

***Mycobacterium insubricum*, Strain FI-06250T**

Catalog No. NR-49077

Product Description: *Mycobacterium insubricum* (*M. insubricum*), strain FI-06250T was isolated in 2006 from sputum of a 62-year-old patient with chronic obstructive pulmonary disease in Varese, Italy.

Lot¹: 64362401

Manufacturing Date: 12JUL2016

TEST	SPECIFICATIONS	RESULTS
Phenotypic Analysis^{2,3} Cellular morphology Colony morphology ⁴ Growth on MacConkey agar (without crystal violet) Growth rate Growth at 45°C Growth at 55°C Acid-fast stain Biochemical tests Nitrate reduction Aryl sulfate (3 days) Aryl sulfate (14 days) Iron uptake Growth in the presence of 5% sodium chloride Growth in the presence of thiophene-2-carboxylic acid hydrazide (TCH)	Report results Report results Report results ≤ 7 days Negative Report results Positive (red colonies) Negative Negative Report results Report results Report results Positive	Rods Irregular, low convex, undulate, rough and white Negative 6 days Positive ⁵ Negative Positive (red colonies) Positive ⁶ Negative Negative Negative Positive Positive
Genotypic Analysis Sequencing of 16S ribosomal RNA gene (~ 1990 base pairs) Digital DNA-DNA hybridization (dDDH) ⁷	≥ 99% sequence identity to <i>M. insubricum</i> type strain (GenBank: EU605695.1) ≥ 70% for species identification	99.9% sequence identity to <i>M. insubricum</i> type strain (GenBank: EU605695.1) Not determined ^{8,9} (Table 1)
Purity (post-freeze) Middlebrook 7H10 agar with OADC enrichment ¹⁰ Tryptic Soy agar ¹⁰	Growth consistent with expected colony morphology Report results	Growth consistent with expected colony morphology Growth consistent with expected colony morphology
Viability (post-freeze)⁴	Growth	Growth

¹NR-49077 was produced by inoculation of the deposited material in Middlebrook 7H9 broth with ADC enrichment for 6 days at 37°C in an aerobic atmosphere with 5% CO₂. Broth inoculum was added to Middlebrook 7H10 agar with OADC enrichment kolles, which were grown for 5 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.

³Phenotypic characterization of *M. insubricum* was performed following: Tortoli, E., et al. "*Mycobacterium insubricum* sp. nov." *Int. J. Syst. Evol. Microbiol.* 59 (2009): 1518-1523. PubMed: 19502346.

⁴6 days at 37°C in an aerobic atmosphere with 5% CO₂ on Middlebrook 7H10 agar with OADC enrichment

⁵NR-49077 was deposited as *M. insubricum* and reported to be negative for growth at 42°C. Testing performed in triplicate by BEI Resources indicates growth was observed after 21 days at 45°C in an aerobic atmosphere in Middlebrook 7H9 broth with ADC enrichment and after 7 days at 45°C in an aerobic atmosphere on Middlebrook 7H10 agar with OADC enrichment and Lowenstein-Jensen agar.

⁶NR-49077 was deposited as *M. insubricum* and reported to be negative for nitrate reduction. Testing performed by BEI Resources indicates a positive result.

⁷Relatedness between bacterial strains has traditionally been determined using dDDH. For additional information, refer to Auch, A.F., et al. "Digital DNA-DNA Hybridization for Microbial Species Delineation by Means of Genome-to-Genome Sequence Comparison." *Stand. Genomic Sci.* 2 (2010):

117-134. PubMed: 21304684.

⁸The whole genome of *M. insubricum*, strain FI-06250T (Contig Total Length ~ 4.7 megabase pairs) was sequenced using the Illumina® MiSeq® system and was assembled and analyzed with CLC Genomics Workbench Version 7.0.2.

⁹The required whole genome sequence for the type strain of this species is not available. dDDH testing rules out all species listed in Table 1, however, this does not rule out species for which the type strains whole genome sequences are not available.

