

**Description of *Mycobacterium saopaulense* sp. nov., a rapidly growing
mycobacterium closely related with members of the *Mycobacterium chelonae*-*M.*
abscessus group.**

Christiane Lourenço Nogueira¹, Christopher M. Whipps², Cristianne Kayoko Matsumoto¹, Erica Chimara³, Sara Droz⁴, Enrico Tortoli⁵, Denise de Freitas⁶, Margo Cnockaert⁷, Juan Carlos Palomino⁷, Anandi Martin⁷, Peter Vandamme⁷, Sylvia Cardoso Leão¹⁺

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¹ Departamento de Microbiologia, Imunologia e Parasitologia, Escola Paulista de Medicina, Universidade Federal de São Paulo, São Paulo, SP, Brazil

² SUNY-ESF, State University of New York College of Environmental Science and Forestry, Environmental and Forest Biology, 1 Forestry Drive, Syracuse, NY, USA

³ Núcleo de Tuberculose e Micobacterioses, Instituto Adolfo Lutz, São Paulo, SP, Brazil

⁴ Division of Bacteriology, University of Bern, Switzerland

⁵ Emerging Bacterial Pathogens Unit, San Raffaele Scientific Institute, Milan, Italy

⁶ Departamento de Oftalmologia, Escola Paulista de Medicina, Universidade Federal de São Paulo, São Paulo, SP, Brazil

⁷ Ghent University, Laboratory of Microbiology, Faculty of Sciences, Ghent, Belgium

⁺ Corresponding author:

Sylvia Cardoso Leão

email: sylvia.leao@gmail.com

Table S1: Comparison of phenotypic and biochemical characteristics of the isolates and the type strains included in this study.

Isolates/strains: 1, EPM10906; 2, EPM10695; 3, IAL 3785; 4, JAN1; 5, JAN2, 6, *M. abscessus subsp. abscessus* ATCC 19977^T; 7, *M. abscessus subsp. bolletii* CCUG 50184^T; 8, *M. chelonae* ATCC 35752^T; 9, *M. immunogenum* ATCC 700505^T; 10, *M. salmoniphilum* ATCC 13758^T; 11, *M. franklinii* DSM 45524^T

Characteristics	1	2	3	4	5	6	7	8	9	10	11
Colony morphology	SM	SM	SM	SM	SM	SM	SM	SM	RG	SM	SM
Pigmentation	nc	nc	nc	nc	nc	nc	nc	nc	nc	nc	nc
Growth in the presence of:											
Picric acid	+	+	+	+	+	+	+	+	+	+	+
PNB*	+	+	+	+	+	+	+	+	+	+	+
Nitrite	+	+	+	+	+	+	+	+	+	+	+
5% NaCl	+	+	+	+	+	+	+	+	+	+	+
Nitrate reduction	-	-	-	-	-	-	-	-	-	-	-
Tween 80 hydrolysis	-	-	-	-	-	-	-	-	-	-	-
single-source carbon utilization of:											
mannitol	-	-	-	-	-	-	-	-	+	-	-
inositol	-	-	-	-	-	-	-	-	-	-	-
citrate	+	+	+	+	+	-	-	+	+	-	-

*PNB, *p*-aminobenzoic acid; SM, smooth; RG, rough; nc, nonchromogenic

Table S2. Primers used in genotypic characterization of strains and isolates included in this study

Gene	Primers	Sequence 5'-3'	References
16S rRNA	16S-27F*#	AGAGTTTGATCCTGGCTCAG	(Harmsen <i>et al.</i> , 2003)
16S rRNA	800F*	ATTAGATACCCTGGTAG	(Adékambi & Drancourt, 2004)
16S rRNA	16S-907R*#	CCGTCAATTCCTTTGAGTTT	(Harmsen <i>et al.</i> , 2003)
16S rRNA	16R-1492*#	TACGGCTACCTTGTTACGACTT	(Gomila <i>et al.</i> , 2007)
23S rRNA	23S1R*	CCCAAAGCCTATATATTCAGC	(Matsumoto <i>et al.</i> , 2012)
<i>rpoB</i>	MYCOF1*	TCCGATGAGGTGCTGGCAGA	(Macheras <i>et al.</i> , 2011)
<i>rpoB</i>	MYCOR2*	ACTTGATGGTCAACAGCTCC	(Macheras <i>et al.</i> , 2011)
<i>hsp65</i>	Tb11	ACCAACGATGGTGTGTCCAT	(Telenti <i>et al.</i> , 1993)
<i>hsp65</i>	Tb12	CTTGTCGAACCGCATAACCCT	(Telenti <i>et al.</i> , 1993)
<i>hsp65</i>	hsp667F*	GGCCAAGACAATTGCGTACG	(Selvaraju <i>et al.</i> , 2005)
<i>hsp65</i>	hsp667R*	GGAGCTGACCAGCAGGATG	(Selvaraju <i>et al.</i> , 2005)
ITS	Sp1*	ACCTCCTTTCTAAGGAGCACC	(Roth <i>et al.</i> , 2000)
ITS	Sp2	GATGCTCGCAACCACTATCCA	(Roth <i>et al.</i> , 2000)
ITS	23S1R*	CCCAAAGCCTATATATTCAGC	(Matsumoto <i>et al.</i> , 2012)

