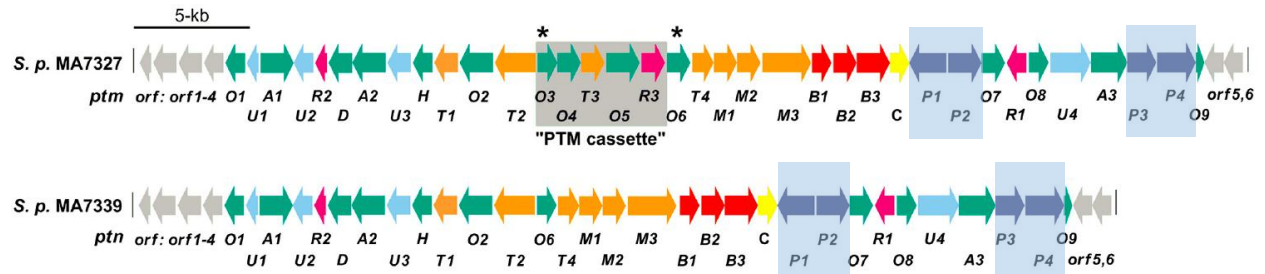
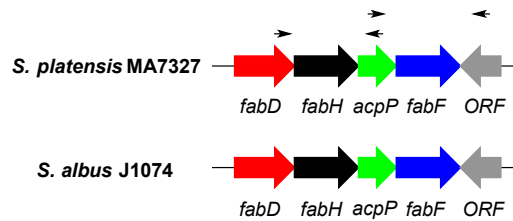


**Figure S1, related to Experimental Procedures and Fig. 1.** Genetic organization of the PTM and PTN biosynthetic clusters in *S. platensis* MA7327 and MA7339 and the FASII loci in *S. platensis* MA7327 and *S. albus* J1074. (A) Genetic organization of the PTM and PTN biosynthetic cluster in *S. platensis* MA7327 and MA7339. Four self-resistance candidates are shaded in light blue and are conserved in both *ptm* and *ptn* clusters. Functional annotations of the four genes are summarized in Table S1. (B) Genetic organization of the FASII loci in *S. platensis* MA7327 and *S. albus* J1074. The *fabH* and *fabF* genes were amplified and sequenced using primers based on homologous regions within the *fab* operons of *S. coelicolor*, *S. avermitilis*, and *S. griseus* (Table S4). Primers are depicted as small black arrows. The ORF downstream of *fabF* is a putative gene encoding a protein homologous to 3-hydroxyisobutyrate dehydrogenases from *Streptomyces* spp.

**A**



**B**



**Fig. S2, related to Experimental Procedures and Fig. S2B.** Protein sequence alignment of *Streptomyces* spp. and *E. coli* FabF and *Streptomyces platensis* MA7327 and MA7339 PtmP3. The conserved catalytic triad of Cys162, His300, and His334 are identified with red asterisks. Conserved residues throughout all protein sequences are shown in red text with yellow highlight; conserved residues to the consensus sequence are shown in blue text with cyan highlight; similar residues to the consensus sequence are shown in blue text with no highlight.

