth consistent with expected colony morphology
Tryptic Soy agar ⁶	Report results	No growth
Viability (post-freeze) ³	Growth	Growth

¹NR-30716 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 49 days at 37°C in an aerobic atmosphere with 5% CO₂ to produce this lot.

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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." <u>Int. J. Syst. Bacteriol.</u> 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*.

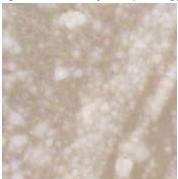
⁵Also consistent with M. africanum, M. bovis, M. canettii, M. caprae and M. microti

⁶Purity of this lot was assessed for 52 days at 37°C in an aerobic atmosphere with 5% CO₂.



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Figure 1: Colony Morphology



Date: 12 DEC 2017

Signature:

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