

**Transcriptional analysis of the cell division-related *ssg* genes in *Streptomyces coelicolor* reveals direct control of *ssgR* by AtrA**

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**Table S1. *Streptomyces* strains used in this study.**

<b>Strains</b>	<b>Description</b>	<b>Reference</b>
<i>S. coelicolor</i> M145	Wild type <i>S. coelicolor</i> A3(2)	(Kieser <i>et al.</i> , 2000)
<i>S. coelicolor</i> J2401	M145 $\Delta$ <i>whiA</i>	(Flärdh <i>et al.</i> , 1999)
<i>S. coelicolor</i> J2402	M145 $\Delta$ <i>whiB</i>	(Flärdh <i>et al.</i> , 1999)
<i>S. coelicolor</i> J2400	M145 $\Delta$ <i>whiG</i>	(Flärdh <i>et al.</i> , 1999)
<i>S. coelicolor</i> J2210	M145 $\Delta$ <i>whiH</i>	(Ryding <i>et al.</i> , 1999)
<i>S. coelicolor</i> J2450	M145 $\Delta$ <i>whiI</i>	(Ainsa <i>et al.</i> , 1999)
<i>S. coelicolor</i> C77	M145 <i>whiJ</i> locus 77	(Ryding <i>et al.</i> , 1999)
<i>S. coelicolor</i> L645	M145 $\Delta$ <i>atrA</i>	(Uguru <i>et al.</i> , 2005)
<i>S. coelicolor</i> GSB1	M145 $\Delta$ <i>ssgB</i>	(Keijser <i>et al.</i> , 2003)
<i>S. coelicolor</i>	M145 $\Delta$ <i>slbR</i>	(Yang <i>et al.</i> , 2012a)

**Table S2. Oligonucleotides used in this study.**

Primer name	DNA sequence (5' to 3')*	5' end	Relative to
<i>Primers for semi-quantitative RT-PCR</i>			
PssgA-for	<u>GTCGAATTC</u> ACCATGGCGCGCTGGCGCGAC	-619	<i>ssgA</i>
PssgA-rev	CTGGGATCCCCGGGTCTCGTAGCGCAGCTC	+70	<i>ssgA</i>
PssgR-for	<u>GTCGAATTC</u> GACTGCCGTGGTGGGTGAAGTG	-512	<i>ssgR</i>
PssgR-rev	<u>GTCGGATCC</u> GCCCGCTGCACGGAGCCGATC	+12	<i>ssgR</i>
PssgB-rev	<u>GTCCGAGCTC</u> GCTCTCCCGAGTGATCACTGGTC	-592	<i>ssgB</i>
PssgB-for	<u>GTCGGATCC</u> GCAGCTGACCGTGGTGTGAT	+21	<i>ssgB</i>
PssgC-rev	<u>GTCGGAATTC</u> GTCGACGCCGGGTTACCCGAGGT	-216	<i>ssgC</i>
PssgC-for	<u>GTCGGATCC</u> TGCACGACCAGGGTCTTGTGCAC	+35	<i>ssgC</i>
PssgD-for	<u>GTCGAATTC</u> GTCCCCTGCGTGCCTGCTTCCC	-291	<i>ssgD</i>
PssgD-rev	<u>GTCGGATCC</u> ACTGCTCGATGACGGTGGAC	+22	<i>ssgD</i>
PssgE-for	<u>GTCGAATTC</u> GAGGTCGGGGCGTTGATGAATC	-223	<i>ssgE</i>
PssgE-rev	<u>GTCGGATCC</u> AGGATGTGGGCTCGTGCCTAC	+38	<i>ssgE</i>
PssgF-for	<u>GTCGAATTC</u> CGCGTGGGCCTGACCGGACATGAC	-401	<i>ssgF</i>
PssgF-rev	<u>GTCGGATCC</u> CTCGAGAGCGCCCGTCATCTG	+99	<i>ssgF</i>
PssgG-for	<u>GTCGAATTC</u> CCTCGACCGGGTCCTCGTCAAG	-467	<i>ssgG</i>
PssgG-rev	<u>GTCGGATCC</u> AGGACGAGCCTGAGCTCCAG	+62	<i>ssgG</i>
<i>Primers for qRT-PCR (atrA mutant)</i>			
SCO3926p1	GACCGCACCGACAAGCTG	+297	<i>ssgA</i>
SCO3926p2	GCTCTGTTCCCTCCGCCAG	+401	<i>ssgA</i>
SCO6722p1	GGGAGAAGGTCCAGCAGA	+171	<i>ssgD</i>
SCO6722p2	GCCACCCTGCACTACGAC	+72	<i>ssgD</i>
SCO3925p1	GCTGTTCTTCTCGGTGA	+155	<i>ssgR</i>
SCO3925p2	CCGGTACGTGGCGTAGTA	+290	<i>ssgR</i>
SCO5085p1	TACTTCGCCCTGATCGAC	+279	<i>actII-ORF4</i>
SCO5085p2	CTCGCCAGAGATCCAGTC	+417	<i>actII-ORF4</i>
SCO4735p1	GGCAAGTGGAAAGGTCAAC	+192	<i>rpsL</i>
SCO4735p2	GTCGTAGCGGTTGTCCAG	+302	<i>rpsL</i>
SCO5087p1	CAACGCGTACCACATGAC	+938	<i>actI</i>
SCO5087p2	GTGCGCGTTGATGTAGTC	+1055	<i>actI</i>
<i>Primers for qRT-PCR (slbR mutant)</i>			
5S rRNA_RTL	GTCATAGCGTAGGGGAAACG	+11	5S rRNA
5S rRNA_RTR	GGCGTCCTACTCTCCCACAG	+104	5S rRNA
<i>ssgA</i> _RTL	TGTCCGAGGAGCTCTTTTC	+17	<i>ssgA</i>
<i>ssgA</i> _RTR	GACCTGAAGTCGGATCAGCA	+243	<i>ssgA</i>
<i>ssgB</i> _RTL	CTCGTTGTGTCGAGCGAGT	+36 <sup>a</sup>	<i>ssgB</i>
<i>ssgB</i> _RTR	AGCTGAGAGCGATGCACAC	+267 <sup>a</sup>	<i>ssgB</i>
<i>ssgG</i> _RTL	AGCTCAGGCTCGTCCTGTC	+46	<i>ssgG</i>
<i>ssgG</i> _RTR	GTCTTCGACGGCCACACC	+250	<i>ssgG</i>
<i>ssgR</i> _RTL	CGTGTACTACGCCACGTACC	+269 <sup>b</sup>	<i>ssgR</i>
<i>ssgR</i> _RTR	AGCCGCTTCAGGAGACTGT	+508 <sup>b</sup>	<i>ssgR</i>
<i>Primers for DACA and EMSA</i>			
sco0608_pETma	AATAGAATTC ATGTCCGAGAGCACGATGC		
sco0608_pET24ma	ATATAAGCTTTCAGCTGCCGTTCCGCGCGG		
<i>ssgA</i> for	TGGCCATCTCACTGCCCTCC		
<i>ssgA</i> _Rev_biotin	CGCAGCTCCACCGGGATGCGG		

