

**Table 4. Exported proteins observed in the cell wall-associated fraction of the *tatC* mutant strain identified by either two-dimensional gel electrophoresis and/or multidimensional protein identification technology**

<b>Sco number</b>	<b>Method of observation</b>	<b>Tat substrate prediction</b>	<b>Product</b>	<b>Putative signal peptide</b>
<b>Twin-arginine signal peptides</b>				
SCO0131 <sup>†</sup>	2D		Putative secreted protein (DNase I-like)	MFRRTLPLVLA AAVGLAAAAAGPATAD
SCO0462	MudPIT (mutual)		Putative oxidoreductase	MRIAVIGAGGVGGYFGARLAAAGNEVTFVARGGHLAAIRRSGLVVHSPLGELRTSPDSVVAS
SCO0494	2D and MudPIT (mutual)		Putative iron-siderophore binding lipoprotein (FhuD type)	MLLRTRTKPWRLAAALSAAALGVLLAGCGSDSD
SCO0736	MudPIT (mutual)	TatP and TatFind	Putative secreted protein	MGDIRRRGAVALGV TALVAPLTLALTAAPAQAA
SCO1230	2D and MudPIT (mutual)	TatP	Putative secreted tripeptidylaminopeptidase	LRKSSIRRRATAFGTAGALVTATLIAGAVSAPAASAA
SCO1725	MudPIT (mutual)		Putative secreted hydrolase (similar to scabies esterase)	MRRFRLVGFLSSLVLAAGAALTGAATAQAA
SCO1860	2D and MudPIT (mutual)	TatP	Putative secreted protein	MSLMRAPHACL TGGPTTLNGNTRMPARRLATVAAATALAAGPATLAGAGSAHAT
SCO2116 <sup>†</sup>	2D and MudPIT ( $\Delta$ <i>tatC</i> )		Putative secreted protein similar in parts to bacterial AmpD (amidase)	LSAEHRRNRNKKRLLVYGVASAVVVAATTGTLALASPGLLGLDADQAAAA

SCO2150	MudPIT ( <i>ΔtatC</i> )		Cytochrome C heme-binding subunit	MKKLSARRRHPLAALVVLLLALACTGGLYAAFAPASKAQAD
SCO2217	2D and MudPIT (mutual)	TatFind	Putative secreted protein	MGRNTRKRRTPLATKIVAGAAALAIGGGGLVWAN
SCO2270	2D and MudPIT ( <i>ΔtatC</i> )		Putative membrane protein	MPVRRRRPTALAAAVATAAALGTTALAALGGATAASAA
SCO2446	2D and MudPIT (mutual)		Putative secreted peptidase (pfam00082, Peptidase_S8, Subtilase family)	MSTGPVRRGAALLSAGLVIALLPAGTSAAQ
SCO2505	MudPIT (mutual)		Putative ABC-transporter metal-binding lipoprotein (cd01019, ZnuA, Zinc binding protein)	MNVRRRRISGIAVTAATALGLGTLSACSSDSSAA
SCO2591	2D* and MudPIT ( <i>ΔtatC</i> )		Putative secreted protein (Integrin?)	LFTSRARLRTRRSRLLTCTALAVAAGMLVTTPAVAA
SCO2920	MudPIT (mutual)		Putative secreted protease pfam05547, Peptidase_M6, Immune inhibitor A peptidase M6	MTRRSWTFRTAATTVAFAAAAATFSAAGVAQAD
SCO3540	2D		Proteinase (putative secreted protein)	MDTRRTHRRTRTGGTRFRATLLTAALLATAACSAGGASTSAGSPAAKAA
SCO3667	MudPIT (mutual)	TatP	Putative secreted solute- binding protein	MSSRARRIAISVAASLMTFSLASCGMLGASESSD
SCO4584	2D and MudPIT (mutual)		Putative membrane protein (lolA lipo-protein sorting protein)	MAPIDSDDNTTAGAGEELRAGRKAARYVVPVAVMGVAAATIGLVPALA

