

***Mycobacterium tuberculosis*, Strain 96-3266**

Catalog No. NR-30935

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Product Description: *Mycobacterium tuberculosis* (*M. tuberculosis*), strain 96-3266 was isolated between 1995 and 2000 from human sputum from an HIV-positive patient infected with pulmonary tuberculosis in North America.

Lot¹: 70005377

Manufacturing Date: 07JUL2018

TEST	SPECIFICATIONS	RESULTS
Phenotypic Analysis² Cellular morphology Colony morphology ³ Growth rate Growth at 26°C Growth at 37°C Acid-fast stain Pigmentation in the dark (Scotochromogen) Photoinduction for 1 hour (Photochromogen) Nonchromogen (no pigment) Biochemical tests Niacin production ⁴ Nitrate reduction Pyrazinamidase	Gram-positive rods Report results ≥ 7 days Negative Positive Positive (red colonies) Negative (no pigment) Negative (no pigment) Positive (no pigment) Positive Positive Positive	Gram-positive rods Irregular, slight peaked, undulate, rough and cream (Figure 1) 21 days Negative Positive Positive (red colonies) Negative (no pigment) Negative (no pigment) Negative (no pigment) Positive (no pigment) Positive Positive Positive
Antibiotic Susceptibility Profile Sensititre™ System ^{5,6} Amikacin Cycloserine Ethambutol Ethionamide Isoniazid Kanamycin Moxifloxacin Ofloxacin Para-aminosalicylic acid Rifabutin Rifampin Streptomycin	Report results Report results Report results Report results Report results Report results Report results Report results Report results Report results Report results Report results Report results	0.25 µg/mL 8 µg/mL ≤ 0.5 µg/mL ⁷ 1.2 µg/mL ⁷⁻⁹ ≤ 0.03 µg/mL ≤ 2.5 µg/mL ^{9,10} 0.5 µg/mL ^{9,11} 1 µg/mL ^{9,12} ≤ 0.5 µg/mL ⁷ ≤ 0.12 µg/mL ⁷ 0.25 µg/mL ^{9,13} 0.5 µg/mL ^{7,9,14}
Genotypic Analysis Sequencing of Heat Shock Protein 65 gene (~ 370 base pairs)	≥ 99% sequence identity to <i>M. tuberculosis</i> type strain (GenBank: AL123456)	100% sequence identity to <i>M. tuberculosis</i> type strain (GenBank: AL123456) ¹⁵
Purity (post-freeze) Middlebrook 7H10 agar with OADC enrichment ¹⁶ Tryptic Soy agar ¹⁷	Growth consistent with expected colony morphology Report results	Growth consistent with expected colony morphology Growth consistent with expected colony morphology
Viability (post-freeze)³	Growth	Growth

¹NR-30935 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 32 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." Biochemical Testing. (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." 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*.
- ⁵Sensititre™ System *Mycobacterium tuberculosis* MIC Plate, Thermo Scientific™, catalog number MYCOTB
- ⁶Minimum Inhibitory Concentration (MIC); No Clinical & Laboratory Standards Institute (CLSI) interpretations of the Sensititre™ System data for *M. tuberculosis* are currently available.
- ⁷For 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.
- ⁸Two MICs were observed for ethionamide (0.6 µg/mL and 1.2 µg/mL) under identical test conditions. The highest MIC is being reported as the test result.
- ⁹Variability 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." Antimicrob. Agents Chemother. 58 (2014): 11-18. PubMed: 24100497.), with the results for a single antibiotic typically within one doubling dilution.
- ¹⁰Two 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.
- ¹¹Two MICs were observed for moxifloxan (0.25 µg/mL and 0.5 µg/mL) under identical test conditions. The highest MIC is being reported as the test result.
- ¹²Two MICs were observed for ofloxacin (0.5 µg/mL and 1 µg/mL) under identical test conditions. The highest MIC is being reported as the test result.
- ¹³Two MICs were observed for rifampin (≤ 0.12 µg/mL and 0.25 µg/mL) under identical test conditions. The highest MIC is being reported as the test result.
- ¹⁴Two MICs were observed for streptomycin (≤ 0.25 µg/mL and 0.5 µg/mL) under identical test conditions. The highest MIC is being reported as the test result.
- ¹⁵Also consistent with *M. africanum*, *M. bovis*, *M. canettii*, *M. caprae* and *M. microti*
- ¹⁶Purity of this lot was assessed for 59 days at 37°C in an aerobic atmosphere with 5% CO₂.
- ¹⁷Purity of this lot was assessed for 21 days at 37°C in an aerobic atmosphere with 5% CO₂.

Figure 1: Colony Morphology



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