ssgB_For	TCGTCCGTCCCCGTACGGTCT		
ssgB_Rev_biotinylated	GCAGGTGCAGCTCGCAGCTGA		
ssgG_For	TCGTCCCGTATCTGCTCGG		
ssgG_Rev_biotin	AGCTCCAGCTCGCGTTCCACG		
ssgR_For	GTGAGAGCCGTGGGACAC		
ssgR_Rev_biotin	CACGGAGCCGATCAGAGT		
ssgR_EM SAF	FAMGCGCAGCGACTGGTGAGAG		
ssgR_EM SAR	GTGCGCCATTCGCGGAGTC		
<i>Primers for EMSA (fluorescent probes)</i>			
SCO4119p1	CTACACACCGTGGGAAAGAA	-305	SCO4119
SCO4119p2	FAM-AGCCGCGGGAACCAACGTCG	-126	SCO4119
ssgRp1	GTGAGAGCCGTGGGACAC	-321	<i>ssgR</i>
ssgRp2	FAM-AGCGAATGCGGTGTCAG	-97	<i>ssgR</i>
actII-4(s2)p1	TTGGGACGTGTCCATGTAATCACC	+15	<i>actII-ORF4</i>
actII-4(s2)p2	FAM-TCGTGCCGCCTGAGGAGCAGCAGC	+115	<i>actII-ORF4</i>

\* Underlined sequences indicate non-homologues sequence added to create restriction sites (in italics) at the ends of the PCR fragments. Restriction sites: GGATCC, BamHI; GAATTC, EcoRI; AAGCTT, HindIII; TCTAGA, XbaI; CTCGAG, XhoI. Positions are relative to the translational start of the indicated gene (for *ssgAsg* and *ssgAsc* we refer to the first of the three possible ATG start sites as +1).