Consensus	VSATNRTVVVTGIGATTEPLGGDAASTWEGLLAGRSGVRPLXQEWAAADLPV	
<i>S. platensis</i> MA7327 PtmP3	-MADNGQVLVTGIGAMTEVGDASSSSWSGLLAGKSGVRAIVEEWAADLPV	49
<i>S. platensis</i> MA7339 PtmP3	-MAENGGQVLVTGIGAMTEVGDASSSSWSGLLAGKSGVRAIVEEWAADLPV	49
<i>S. platensis</i> MA7327 FabF	VNATNRTVVVTGIGATTEPLGGDSASSWEAMLAGRSGVGTLDQDWAALPVE	50
<i>S. albus</i> J1074 FabF	VSPNTNRTVVVTGIGATTEPLGGDAASTWEGLIAGRSGVRPLDDERYADLPV	50
<i>S. avermitilis</i> FabF	VSPNTNRTVVVTGIGATTEPLGGDVASTWEGLVAGRSGVRPLEQEWAEEQAV	50
<i>S. coelicolor</i> FabF	VSSNTNRTVVVTGIGATTEPLGGDAASTWEGLVAGRSGVRPLEQEWAADQAV	50
<i>S. griseus</i> FabF	VNSTNRTVVVTGIGATTEPLGGDSASTWEGLMAGRSGVRFLEGERFAELPVE	50
<i>E. coli</i> FabF	--MSKFRVVVTGLGMLSEVGNVSESTWKALLAGQSGISLIDHFDTSAYAT	48
Consensus	RIAAQAAVDPAAE-VLPRPQARKLDRSAQFALIAAREAWADAGXTXXAGED	
<i>S. platensis</i> MA7327 PtmP3	SLAAGLTEDPAA-ALPRTQARKLDRGEQLAMLSREAWQDAG-----A	91
<i>S. platensis</i> MA7339 PtmP3	GIAAGLTEDPAA-ALPRTQARKLDRGEQLAMLSREAWQDAG-----A	91
<i>S. platensis</i> MA7327 FabF	RIAAQAAVDPAD-VLPRPQARKLDRSAQFALIAAREAWADAGFTARAGED	99
<i>S. albus</i> J1074 FabF	RIAAQIAVEPAE-VLPRFLVRKLDLSAQFALIAAREAWADAGYTAMAGED	99
<i>S. avermitilis</i> FabF	RIAAQIAVEPGE-VIPRPQARRLDRSAQFALIAAKEAWADAGFTDKAGAD	99
<i>S. coelicolor</i> FabF	RIAAQAAVDPSE-VIPRPQARRLDRSAQFALIAAQEAWKADAGYAGKAGES	99
<i>S. griseus</i> FabF	RIAAQAAVDPGD-VLPRFLARKLDRSAQFALIAAREAWADAGFTAKAGED	99
<i>E. coli</i> FabF	KFAGLVKDFNCEDIISRKEQRKMDAFIQYGIVAGVQAMQDSC-----LEI	93
Consensus	PEVD-----PERLGAVIASGIGGVTTLLDQYDVLKEKGVRRVSPHTVPM	
<i>S. platensis</i> MA7327 PtmP3	PEVE-----PERLAVVIGTGTGGVLTTLGQDDLFEESGARRLSFFAVPMP	135
<i>S. platensis</i> MA7339 PtmP3	PEVE-----PERLAVVIGTGTGGVLTTLGQDDLFEESGARRLSFFAVPMP	135
<i>S. platensis</i> MA7327 FabF	TSVD-----PDRLGAVIASGIGGVTTLLGQYDVLKEKGVRRVSPHTVPM	143
<i>S. albus</i> J1074 FabF	EQIS-----PERLGTVIASGIGGVTTLLDQYDVLKEKGVRRVSPHTVPM	143
<i>S. avermitilis</i> FabF	PAVD-----PDRLGTVIASGIGGVTTLLDQYDVLKEKGVRRVSPHTVPM	143
<i>S. coelicolor</i> FabF	PAEDGAAHVDPDRLGAVIASGIGGVTTLLDQYDVLKEKGVRRVSPHTVPM	149
<i>S. griseus</i> FabF	ESVA-----PERLGSVIASGIGGVTTLLDQYDVLKEKGVRRVSPHTVPM	143
<i>E. coli</i> FabF	TEEN-----ATRIGAAIGSGIGGLGLIEENHTSLMNGSPRKISFFVPS	137
*		
Consensus	LMPNGFPXANVGLDVGARAGVHTPVSACASGAEAIGYAIEMIRTGRADV	
<i>S. platensis</i> MA7327 PtmP3	LMPNGFPAAWVSMDLGAKGGARTPVSACASGAEALAMGLDLLRAGRVDVVV	185
<i>S. platensis</i> MA7339 PtmP3	LMPNGFPAAWVSMDLGAKGGARTPVSACASGAEALAMGLDLLRAGRVDVVV	185
<i>S. platensis</i> MA7327 FabF	LMPNGFSANVGLDVNARAGVHTPVSACASGSEAIGYAIEMIRTGRADFVI	193
<i>S. albus</i> J1074 FabF	LMPNSESANVGLVNVNARAGVHTPVSACASGAEAIGYAVEMIRTGRADV	193
<i>S. avermitilis</i> FabF	LMPNGFSANVGLAVGARAGVHTPVSACASGAEAIGYAIEMIRTGRADV	193
<i>S. coelicolor</i> FabF	LMPNGFSANVGLAVGARAGVHTPVSACASGAEAIGYAIEMIRTGRADV	199
<i>S. griseus</i> FabF	LMPNGFPAANVGLVNAQAGVHTPVSACASGAEAIGYAVEMIRTGRADV	193
<i>E. coli</i> FabF	TIVNMVAGHLTIMYGLRGPSSISIATACTSGVHNIGHAARIYAGDADVMV	187
Consensus	AGGTEAAIHPLPIAAFNMAMSKNNDXPXGASRPFDTARDGFVLGEGAG	
<i>S. platensis</i> MA7327 PtmP3	AGGVEACLHPFTIAAFQMKALSTQCENPEAVSRPFDVARSGFVMGEGAG	235
<i>S. platensis</i> MA7339 PtmP3	AGGVEACLHPFTIAAFQMKALSTQCENPEAVSRPFDVARSGFVMGEGAG	235
<i>S. platensis</i> MA7327 FabF	AGGTEAAIHPLPIAAFNMAMSKNNDPQGASRPFDTARNGFVMGEGAG	243
<i>S. albus</i> J1074 FabF	AGGTEAAIHPLPVVAFANMMAMSKNNDPEKASRPYDTGRDGFVLGEGAG	243
<i>S. avermitilis</i> FabF	AGGTEAAIHPLPIAAFNMAMSKNNDPQGASRPYDVARDGFVLGEGAG	243
<i>S. coelicolor</i> FabF	AGGTEAAIHPLPIAAFNMAMSKNNDPQGASRPFDTARDGFVLGEGAG	249
<i>S. griseus</i> FabF	AGGTEAAIHPLPIAAFANMMAMSKNEEPETASRPYDTGRDGFVLGEGAG	243
<i>E. coli</i> FabF	AGGAEKASTPLGVGGFGAARALSTRNDNPQAASRPWDKERDGFVLGDGAG	237

**Fig. S2 continued.**

Consensus	VXVLESAEHAAARGARVYAEXVGGISADXHHIXQPEPEGRGIAHALQNL	
S. platensis MA7327 PtmP3	IMVLERAEFARARGAKTYGTLASAVSSSNHITASDAEG--QVHAIELA	283
S. platensis MA7339 PtnP3	IMVLERAGFARARGATTYGTLAGSAVSSSNHITASDAAG--QVHAIELA	283
S. platensis MA7327 FabF	VIVLESAEHAARKRGARVYAEAVGQGISADSHDVVQPEPESGNGIAHALQDL	293
S. albus J1074 FabF	VVVLESAEHAAARGASVYCEVLGQQLSADSHHIAQPEPTCRGIAAALQHL	293
S. avermitilis FabF	VIVLESAEHAARKRGARVYAEAVGQGISADSHDIVQPEPEGRGISHALQNL	293
S. coelicolor FabF	VLVLESAEHAAARGARVYAEAVGQGISADSHDIVQPEPEGRGISAALQNL	299
S. griseus FabF	VVVLESAEHAAARGARVYCEVLGQQLSADAHHIAQPEPESGRGIAAAMQNL	293
E. coli FabF	MLVLEEYEHAKKRGAKIYAEVLVGFMSSDAYHMTSPENGAALAMANA	287