SCO5022	MudPIT ( <i>ΔtatC</i> )	TatFind	Putative lipoprotein	MNRRRPTLLAALTLTAAAALTLSACGS
SCO5260	MudPIT ( <i>ΔtatC</i> )		Secreted protein (periplasmic binding protein AtrA)	MTARSTRRTTAAHSRLAAVGAIAVAGALLLTGCGDQTENGD
SCO5447	2D		Putative neutral zinc metalloprotease	MLSSSTPHRRTSHTTSHTSRRTAHRRAAAVALAGVAALIATAVQSGTATAA
SCO5477 <sup>†</sup>	2D and MudPIT (mutual)		Putative oligopeptide- binding lipoprotein	MTTQRTSGRRKQALAAAAVVAALLTTAACGGGGDDDSGGG
SCO5650	MudPIT ( <i>ΔtatC</i> )		Putative membrane protein	MAVLSIISRTASRRGVRALGVVAASAALALSASGNALAC
SCO6096	MudPIT ( <i>ΔtatC</i> )		Putative lipoprotein	MPATSALRRTLAVIAALPLLTLAACGYGSQAK
SCO6108	2D and MudPIT (mutual)		Esterase fusH	MATLIPKKGSTLLNKGIRTRRARGALAGGTVLTAALLTAVPAAQAI
SCO6197	2D and MudPIT (mutual)	TatP	Putative secreted protein	MSTTSVNGSRKRRVLLRRALPVCVAAGVASIVVFGSGSSPDAAAR
SCO6407	2D and MudPIT (mutual)		Putative gamma- glutamyltranspeptidase (putative secreted protein)	MRRPVARNLTVLAVSAAVVVSVGAAAP
SCO6572	2D		Putative glycosyl hydrolase (putative secreted protein)	MRSVGAGRWRRVASQVGRSVTVWAVSTLTMLVLAGILPDFRLQSPDGDSATDIAM TAAVGAGA
SCO6590	2D and MudPIT (mutual)		Putative secreted esterase (trypsin like serine protease)	MSSIRRSRSAVLGAVLTAGSLALSTAAAVAVTGGTPVADSD

SCO6608	2D and MudPIT (mutual)		Putative secreted protein (exopolysaccharide biosynthesis)	MKLRKRRLCAATGVVLGALTALAALPPAAVAS
SCO6736	2D and MudPIT (mutual)		Putative metallopeptidase	MPLRLGAFVHRRFIVPGALGAASLMLAIPASAAA
SCO7657	2D and MudPIT (mutual)	TatP	Putative secreted protein	MTRTRTTRLRRPLL TGSTAVAAMAVTAGLVAATAEPAKAA
SCO7677 <sup>†</sup>	2D and MudPIT (mutual)	TatP and TatFind	Putative secreted solute-binding protein	MSRQIDRRSFLRRGAAGAAALAVGPGLLAACSTDEPGSA
<b>Non twin-arginine signal peptides</b>				
SCO0297 <sup>†</sup>	2D and MudPIT (mutual)		Putative secreted protein	MRSTGPVSRIGRSLAAVTATAAAGAVAALGLAASPAAAA
SCO0324	2D		Putative secreted protein (superfamily Quinoprotein amine dehydrogenase)	MSPKSKRTRAALAALVVAAGSLVTAGAARAE
SCO0472	MudPIT (mutual)		Putative secreted protein (Quinoprotein amine dehydrogenase)	LNTSIRNRAVTGTALALAVSAVLTACGGDTSSSD
SCO0762 <sup>†</sup>	2D and MudPIT (mutual)		Putative secreted protein	MRNTARWAATLGLTATAVCGPLAGASLAS
SCO2008	MudPIT (mutual)		Putative branched chain amino acid binding protein	MPGKGLIVRQRSLIAITAALAAGALTLTACGS
SCO2096	MudPIT ( $\Delta$ tatC)		Putative membrane protein (Transglutaminase/protease-	MMSGRARLAMCAWAATLMAACALLPLVEPATWILQAA