¹⁰Purity of this lot was assessed for 8 days at 37°C in an aerobic atmosphere with 5% CO₂.

Table 1: Digital DNA-DNA hybridization (dDDH)

Species	Strain	Accession #	GGD vs. NR-49077 (Deposited as: <i>M. insubricum</i>)
<i>M. abscessus</i> subsp. <i>abscessus</i>	Hauduroy L948 ^T	NC_010397.1	20.1
<i>M. abscessus</i> subsp. <i>bolletii</i>	BD ^T	AHAS00000000.1	20.2
<i>M. abscessus</i> subsp. <i>massiliense</i>	CCUG 48898 ^T	NZ_AP014547.1	20.1
<i>M. africanum</i>	ATCC 25420 ^T	ATCC	19.9
<i>M. agri</i>	90012 ^T	ATCC	20.4
<i>M. algericum</i>	TBE 500028/10 ^T	ATCC	20.7
<i>M. alsense</i>	TB 1906 ^T	ATCC	20.3
<i>M. arabiense</i>	YIM 121001 ^T	ATCC	20.1
<i>M. aromaticivorans</i>	JS19b1 ^T	JALN00000000.2	20.4
<i>M. arosiense</i>	T1921 ^T	ATCC	20.4
<i>M. arupense</i>	AR30097 ^T	ATCC	20.8
<i>M. asiaticum</i>	ATCC 25276 ^T	CCBD00000000.1	19.9
<i>M. aubagnense</i>	U8 ^T	ATCC	20.5
<i>M. aurum</i>	ATCC 23366 ^T	CVQQ01000001.1	20
<i>M. austroafricanum</i>	E9789-SA12441 ^T	HG964450.1	20.8
<i>M. avium</i> subsp. <i>avium</i>	ATCC 25291 ^T	ACFI00000000.1	20.7
<i>M. avium</i> subsp. <i>paratuberculosis</i>	ATCC 19698 ^T	AGAR00000000.1	21
<i>M. avium</i> subsp. <i>silvaticum</i>	6409 ^T	AYOC00000000.1	21
<i>M. bacteremicum</i>	ATCC 25791 ^T	ATCC	21
<i>M. boenickei</i>	W5998 ^T	ATCC	20.8
<i>M. bohemicum</i>	CIP 105808 ^T	CSTD01000001.1	20.5
<i>M. bouchedurhonense</i>	4355387 ^T	MVHL00000000.1	20.6
<i>M. bourgelatii</i>	MLB-A84 ^T	ATCC	20.3
<i>M. bovis</i>	ATCC 19210 ^T	ATCC	20
<i>M. branderi</i>	ATCC 51789 ^T	ATCC	20.1
<i>M. brisbanense</i>	W6743 ^T	ATCC	20.7
<i>M. brumae</i>	CR-270 ^T	FJNX00000000.1	23.7
<i>M. canariasense</i>	502329 ^T	BCSY00000000.1	22.2
<i>M. caprae</i>	spc-1 ^T	ATCC	19.9
<i>M. celatum</i>	ATCC 51131 ^T	BBUN00000000.1	20.7
<i>M. celeriflavum</i>	AFPC-000207 ^T	ATCC	21.1
<i>M. chelonae</i>	CM 6388 ^T	CP010946.1	20
<i>M. chimaera</i>	FI-01069 ^T	MRBR00000000.1	20.4
<i>M. chitae</i>	ATCC 19627 ^T	ATCC	21.3
<i>M. chlorophenicolum</i>	PCP-I ^T	JYNL00000000.1	20.6
<i>M. chubuense</i>	48013 ^T	NC_018027.1	20.7
<i>M. colombiense</i>	10B ^T	AFVW00000000.2	20.5
<i>M. conceptionense</i>	D16 ^T	CTEF00000000.1	20.7
<i>M. confluentis</i>	1389/90 ^T	ATCC	21