\*

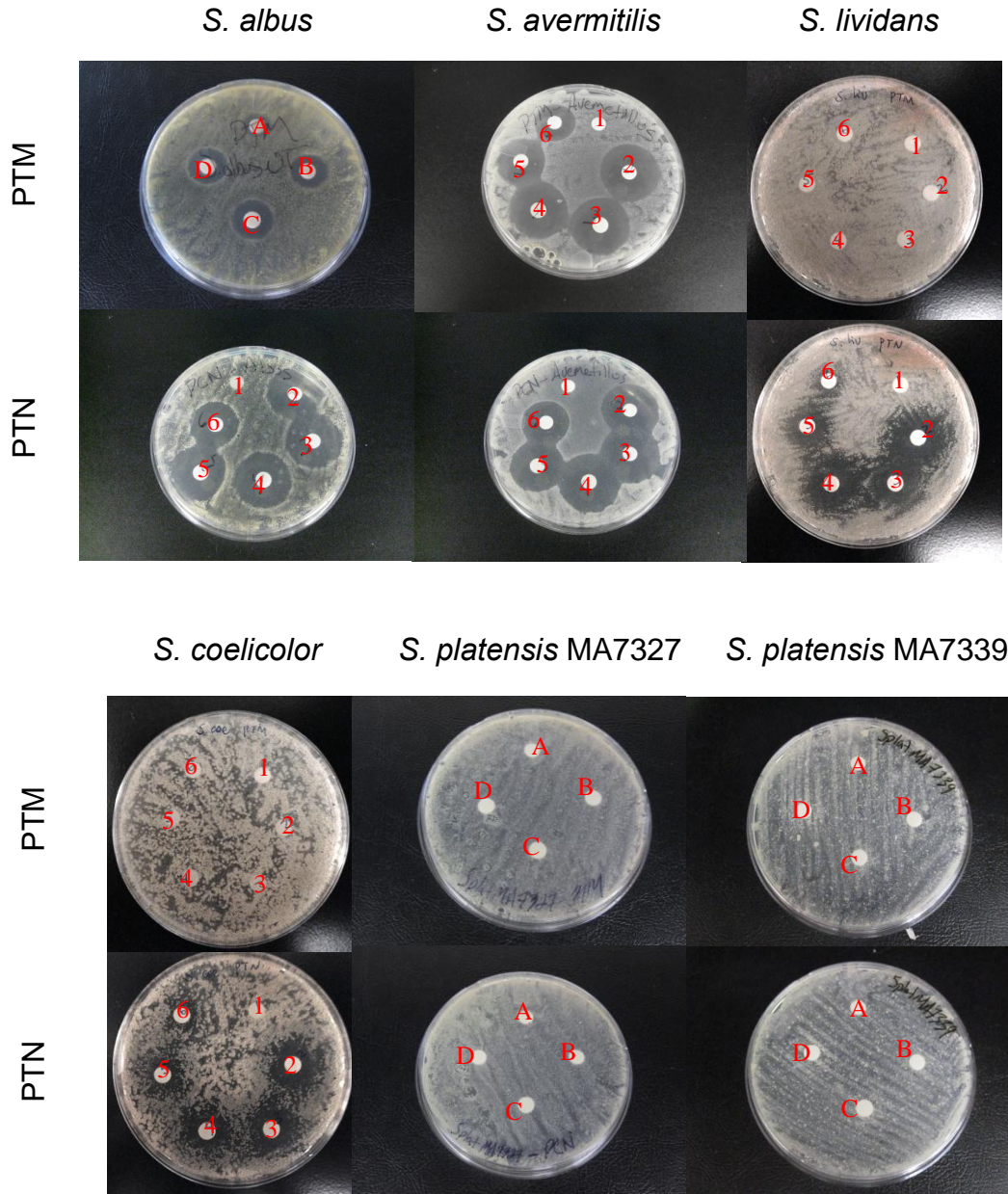
Consensus	LDDADLXPXEIVHVNAHATSTPQGDLAEXKALRKVXGDDXDHVAVSATKS	
S. platensis MA7327 PtmP3	LRDADLTQQDIGHVHAHATSTESGDLAEAEALGRAVCG---THASVTATKS	330
S. platensis MA7339 PtnP3	LRDADLTQRDIGHVHAHATSTESGDLAEAEALGRAVCG---THASVTATKS	330
S. platensis MA7327 FabF	LDSNDLDPAEIVHVNAHATSTPQGDIAELKALRKVFGDDTDHMAVSATKS	343
S. albus J1074 FabF	MDDTDLKPSEVVHLNAHATSTPQGDIAEVLKALRQILGEDLEHVVAISATKS	343
S. avermitilis FabF	LDSTDLNPAEIVHVNAHATSTPAGDVAELKALRKVFGDDADHFVAVSATKS	343
S. coelicolor FabF	LDGNDLDPAEIVHVNAHATSTPAGDIAELKALRKVFGDDVDHMAVSGTKS	349
S. griseus FabF	LDSSDLKPSEVVHLNAHATSTPQGDIAEIKALRKVFGDDLHDHVAISATKS	343
E. coli FabF	LRDAGIEASQIGYVNAHGTSTPAGDKAEAQAVKTIIFGEAASRVLVSSATKS	337