			like)	
SCO2271	MudPIT ( <i>ΔtatC</i> )		Hypothetical secreted protein	MPARLRALVTVLCAAVLAALLPAAAAHAE
SCO2483	2D		Putative secreted protein	MHSTTSRGAVLATLLALIGVFATYRALPAGAD
SCO2892	2D and MudPIT (mutual)		Putative secreted protein	MTRGRDGGAGAPPTKHRALLAAIVTLIVAISAAIYAGASAD
SCO3053	2D and MudPIT (mutual)		Putative secreted esterase (FusH-like)	MSRFRSRAAVTAGLTAIAVAAGCLVVGPVQAA
SCO3176	2D		Putative membrane protein	MRPSSLRSPWTRAAVAGAAAAGLAAAGLAGVAHAA
SCO3487	2D		Putative hydrolase	MTVHKRACTTPPPRASRSFRVRWPVLIAAACAGLVLATTSPPAVAA
SCO3966	MudPIT ( <i>ΔtatC</i> )		Putative secreted protein (thioredoxin-like)	MRKKTFAAALLAAACLTLACGSGADSD
SCO4589	2D		Putative aminopeptidase (putative secreted protein)(Zn dependent)	MQLLPSGRALTAGAVAVVTLMAGGSAAGAASAPVHPATPTGTAAAAAA
SCO4891	MudPIT ( <i>ΔtatC</i> )		Putative secreted protein	MRTRTKLTTVLASGTLAAGLLVGGASSASAG
SCO5113 <sup>†</sup>	2D and MudPIT (mutual)		BldKB, putative ABC transport system lipoprotein	MSILRNRTATAAIVAVAAGALTLTACGGGSDSSAK
SCO5232	MudPIT ( <i>ΔtatC</i> )		Putative sugar transporter sugar binding protein	MKRKLIAAIGIAGMMVSIACGG

SCO5420	2D and MudPIT ( <i>ΔtatC</i> )		Cholesterol esterase	MESQVRGGTRWKRFVVMVPSVAATAAIGVALAQQGALAA
SCO5660	MudPIT (mutual)		Putative secreted peptidase	MRARSHAVLAASVVLALAAGPLAAPFAA
SCO5776	MudPIT (mutual)		Glutamate binding protein	MKLRKVSAAAVLALALTATACGG
SCO6109	2D and MudPIT (mutual)		Putative secreted hydrolase (fusH-like)	MFAMPRSARLTALSAALLAGALASSSTPALATAGA
SCO6199	2D and MudPIT (mutual)		Secreted esterase	MHRTRPGGHPRSIRFATLAISLTAGSALMTASPAVAVTGPAAAD
SCO6591	2D and MudPIT (mutual)		Putative secreted protein	MKKRSSFVRVLGAAATAGALAWAVLAQGNAGAA
SCO6710	2D		Putative glycosyl hydrolase	MWAALLAGLLMVLGLQATSASGQPAKAPAAAAA
SCO6723	2D and MudPIT (mutual)		Putative oxidoreductase (putative secreted protein) (sialidase?)	MALAGAVCAAALAALTAVPAQAH
SCO7250	2D and MudPIT (mutual)		Conserved hypothetical protein (Amidase_2, N-acetylmuramoyl-L-alanine amidase)	MPALATAALLPLLGAAPSAAAPTAAP

Proteins are listed by SCO number and the signal peptide sequences are given in the right column. Method of observation indicates which technique was used to identify a particular protein (2D is two-dimensional gel electrophoresis; MudPIT is multidimensional protein identification technology. Bracketed after MudPIT is indicated '*ΔtatC*' if the protein was found in the cell-wall fraction of the *ΔtatC* mutant

strain only, and 'mutual' if it was found in the cell-wall fractions of both the M145 and the  $\Delta tatC$  mutant strains. The column annotated 'Tat substrate prediction' indicates whether a particular protein has been identified as a putative Tat substrate by either of the two prediction programs TatFind or TatP.

\*This protein migrated in a unique position on two-dimensional gel analysis of the extracellular fraction from the M145 strain. However it was detected in the *tatC* mutant strain by MudPIT analysis.

† These proteins have also been identified in the extracellular fraction of *S. coelicolor* grown in liquid medium (Kim DW, Chater K, Lee KJ, Hesketh A (2005) *J Bacteriol* 187: 2957-2966).