Species	Strain	Accession #	GGD vs. NR-49077 (Deposited as: <i>M. insubricum</i>)
<i>M. conspicuum</i>	3895/92 ^T	LQOR00000000.1	20.3
<i>M. cookii</i>	NZ2 ^T	ATCC	20
<i>M. cosmeticum</i>	LTA-388 ^T	CCBB00000000.1	20.9
<i>M. crocinum</i>	czh-42 ^T	BBHD00000000.1	22.3
<i>M. diernhoferi</i>	41001 ^T	ATCC	20.8
<i>M. doricum</i>	FI-13295 ^T	ATCC	20.8
<i>M. duvalii</i>	ATCC 43910 ^T	ATCC	20.4
<i>M. elephantis</i>	484 ^T	ATCC	22.6
<i>M. engbaekii</i>	ATCC 27353 ^T	ATCC	20.6
<i>M. europaeum</i>	FI-95228 ^T	ATCC	20.3
<i>M. fallax</i>	ATCC 35219 ^T	ATCC	31.4
<i>M. farcinogenes</i>	IEMVT 75 ^T	CCAY00000000.1	20.6
<i>M. flavescens</i>	ATCC 14474 ^T	ATCC	20.2
<i>M. florentinum</i>	FI-93171 ^T	ATCC	20.3
<i>M. fluoranthenivorans</i>	FA4 ^T	BBFT00000000.1	23.8
<i>M. fortuitum</i> subsp. <i>fortuitum</i>	ATCC 6841 ^T	CP014258.1	20.6
<i>M. fortuitum</i> subsp. <i>acetamidolyticum</i>	NCH E11620 ^T	BCSZ00000000.1	20.6
<i>M. fragae</i>	HF8705 ^T	ATCC	19.8
<i>M. franklinii</i>	DSM 45524 ^T	ATCC	21.9
<i>M. frederiksbergense</i>	Fan9 ^T	ATCC	22.4
<i>M. gadium</i>	ATCC 27726 ^T	ATCC	19.9
<i>M. gastris</i>	ATCC 15754 ^T	AZYN00000000.1	19.9
<i>M. genavense</i>	2289 ^T	JAGZ00000000.1	20.1
<i>M. gilvum</i>	ATCC 43909 ^T	ATCC	21.5
<i>M. gordonae</i>	ATCC 14470 ^T	ATCC	20.5
<i>M. haemophilum</i>	ATCC 29548 ^T	CP011883.2	19.5
<i>M. hassiacum</i>	3849 ^T	ARBU00000000.1	20.7
<i>M. heckeshornense</i>	S369 ^T	ATCC	19.8
<i>M. heidelbergense</i>	2554/91 ^T	ATCC	20.4
<i>M. heraklionense</i>	GN-1 ^T	ATCC	20.6
<i>M. hiberniae</i>	Hi 11 ^T	LQOZ00000000.1	20.4
<i>M. hippocampi</i>	BFLP-6 ^T	ATCC	21.4
<i>M. hodleri</i>	EMI2 ^T	BBGO00000000.1	23.1
<i>M. holsaticum</i>	1406 ^T	ATCC	20.4
<i>M. houstonense</i>	W5198 ^T	FJVO00000000.1	21.3
<i>M. immunogenum</i>	BH29 ^T	ATCC	20
<i>M. interjectum</i>	4185/92 ^T	FJVQ00000000.1	20.5
<i>M. intermedium</i>	1669/91 ^T	ATCC	20.3
<i>M. intracellulare</i>	ATCC 13950 ^T	NC_016946.1	20.3
<i>M. iranikum</i>	M05 ^T	ATCC	20.9
<i>M. kansasii</i>	ATCC 12478 ^T	NC_022663.1	20.1
<i>M. komossense</i>	Ko 2 ^T	ATCC	20.2
<i>M. koreense</i>	01-305 ^T	ATCC	21.5
<i>M. kubicae</i>	ATCC 700732 ^T	ATCC	19.9
<i>M. kumamotoense</i>	CST 7247 ^T	ATCC	20.6
<i>M. kyorinense</i>	KUM 060204 ^T	BBKA00000000.1	20.2