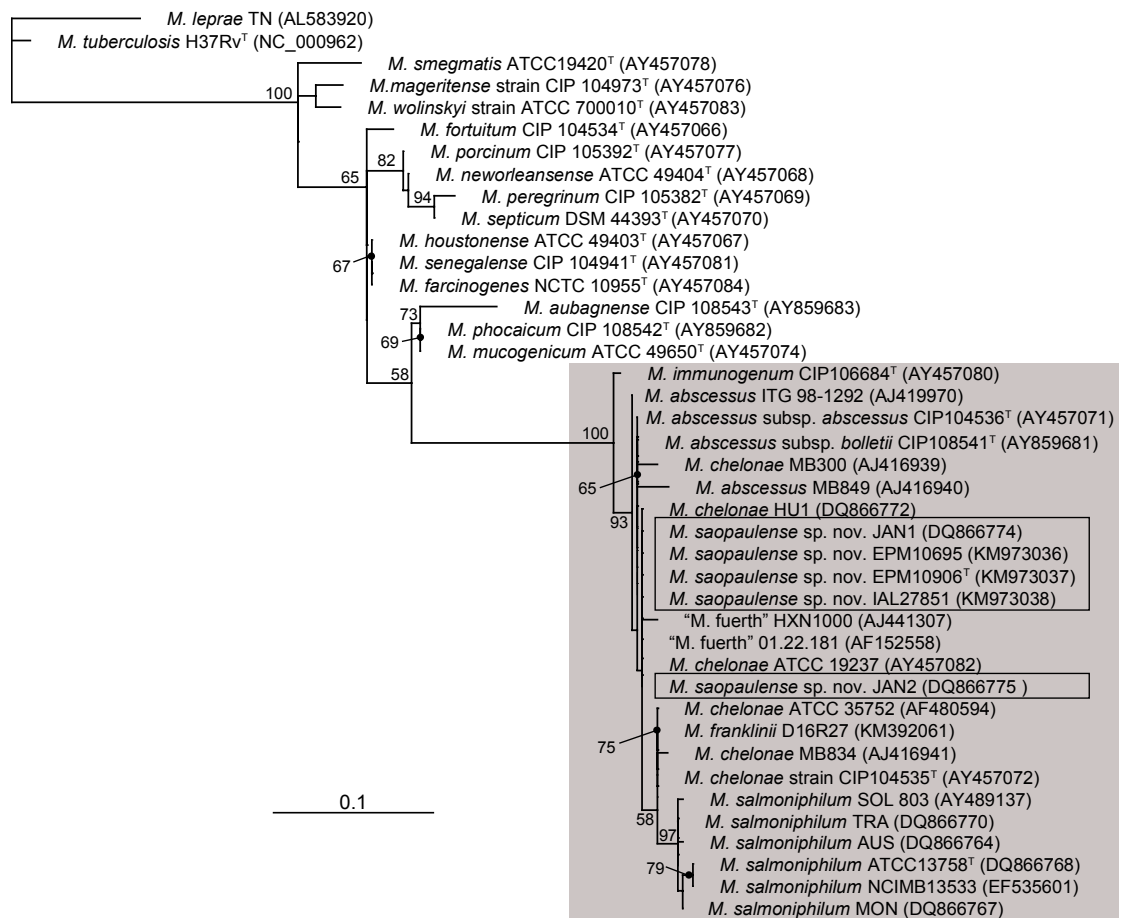
*Primers used for PCR amplification and sequencing of 16S rRNA, *hsp65*, *rpoB* and ITS

Primers were modified based on *M. abscessus* subsp. *abscessus* ATCC 19977 genome

Table S3. Species, strains and GenBank accession numbers of sequences used for phylogenetic analyses. Number of characters in each DNA sequence alignment shown in final row.

<i>Mycobacterium</i> species	Strain	16S rRNA	<i>hsp65</i>	<i>rpoB</i>
<i>M. abscessus</i> subsp. <i>abscessus</i>	CIP 104536 ^T	AY457071	AY458075	AY147164
<i>M. abscessus</i> subsp. <i>boletti</i>	CIP 108541 ^T	AY859681	AY859675	AY859692
<i>M. aubagnense</i>	CIP 108543 ^T	AY859683	AY859677	AY859694
<i>M. chelonae</i>	CIP104535 ^T	AY457072	AY458074	AY147163
<i>M. fortuitum</i>	CIP 104534 ^T	AY457066	AY458072	AY147165
<i>M. franklinii</i>	DSM 45524 ^T	HQ153090	KM392059	HQ153091
<i>M. immunogenum</i>	CIP 106684 ^T	AY457080	AY458081	AY262739
<i>M. mucogenicum</i>	ATCC 49650 ^T	AY457074	AY458079	AY147170
<i>M. phocaicum</i>	CIP 108542 ^T	AY859682	AY859676	AY859693
<i>M. salmoniphilum</i>	ATCC 13758 ^T	DQ866768	DQ866777	DQ866790
<i>M. saopaulense</i>	JAN1	DQ866774	DQ866786	DQ866800
<i>M. saopaulense</i>	JAN2	DQ866775	DQ866787	DQ866801
<i>M. saopaulense</i>	EPM 10695	KM973036	KM973025	KM973028
<i>M. saopaulense</i>	EPM 10906 ^T	KM973037	KM973026	KM973029
<i>M. saopaulense</i>	IAL 3785	KM973038	KM973027	KM973030
Characters in alignment		1387	401	676

Figure S1. Estimate of phylogeny of *Mycobacterium* species based on small subunit (16S) ribosomal DNA sequences. Names and strains are shown, with Genbank accession numbers in parentheses. A superscript ‘T’ represents sequences from type species. Bootstrap values greater than 50% are shown at nodes. The *M. chelonae*-*M. abscessus* group has 100% bootstrap support and is shown in a grey box. Within the group, most relationships are unresolved using 16S, and species are intermixed with the exception of *M. immunogenum* and *M. salmoniphilum*.



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