\*

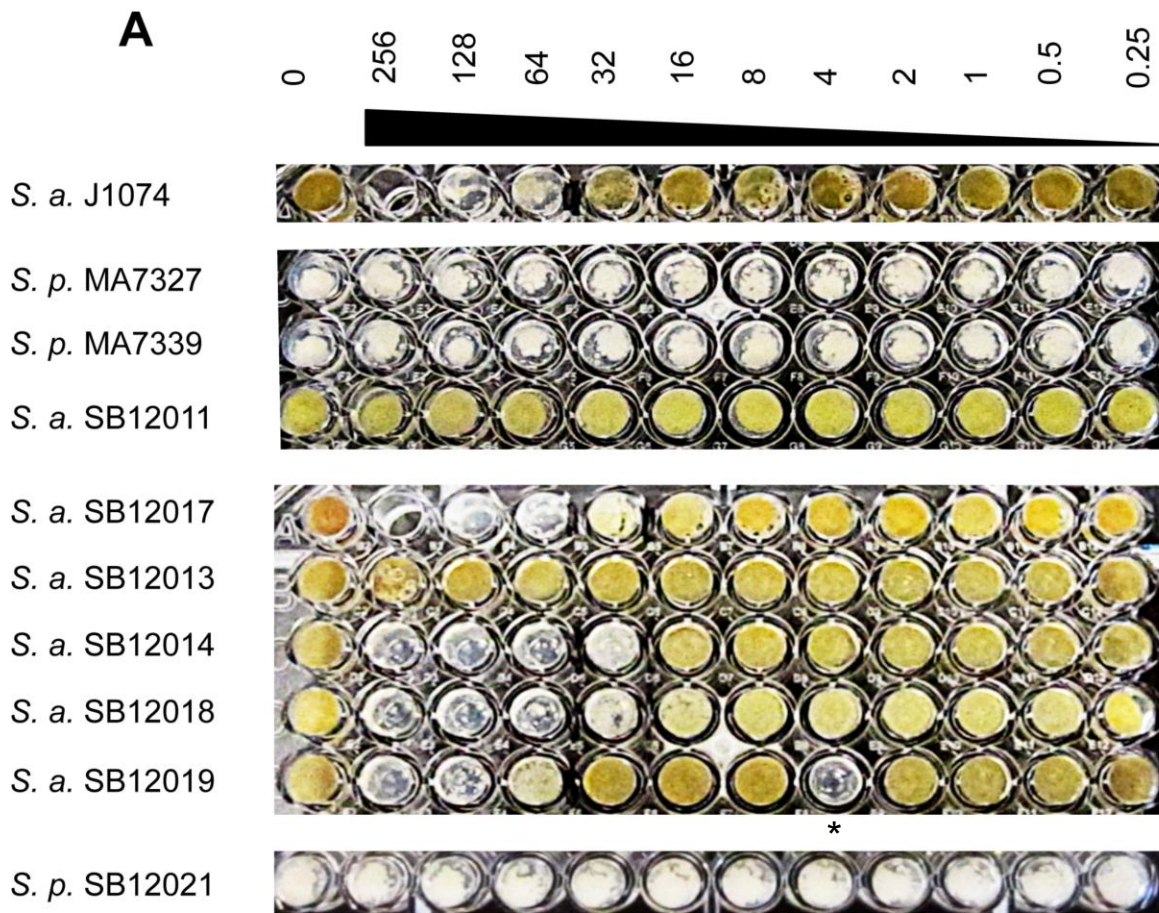
Consensus	MTGHLLGGAGGVEXVAXVLALXHRVAPPTINVDELDPEDID---ADIVRGE	
S. platensis MA7327 PtmP3	MTGHMLGASGAVGALAALLALKDGVAPAIRNLDDELDPEDID---LDVVQGE	377
S. platensis MA7339 PtnP3	MTGHMLGASGAVGAMAALLALKDGVAPAIRNLDDELDPEDID---LDVVQGE	377
S. platensis MA7327 FabF	MTGHLLGGAGGVETVAAILALYHRVAPPTINVDELDPEDID---ADIVRGE	390
S. albus J1074 FabF	MTGHLLGGAGGDIETVATVLAALKHRKAPPTLNVDDELDPATEAGADVVRGE	393
S. avermitilis FabF	MTGHLLGGAGGVESVASVLALYHRVAPPTINVDNLDPEAEAN-ADIVRGE	392
S. coelicolor FabF	MTGHLLGGAGGVESVATVLAALYHRVAPPTINVDNLDPEAEAN-ADIVRGE	398
S. griseus FabF	MTGHLLGGAGGDIETVATVLAALHRRIAPPTINVDDELDEAID---ADIVRGE	390
E. coli FabF	MTGHLLGAAGAVESIYSILALRDQAVPPTINLDNPDEGCD---LDFVPHE	384

Consensus	ARKLP-EGRIAAALNNSFGFGGHNVVLAFRXV	
S. platensis MA7327 PtmP3	NRTGQWN---ASLANSFGFGGHNVSIVFTK	404
S. platensis MA7339 PtnP3	NRTGEWN---ASLANSFGFGGHNVSIVFTK	404
S. platensis MA7327 FabF	PRKLPFAEGRIAAALNDSFGFSGHNVVLAFRTV	421
S. albus J1074 FabF	ARELE-EGTVAAINNSFGFGGHNVVLAFRTV	423
S. avermitilis FabF	ARKLPVEGRIAAALNDSFGFGGHNVVLAFRTV	423
S. coelicolor FabF	ARKLPVEGRIAAALNDSFGFGGHNVVLAFRSV	429
S. griseus FabF	PRVLE-EGPIAAINNSFGFGGHNVVLAFRSV	420
E. coli FabF	ARQVS--GMEYTLCSNFGFGGCTNGSLIFKKI	413

**Fig. S3, related to Experimental Procedures.** Disk diffusion assay for PTM and PTN susceptibility of naïve *Streptomyces* spp. *Streptomyces* spp. were assayed for resistance against PTM and PTN. *S. albus* J1027, *S. avermitilis* SUK-A4, *S. lividans* K4-114, *S. coelicolor* CH999, *S. platensis* MA7327, and *S. platensis* MA7339 were challenged with (1) 0  $\mu\text{g}$ , (2) 200  $\mu\text{g}$ , (3) 150  $\mu\text{g}$ , (4) 100  $\mu\text{g}$ , (5) 50  $\mu\text{g}$ , and (6) 25  $\mu\text{g}$  or (A) 5  $\mu\text{g}$ , (B) 10  $\mu\text{g}$ , (C) 20  $\mu\text{g}$ , and (D) 40  $\mu\text{g}$  of PTM or PTN.