Species	Strain	Accession #	GGD vs. NR-49077 (Deposited as: <i>M. insubricum</i>)
<i>M. lacus</i>	NRCM 00-255 ^T	ATCC	20.4
<i>M. lentiflavum</i>	2186/92 ^T	ATCC	19.9
<i>M. litorale</i>	F4 ^T	ATCC	21.7
<i>M. llatzerense</i>	MG13 ^T	ATCC	21.9
<i>M. longobardum</i>	FI-07034 ^T	ATCC	20.8
<i>M. madagascariense</i>	P2 ^T	ATCC	20.2
<i>M. mageritense</i>	938 ^T	CCBF00000000.1	20.8
<i>M. malmesburyense</i>	WCM 7299 ^T	CVTB00000000.1	20.4
<i>M. malmoense</i>	ATCC 29571 ^T	ATCC	20.4
<i>M. manitobense</i>	NRCM 01-154 ^T	ATCC	20.8
<i>M. mantenii</i>	04-1474 ^T	ATCC	20.4
<i>M. marinum</i>	ATCC 927 ^T	ATCC	20
<i>M. marseillense</i>	5356591 ^T	ATCC	21
<i>M. microti</i>	ATCC 19422 ^T	ATCC	20
<i>M. minnesotense</i>	DL49 ^T	ATCC	20.5
<i>M. monacense</i>	B9-21-178 ^T	ATCC	20.8
<i>M. montefiorensis</i>	ATCC BAA-256 ^T	ATCC	20
<i>M. moriokaense</i>	NCH E11715 ^T	ATCC	20.2
<i>M. mucogenum</i>	MO76 ^T	ATCC	20.9
<i>M. murale</i>	MA112/96 ^T	ATCC	22.5
<i>M. nebraskense</i>	UNMC-MY 1349 ^T	ATCC	20.3
<i>M. neoaurum</i>	ATCC 25795 ^T	JMDW00000000.1	20.9
<i>M. neworleansense</i>	W6705 ^T	CWKH00000000.1	20.7
<i>M. nonchromogenicum</i>	ATCC 19530 ^T	ATCC	20.8
<i>M. noviomagense</i>	NLA000500338 ^T	ATCC	20.1
<i>M. novocastrensis</i>	73 ^T	BCTA00000000.1	20
<i>M. obuense</i>	47001 ^T	JYNU00000000.1	20.3
<i>M. pallens</i>	czh-8 ^T	BBHE00000000.1	22
<i>M. palustre</i>	E846 ^T	ATCC	20.4
<i>M. paraense</i>	IEC26 ^T	ATCC	20.8
<i>M. paraffinicum</i>	ATCC 12670 ^T	ATCC	20.2
<i>M. parafortuitum</i>	ATCC 19686 ^T	ATCC	20.5
<i>M. paragordonae</i>	49061 ^T	ATCC	20.5
<i>M. paraintracellulare</i>	MOTT-64 ^T	CP003324.1	20.3
<i>M. parakoreense</i>	299 ^T	ATCC	20.4
<i>M. parascrofulaceum</i>	HSC-68 ^T	ADNV00000000.1	20.4
<i>M. paraseoulense</i>	31118 ^T	ATCC	20.2
<i>M. parmense</i>	MUP 1182 ^T	ATCC	20.3
<i>M. peregrinum</i>	ATCC 14467 ^T	ATCC	21
<i>M. persicum</i>	JC290 ^T	MVIF00000000.1	19.6
<i>M. phlei</i>	ATCC 11758 ^T	ANBO00000000.1	21
<i>M. phocaicum</i>	N4 ^T	ATCC	20.9
<i>M. pinnipedii</i>	6482 ^T	ATCC	19.9
<i>M. porcinum</i>	E10241-1 ^T	MVIG00000000.1	20.9
<i>M. poriferae</i>	47 ^T	ATCC	20.8
<i>M. pseudoshottsii</i>	L15 ^T	BCND00000000.1	20.2