<sup>a</sup> based on the re-annotated translational start site for *ssgB*, which lies 66 nt downstream of the start predicted in StrepDB.

<sup>b</sup> based on the translational start site predicted in StrepDB. The true start of *ssgR* is most likely further upstream (see the manuscript).

**Table S3. Identification of peptides by DACA assay.**

Protein (SCO Reference) <i>a</i>	ID	peptide (aa sequence)	MH+ <i>b</i>	z	Time point	Xcorrelation score (peptide ID) <i>c</i>			
						<i>ssgA</i>	<i>ssgB</i>	<i>ssgG</i>	<i>ssgR</i>
SlbR (SCO0608)	1	R.SAAEVTTALSEAHSGR.T	1586,8	2	48h	20.25 (1,3)	15.18* (1,3)		
	2	K.VTIPGFLTFER.T	1279,7	2	72h				
	3	R.ATAQPTLVLEVR.E	1297,7	2					
AdpA (SCO2792)	4	R.LLETSYVDEVAGR.C	1653,8	2	48h	10.17 (4)	20.17 (4,6)		
	5	R.LLVCAGEDGPLR.T	1299,7	2	72h	30.2 (4,5,6)	120.20 (4,5)	20.17* (4,5,6)	
	6	R.TTGGLELTAPQGLEAISR.A	1814,0	2					
HupA (SCO2950)	7	R.SELVAALADR.A	1044,6	2	48h	15.21* (7,9,10)	15.23* (7,9)	10.21* (9)	50.25 (7,8,9,10)
	8	K.VTIPGFLTFER.T	1279,7	2	72h	20.21 (7,9)	15.22* (7,9)	10.22* (7,9)	40.25 (7,8,9,10)
	9	R.NPQTGEPIQIPAGYSVK.V	1798,9	2					
	10	R.TARNPQTGEPIQIPAGYSVK.V	2127,1	3					
FruR (SCO3198)	11	R.MAAAIPPDASLTVVTHSLPIAAR.L	2302,3	3	48h	10.22 (11)	20.26 (11,14)	10.15 (11)	20.22 (11,12)
	12	R.LADHPGIQLHIVGGR.V	1582,9	3	72h	20.18 (11/13)		10.19 (11)	
	13	R.ADVAVVAANGFSVEHGLTTPDLAEAAVKR.A	2908,5	3					
	14	R.AALAELEPAEGLLILDAGSTVAR.M	2139,2	2					
Lsr2 domain protein (SCO3375)	15	K.TYEIDLTTANADK.L	1454,7	2	48h				
	16	K.TYEIDLTTANADKLR.G	1723,9	2	72h		20.20 (15,16)		
sugar diacid regulator (SCO3606)	17	R.FGLVLSHAHAVAVAQGPPEAYDDTAPVTR.V	2892,5	3	48h				
	18	R.AADMLVYPVLAR.D	1318,7	2					
	19	RVAVGRPQSGAGGVVHSYEEALTTLEADRL	2982,5	3	72h	10.19(19)	10.16 (17)		10.15(18)
XRE-family protein (SCO3859)	20	R.LIDDLGLNQAR.L	1227,7	2	48h				
	21	R.VQLLQDLAQVADGVSVAEATER.M	2428,2	3	72h	20.17 (20,21)	30.26 (20,21)		
	22	R.SVAAAGDIIDAADSLSSHPLEAEFLR.V	2742,4	3					
AtrA (SCO4118)	23	R.TALGQEDEPWSALS.R.F	1659,7	2	48h				30.22 (23,24,25)
	24	R.EVFGELGYGAPMEDVAR.R	1839,9	2	72h				40.22 (23,24,25,26)
	25	R.LLDILLEGLR.S	1154,7	2					
	26	R.VGVSDDAGTDGGVGGPGAAAAGDPR.V	2126,0	3					
LexA, SOS regulator (SCO5803)	27	R.GSDQAASVQPTDTAGKPAASYVPLVGR.I	2643,3	3	48h		20.24 (27,28)		
	28	R.IAAGGPILAEESVEDVFPLPR.Q	2180,2	2	72h		30.22 (27,28,29)		
	29	R.RQPVAENGDIVAAMLDEATVTKR.F	2440,3	3					

**Footnotes**

*a* Refers to the StrepDB database reference number

*b* MH+: Calculated m/z of the peptide with z = 1

*c* XCorrelation score: cross-correlation peptide score by SEQUEST database search. Peptide numbers between brackets (Refer to column peptide ID). Default threshold of 10 was used in identifying the peptides.

\* averaged Xcorr for peptides found in both of two independent experiments