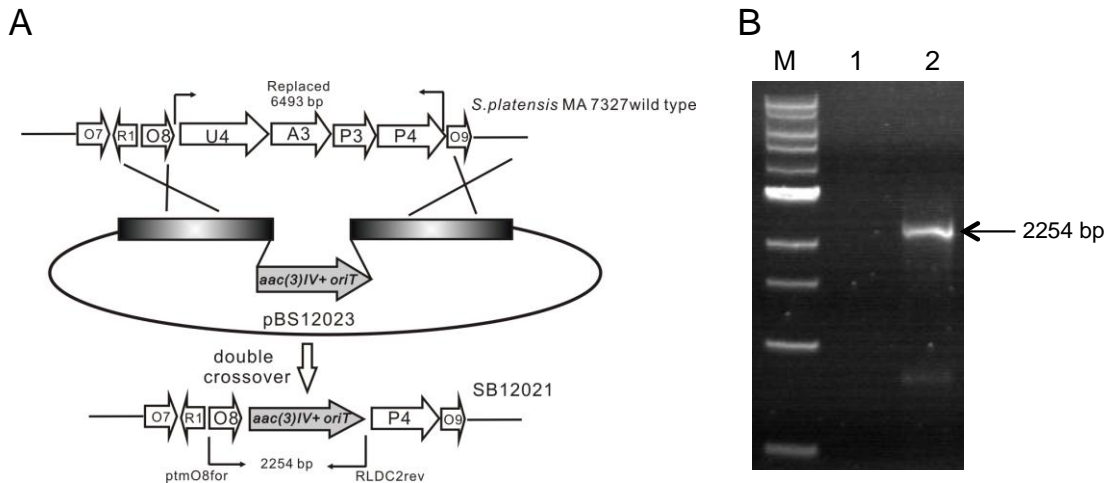


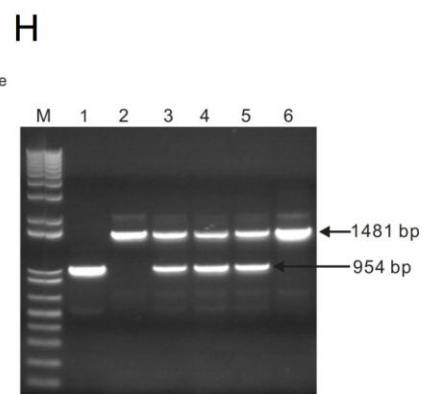
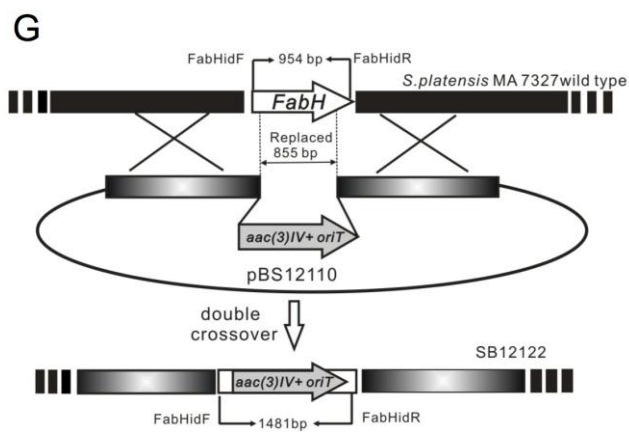
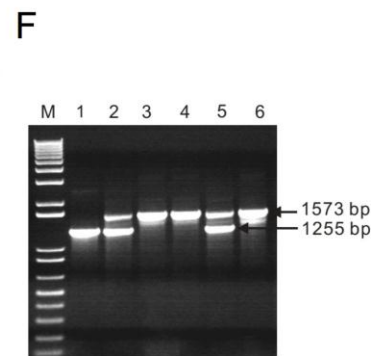
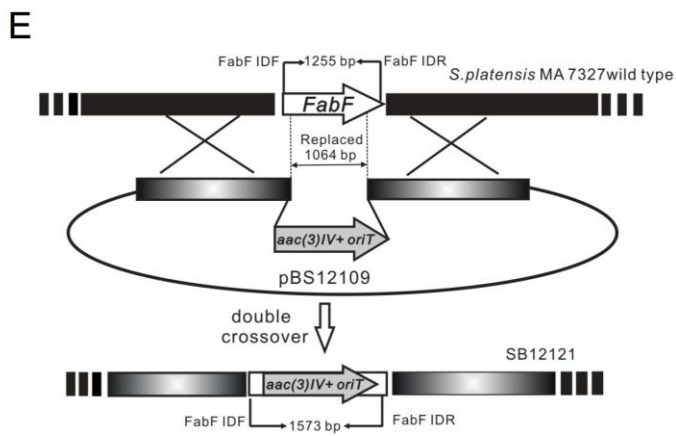
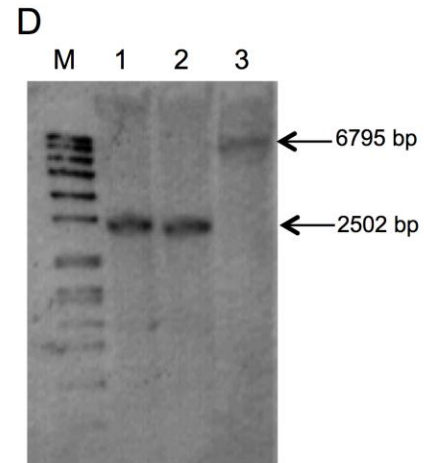
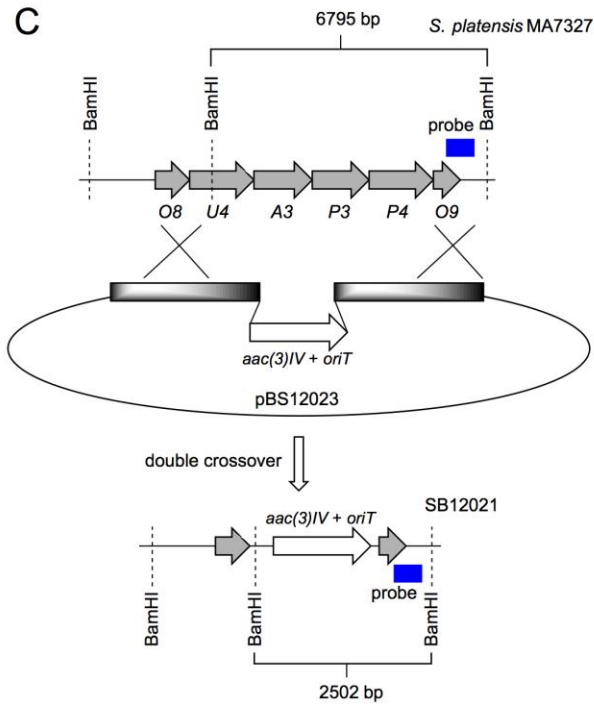
**Fig. S4, related to Table 1.** Minimum inhibitory concentrations (MIC) to PTM and PTN for selected *Streptomyces* species by 96-well plate assay. *S. albus* J1074, *S. platensis* MA7327, *S. platensis* MA7339, *S. albus* SB12011, *S. albus* SB12017, *S. albus* SB12013, *S. albus* SB12014, *S. albus* SB12018, *S. albus* SB12019, and *S. platensis* SB12021 (rows 1-10, respectively) challenged with PTM (panel A) or PTN (panel B). Final concentrations of PTM and PTN were 0, 256, 128, 64, 32, 16, 8, 4, 2, 1, 0.5, and 0.25  $\mu\text{g/mL}$  (columns 1-12, respectively) and 0, 64, 32, 16, 8, 4, 2, 1, 0.5, 0.25, 0.125, and 0.0625  $\mu\text{g/mL}$ , respectively (columns 1-12, respectively). The asterisk denotes a well (i.e., *S. a.* SB12019 at 4) with poor growth; however, it was ignored as the adjacent wells showed no inhibition.