Species	Strain	Accession #	GGD vs. NR-49077 (Deposited as: <i>M. insubricum</i>)
<i>M. psychrotolerans</i>	WA101 ^T	ATCC	20.8
<i>M. pulveris</i>	NCH 33505 ^T	ATCC	20.6
<i>M. pyrenivorans</i>	17A3 ^T	BBHB00000000.1	22.1
<i>M. rhodesiae</i>	02002 ^T	ATCC	20.8
<i>M. riyadhense</i>	NLA000201958 ^T	ATCC	19.7
<i>M. rufum</i>	JS14 ^T	JROA00000000.1	21.1
<i>M. rutilum</i>	czh-117 ^T	BBHF00000000.1	23.9
<i>M. salmoniphilum</i>	SC ^T	ATCC	19.9
<i>M. saopaulense</i>	EPM 10906 ^T	CP010271.1	20.1
<i>M. saskatchewanense</i>	00-250 ^T	ATCC	20.1
<i>M. scrofulaceum</i>	ATCC 19981 ^T	ATCC	20.2
<i>M. sediminus</i>	YIM M13028 ^T	ATCC	21.2
<i>M. senegalense</i>	IEMVT 378 ^T	ATCC	20.9
<i>M. senuese</i>	05-832 ^T	ATCC	20.6
<i>M. seoulense</i>	03-19 ^T	ATCC	20.1
<i>M. septicum</i>	W4964 ^T	CBMO00000000.1	20.8
<i>M. setense</i>	ABO-M06 ^T	JTJW00000000.1	20.6
<i>M. sherrisii</i>	4773 ^T	ATCC	20.7
<i>M. shimoidei</i>	E4796 ^T	ATCC	20.2
<i>M. shinjukuense</i>	GTC 2738 ^T	ATCC	20.1
<i>M. shottsii</i>	M175 ^T	ATCC	20.5
<i>M. simiae</i>	ATCC 25275 ^T	CBMJ00000000.2	20.1
<i>M. smegmatis</i>	ATCC 19420 ^T	LN831039.1	20.9
<i>M. sphagni</i>	Sph 38 ^T	ATCC	20.7
<i>M. stomatepiae</i>	T11 ^T	ATCC	20.2
<i>M. szulgai</i>	ATCC 35799 ^T	ATCC	20
<i>M. terrae</i>	ATCC 15755 ^T	ATCC	21.8
<i>M. thermoresistibile</i>	ATCC 19527 ^T	BCTB00000000.1	20.4
<i>M. timonense</i>	5351974 ^T	ATCC	20.6
<i>M. tokaiense</i>	47503 ^T	ATCC	22.1
<i>M. triplex</i>	90-1019 ^T	CCAU00000000.1	20.2
<i>M. triviale</i>	ATCC 23292 ^T	ATCC	21.3
<i>M. tuberculosis</i>	H37Rv ^T	NC_000962.3	19.9
<i>M. tusciae</i>	FI-25796 ^T	ATCC	21.2
<i>M. vaccae</i>	ATCC 15483 ^T	BCRS00000000.1	20.9
<i>M. vanbaalenii</i>	PYR-1 ^T	NC_008726.1	20.7
<i>M. vulneris</i>	NLA000700772 ^T	CCBG00000000.1	21
<i>M. wolinskyi</i>	MO739 ^T	ATCC	20.1
<i>M. xenopi</i>	ATCC 19250 ^T	LQQB00000000.1	19.6
<i>M. yongonense</i>	05-1390 ^T	NC_021715.1	20.2
<i>Nocardia asteroides</i>	NBRC 15531 ^T	BAFO00000000.2	19.9

Date: 01 NOV 2017

Signature:



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