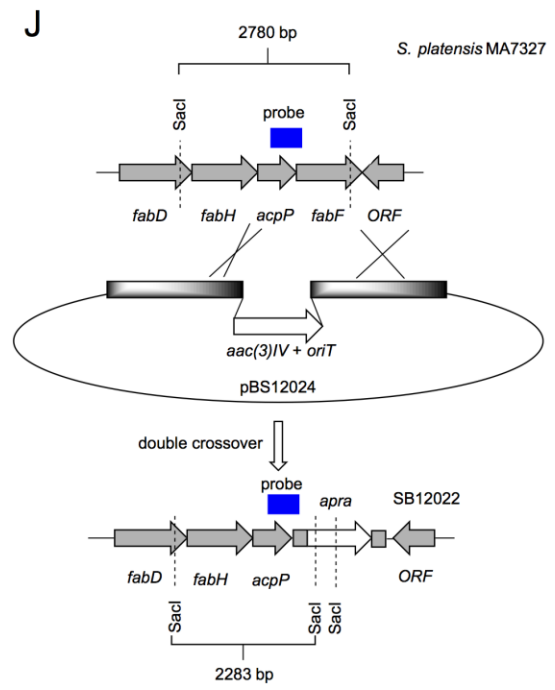
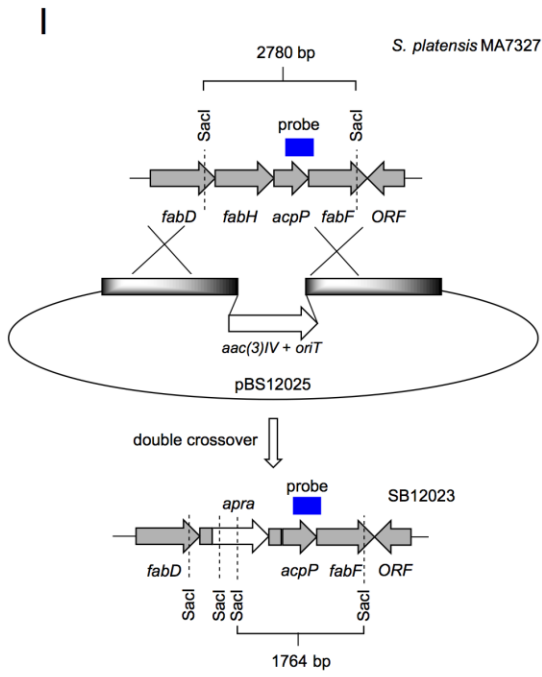


**Fig. S5, related to Experimental Procedures and Table 1.** Construction and Southern analyses of the  $\Delta ptmU4-ptmP4$  mutant *S. platensis* SB12021, the  $\Delta fabF$  mutant *S. platensis* SB12022, the  $\Delta fabH$  mutant of *S. platensis* SB12023. (A) Schematic representation of the deletion of *S. platensis ptmU4-ptmP4* by insertion of an apramycin resistance-oriT cassette (*aac(3)IV+oriT*). (B) PCR verification of WT and double crossover mutant genotypes, using the primers ptmO8for and RLDC2rev. Lane 1, *S. platensis* MA7327; lane 2, *S. platensis* SB12021. (C) Schematic representation of the Southern analysis constructs for *S. platensis* SB12021. (D) Southern blot verification of WT (6795 bp) and double crossover (2502 bp) mutant genotypes. Lanes 1-2, *S. platensis* SB12021; lane 3, *S. platensis* MA7327. (E) Schematic representation of the inactivation of *S. platensis fabF*. (F) PCR verification of WT, single crossover mutant, and double crossover mutant genotypes, using the primers FabF\_IDF and FabF\_IDR. Lane 1, *S. platensis* MA7327; lanes 2 and 5, single crossover mutants; lanes 3 and 4, double crossover mutants, *S. platensis* SB12022 and SB12024; lane 6, pBS12024 positive control. (G) Schematic representation of the inactivation of *S. platensis fabH*. (H) PCR verification of WT, single crossover mutant, and double crossover mutant genotypes, using the primers FabF\_IDF and FabF\_IDR. Lane 1, *S. platensis* MA7327; lane 2, double crossover mutants, *S. platensis* SB12023 (or SB12025); lanes 3-5, single crossover mutants; lane 6, pBS12025 positive control. (I) Schematic representation of the inactivation of *S. platensis fabH*. (J) Schematic representation of the inactivation of *S. platensis fabF*. (K) Southern blot verification of WT (2780 bp) and double crossover mutants of *fabH* (1764 bp) and *fabF* (2283 bp) genotypes. Lanes 1 and 4, *S. platensis* MA7327; lane 2, *S. platensis* SB12023; lane 3, *S. platensis* SB12022.

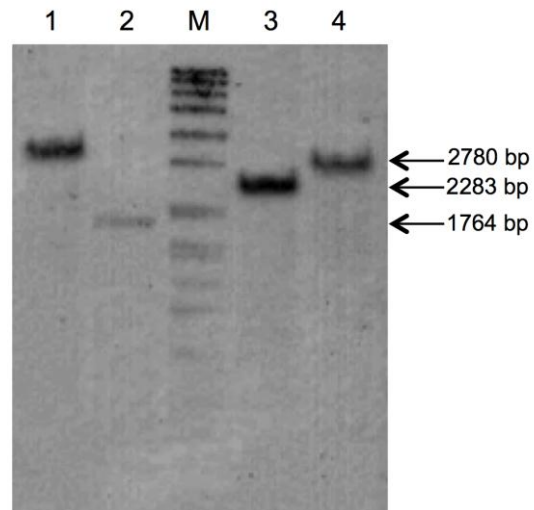








**K**



**Table S1, related to Fig. 2A.** Functional analysis of four candidate resistance genes from the *ptm/ptn* gene clusters in *Streptomyces platensis* MA7327 and *Streptomyces platensis* MA7339.

Gene	# of AA*	PTM/PTN % Identity	Protein homolog	% Identity/ % Similarity	Proposed function
<i>ptmP1/ptnP1</i>	474	96	PhIA (AAB48109)	23/34 <sup>§</sup>	Putative resistance
			PhIB (AAB48107)	21/36 <sup>¶</sup>	Unknown
<i>ptmP2/ptnP2</i>	414	99	PhIC (AAB48108)	22/35	Putative resistance
<i>ptmP3/ptnP3</i>	405	96	TiKASII (1J3N_A)	46/63	Putative resistance
<i>ptmP4/ptnP4</i>	520	97	Pep (AAG31689)	30/48	Efflux pump

\*AA, amino acids.

<sup>§</sup>Values from alignment of full-length PhIA with residues 1-334 of PtmP1.

<sup>¶</sup>Values from alignment of full-length PhIB with residues 335-474 of PtmP1.

**Table S2, related to Experimental Procedures.** Strains and plasmids used in this study.

Strain	Genotype, Description	Reference/Source
<i>E. coli</i> DH5 $\alpha$	<i>E. coli</i> host for cloning	Commercial
<i>S. platensis</i> MA7327	Wildtype PTM/PTN producer	(Singh, et al., 2006; Wang, et al., 2006)
<i>S. platensis</i> MA7339	Wildtype PTN producer	(Singh, et al., 2007; Wang, et al., 2007)
<i>S. platensis</i> SB12001	PTM/PTN overproducing strain	(Smanski, et al., 2009)
<i>S. platensis</i> SB12002	PTM/PTN overproducing strain	(Smanski, et al., 2009)
<i>E. coli</i> ET12567/pUZ8002	Methylation-deficient <i>E. coli</i> host for intergeneric conjugation; contains pUZ8002, a nontransmissible <i>oriT</i> mobilizing plasmid	(MacNeil, et al., 1992)
<i>E. coli</i> BW25113/pIJ790	<i>E. coli</i> host for PCR targeting	(Gust, et al., 2003)
<i>E. coli</i> BW25113/pIJ790/pBS12005	<i>E. coli</i> host for PCR targeting of the <i>ptm</i> cluster	This study
<i>S. albus</i> J1074	Naïve host for expression constructs	(Chater and Wilde, 1980)
<i>S. avermitilis</i> SUK-A4	Naïve host for susceptibility assay	(Komatsu, et al., 2010)
<i>S. lividans</i> K4-114	Naïve host for susceptibility assay	(Ziermann and Betlach, 1999)
<i>S. coelicolor</i> CH999	Naïve host for susceptibility assay	(Kieser T, et al., 2000; McDaniel, et al., 1993)
<i>S. albus</i> SB12009	<i>S. albus</i> J1074 harboring pBS12016	This study
<i>S. albus</i> SB12010	<i>S. albus</i> J1074 harboring pBS12017	This study
<i>S. albus</i> SB12011	<i>S. albus</i> J1074 harboring pBS12018	This study
<i>S. albus</i> SB12012	<i>S. albus</i> J1074 harboring pBS12019	This study
<i>S. albus</i> SB12013	<i>S. albus</i> J1074 harboring pBS12020	This study
<i>S. albus</i> SB12014	<i>S. albus</i> J1074 harboring pBS12021	This study
<i>S. albus</i> SB12015	<i>S. albus</i> J1074 harboring pBS12026	This study
<i>S. albus</i> SB12016	<i>S. albus</i> J1074 harboring pBS12027	This study
<i>S. albus</i> SB12017	<i>S. albus</i> J1074 harboring pBS12028	This study
<i>S. albus</i> SB12018	<i>S. albus</i> J1074 harboring pBS12029	This study
<i>S. albus</i> SB12019	<i>S. albus</i> J1074 harboring pBS12030	This study
<i>S. platensis</i> SB12020	<i>S. platensis</i> MA7327 harboring pBS12022	This study
<i>S. platensis</i> SB12021	<i>S. platensis</i> MA7327 $\Delta ptmU4-ptmP4::aac(3)IV$	This study
<i>S. platensis</i> SB12022	<i>S. platensis</i> MA7327 $\Delta fabF::aac(3)IV$	This study
<i>S. platensis</i> SB12023	<i>S. platensis</i> MA7327 $\Delta fabH::aac(3)IV$	This study
<i>S. platensis</i> SB12024	<i>S. platensis</i> SB12020 $\Delta fabF::aac(3)IV$	This study
<i>S. platensis</i> SB12025	<i>S. platensis</i> SB12020 $\Delta fabH::aac(3)IV$	This study
Plasmid	Description	Reference/Source
pGEM-T Easy	TA cloning vector	Promega
pCR2.1-TOPO	TA cloning vector	Invitrogen

**Table S2** continued.

pIJ773	Construct containing the apramycin resistance cassette ( <i>aac (3)IV+oriT</i> )	(Gust, et al., 2003)
pBS12005	Cosmid harboring partial <i>ptm</i> gene cluster	(Smanski, et al., 2011)
pBS12013	The ermE* promotor cloned as an 0.3 kb EcoRI/BamHI fragment from pWHM860pro into the same site of pBS9010 (Galm, et al., 2009)	This study
pBS12014	Cosmid harboring <i>fab</i> cluster from <i>S. platensis</i> MA7327	This study
pBS12015	0.3 kb EcoRI/BamHI fragment from pWHM1250 containing the constitutive ermE* promotor cloned into the same sites of pSET152	This study
pBS12016	pSET152ermE* derivative pBS12015 harboring <i>ptmP1</i>	This study
pBS12017	pSET152ermE* derivative pBS12015 harboring <i>ptmP2</i>	This study
pBS12018	pSET152ermE* derivative pBS12015 harboring <i>ptmP3</i>	This study
pBS12019	pSET152ermE* derivative pBS12015 harboring <i>ptmP4</i>	This study
pBS12020	pSET152ermE* derivative pBS12015 harboring <i>S. platensis</i> MA7327 <i>fabF</i>	This study
pBS12021	pSET152ermE* derivative pBS12015 harboring <i>S. platensis</i> MA7327 <i>fabH</i>	This study
pBS12022	pSET152ermE* derivative pBS12013 harboring <i>ptmP3</i>	This study
pBS12023	pBS12005 $\Delta ptmU4-ptmP4::aac(IV)3$ , generated by PCR targeting with primers RLDC2for and RLDC2rev	This study
pBS12024	pBS12014 $\Delta fabF::aac(IV)3$ , generated by PCR targeting with primers $\Delta FabF$ -Forward and $\Delta FabF$ -Reverse	This study
pBS12025	pBS12014 $\Delta fabH::aac(IV)3$ , generated by PCR targeting with primers $\Delta FabH$ -Forward and $\Delta FabH$ -Reverse	This study
pBS12026	pSET152ermE* derivative pBS12015 harboring <i>ptmP3(C162A)</i>	This study
pBS12027	pSET152ermE* derivative pBS12015 harboring <i>ptmP3(C162Q)</i>	This study
pBS12028	pSET152ermE* derivative pBS12015 harboring <i>ptmP3(C162L)</i>	This study
pBS12029	pSET152ermE* derivative pBS12015 harboring <i>S. albus</i> J1074 <i>fabF</i>	This study
pBS12030	pSET152ermE* derivative pBS12015 harboring <i>S. albus</i> J1074 <i>fabH</i>	This study

**Table S3, related to Experimental Procedures.** Primers used in this study.

Primer	Nucleotide Sequence (5'-3')	Function
PtmP1For	GACCTCGACGTCGCTGAC	Amplification of <i>ptmP1</i>
PtmP1Rev	TGGTTCCGTGCGCAGTAC	Amplification of <i>ptmP1</i>
PtmP2For	CGTGGTGTGATGACCCGTC	Amplification of <i>ptmP2</i>
PtmP2Rev	GCCAGTTCGACGGCCATG	Amplification of <i>ptmP2</i>
PtmP3For	TGTGGAAGGGACACAACCTACC	Amplification of <i>ptmP3</i>
PtmP3Rev	GTGGTCACTTGGTGAACACCAGG	Amplification of <i>ptmP3</i>
PtmP4For	TGTTACCAAGTGACCACGGC	Amplification of <i>ptmP4</i>
PtmP4Rev	CGTCATCACTCACTCCAGGATG	Amplification of <i>ptmP4</i>
PtmP3C162A For	GCCCGTCAGCGCCGCGCTCCGGTGCCG	Site-directed mutagenesis of <i>ptmP3</i>
PtmP3C162A Rev	CGGCACCGGACGCGGCGGCGCTGACGGGC	Site-directed mutagenesis of <i>ptmP3</i>
PtmP3C162Q For	GCCCGTCAGCGCCAGGCGTCCGGTGCCG	Site-directed mutagenesis of <i>ptmP3</i>
PtmP3C162Q Rev	CGGCACCGGACGCGCTGGGCGCTGACGGGC	Site-directed mutagenesis of <i>ptmP3</i>
SeqC162mut Up	GACGACATCTTCGAGGAGTCCG	Sequencing of PtmP3 C162 mutations
SeqC162mut Down	CGGACTCCTCGAAGATGTCTGC	Sequencing of PtmP3 C162 mutations
RLDC2for	ACACCAGCCGTACTIONACGCGGATCGGAGGGCC GAGTCGTGATTCCGGGGATCCGTCGACC	Amplification of <i>ptmU4- ptmP4</i> disruption cassette
RLDC2rev	CTACTTGCCGGGCTGCCGGGTACGGACGAGC AGTCCGGCTGTAGGCTGGAGCTGCTTC	Amplification of <i>ptmU4- ptmP4</i> disruption cassette
ptmO8for	AAAACCTCTATTTCCAGTCGGTAGCGTTAAAT CAGTTAACTAAG	Confirmation of $\Delta$ <i>ptmU4- ptmP4</i> in <i>S. platensis</i>
U4-P4SouthF	GTCGAGGCCATCTTCTACGA	Probe for Southern analysis of $\Delta$ <i>U4-P4</i>
U4-P4SouthR	GAACATCCAGCGGTTCTCC	Probe for Southern analysis of $\Delta$ <i>U4-P4</i>
UpstreamFab H(2)	TGGGACCTGTGCATGGAGAC	Amplification/sequencing of <i>Streptomyces</i> /sequencing <i>fabH</i>
Downstream FabH(1)	GGTCGTCGGTGAAGGACTTGTC	Amplification/sequencing of <i>Streptomyces fabH</i>
Downstream FabH(2)	TCGACCATGGACAGCGAGTCG	Amplification/sequencing of <i>Streptomyces fabH</i>
UpstreamFab F(1)	GGACAAGTCCTTCACCGACGAC	Amplification/sequencing of <i>Streptomyces fabF</i>
UpstreamFab F(2)	GCTGTCCATGGTCGAGGTCCG	Amplification/sequencing of <i>Streptomyces fabF</i>
Downstream FabF(1)	TCCGCTGGCTGCACCAGG	Amplification/sequencing of <i>Streptomyces fabF</i>
Downstream FabF(2)	ACGACGAACTGGAGCCCTTCC	Amplification/sequencing of <i>Streptomyces fabF</i>

**Table S3** continued.

Primer	Nucleotide Sequence (5'-3')	Function
FabFinFor	GCGGCCAGTACGACGTGCTC	Sequencing of <i>Streptomyces fabF</i>
FabFinRev	AGCCGTCTCAAGCTCGTCGTC	Sequencing of <i>Streptomyces fabF</i>
$\Delta$ FabH-Forward	GGCGGCTACCGCCCGACCCGGGTCGTGCCC AACGAGGAGATTCCGGGGATCCGTCGACC	Amplification of <i>fabH</i> disruption cassette
$\Delta$ FabH-Reverse	CAACAGTCGCTCCATGGCGAGCGGAATGGAG GCGGCCGATGTAGGCTGGAGCTGCTTC	Amplification of <i>fabH</i> disruption cassette
$\Delta$ FabF-Forward	ACACCGCTGGGTGGCGACAGCGCTTCGTCCT GGGAGGCCATTCCGGGGATCCGTCGACC	Amplification of <i>fabF</i> disruption cassette
$\Delta$ FabF-Reverse	CTCGGCGGGCAGCTTGCGGGGCTCGCCGCG GACGATGTCTGTAGGCTGGAGCTGCTTC	Amplification of <i>fabF</i> disruption cassette
FabHscreen For	TCCACCGTTTTCGCACTTCA	Isolation of cosmids containing <i>S. platensis fabH</i>
FabHscreen Rev	GGCCGTTGGGCATCAGCAT	Isolation of cosmids containing <i>S. platensis fabH</i>
FabFscreen For	ATCGCACCGTGGTCGTCAC	Isolation of cosmids containing <i>S. platensis fabF</i>
FabFscreen Rev	GGCCGTTGGGCATCAGCAT	Isolation of cosmids containing <i>S. platensis fabF</i>
FabH_IDF	GACCTCGAAGGTCAAGCCC	Confirmation of $\Delta$ <i>fabH</i> in <i>S. platensis</i>
FabH_IDR	GTCGCCAACAGTCGCTCCA	Confirmation of $\Delta$ <i>fabH</i> in <i>S. platensis</i>
FabF_IDF	AACGCGACCAATCGCACCG	Confirmation of $\Delta$ <i>fabF</i> in <i>S. platensis</i>
FabF_IDR	TCCGGAAGGCCAGCACCAC	Confirmation of $\Delta$ <i>fabF</i> in <i>S. platensis</i>
fabFH_South F	CGGATCTTCACCGCAAGTAA	Probe for Southern analysis of $\Delta$ <i>fabF/H</i>
fabFH_South R	GTTACGCGGAATTCTTACTCCA	Probe for Southern analysis of $\Delta$ <i>fabF/H</i>

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