

SUPPORTING INFORMATION

Chitin-induced gene expression involved in secondary metabolic pathways in *Streptomyces coelicolor* A3(2) grown in soil

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Table S1. Primers used for real-time RT-PCR.

Gene	Primer name	Sequence	Secondary metabolite gene cluster	Reference
sco3217 (<i>cdaR</i>)	cdaR-F183 cdaR-R302	CTCCGCTAATCAGGTCGTTTCT CGGAAGATTTTGCGCAGTGT		
SCO3230 (<i>cdaSyn</i>)	cdaSyn-F12302 cdaSyn-R12422	GCCCCGGCGTCGAGTCCA GAGGCGGGCCTGTTTCGTTCTG	Calcium-dependent antibiotic (SCO3210-49) Non-ribosomal peptide synthetase (NRPS)	Bentley <i>et al.</i> , 2002
SCO3245 (<i>hcmO</i>)	hcmO-F219 hcmO-R343	CATGGAGGTGCGGGACTGGTC GAGCATGGTGTGCAGGTCGG		
SCO5085 (<i>actII4</i>)	actII4-F485 actII4-R604	TGCGGCTTTTTGGAATGC CGTGCAGGGTCTCGTTCAG		
SCO5077 (<i>actVA2</i>)	actVA2-F136 actVA2-R258	ATCGGCGGGCACAACT CACTTCGATACCGGTACGTT	Actinorhodin (SCO5071-5092) Type II polyketide synthase (PKS)	Bentley <i>et al.</i> , 2002
SCO5087(<i>actI-orf1</i>)	actI-orf1-F250 actI-orf1-R353	GACCCCTCGCCCTACCGTTCA TGCGAGGCCCGGTCCATC		
SCO5877 (<i>redD</i>)	red-F195 red-R319	GGACCACGGACCCAGCCTGTA CCACCAGCGTACGGACCTTGC	Prodiginines antibiotic (SCO5877-98) NRPS; type I modular PKS	Bentley <i>et al.</i> , 2002
SCO5880 (<i>redY</i>)	redY-F147 redY-R261	GGATGCCCCATTCCACTACTT CGCCATCGTGTCGAACAC		
SCO6266 (<i>scbA</i>)	scbA-F221 scbA-R315	TGCTGATCGCCGAGACCC TCGAGGTGGCAGGTGTAGTCC		
SCO6276 (<i>cpkD</i>)	cpkD-F1092 cpkD-R1182	CGACGGCTGCTGGGAGATGG TCCTGCACCTGGGCGATGAAG	Cryptic polyketide synthase (SCO6265-86) Type I modular PKS	Gottelt <i>et al.</i> , 2010
SCO6282 (<i>cpkI</i>)	cpkI-F362 cpkI-R453	AGGCCGAGTACGACAAGGTG CCGGAGGAGATGTTGACGATA		
SCO6430 ^a	SCO6430-F458	TGCAGTCCACCCAGATGTTC		

	SCO6430-R579	CCAGACGGTGACCACGTACA		
SCO6433	SCO6433-F190 SCO6433-R283	GCGGTGTACGAGGAGGACGAG CGGGGACGACGATGGTGT	Unknown NRPS (SCO6429-40)	Challis & Hopwood, 2003
SCO6435	SCO6435-F109 SCO6435-R208	GGCATGGGGGTGTGCACCGTC CGTCCGCTCCCCGTGACCTC		
SCO6764	SCO6764-F649 SCO6764-R744	CCGTTCCCGCTGGACGAACTG GCGTGCAGGGCCTTGTCGATG	Squalene-hopene cyclase cluster (6759-6771)	Challis & Hopwood, 2003
SCO6766 ^a	SCO6766-F35 SCO6766-R91	CTACATACCTGGCCGAACAGAAG CCACGATGAGCGGGAAC		
SCO0489 (<i>cchA</i>) ^a	cchA-F3 cchA-R66	GAGCACCAACCCCTTCGA CTGGCCCTCGTCTGTCAC	Coelichelin (SCO0489-99); NRPS	Patel <i>et al.</i> , 2010
SCO0494 (<i>cchF</i>)	cchF-F216 cchF-R308	GGTCGTCTCCGTGGCTACAC TGATGTCGGGGCTCTGGC		
SCO0381 ^a	SCO0381-F1280 SCO0381-R1351	GCCCGGACATCCGAAGAC CGCTGCGTCCGCTGATCT		
SCO0392	SCO0392-F1117 SCO0392-R1208	CTCGTCTGCCGTGGAACCTG CGGCCTCGACCACGCTCAGTT	Unknown deoxysugar (SCO0381-401)	Challis & Hopwood, 2003
SCO0398	SCO0398-F173 SCO0398-R261	AGGACTCGCGCATCCGGTACA TGCGAGGCCCACTTGAACAGC		
SCO5231 (<i>dasR</i>) ^b	dasR-F247 dasR-R370	TTCGTGCCAAGCCCAAGGT GGTCGTGCGGGTGTAGT	Pleiotropic regulator for antibiotic production and chitin metabolism; nutrient-sensing protein	Rigali <i>et al.</i> , 2006
SCO5820 (<i>hrdB</i>) ^b	hrdB-F hrdB-R	TCGACTACCAAGGGCTACAA ACCATGTGACCGGGATAC	Major sigma factor in streptomycetes	Bentley <i>et al.</i> , 2002
SCO5920	SCO5920 -F SCO5920 -R	AACCGCACACGCACGAACGA TGCCGCCCTTCCGGAGTCG	DEAD-box RNA helicase	Bentley <i>et al.</i> , 2002
SCO5921 (<i>scoF5</i>)	scoF5-F scoF5-R	GTGGCGGGCTGATGTGT TGCGCGATGTCGAAGTTCACC	Cold shock protein	Bentley <i>et al.</i> , 2002

^a The primer was already reported by Tanaka *et al.*, 2010.

^b The primer was already reported by Nazari *et al.*, 2011.

References for Table S1.

1. Bentley SD, Chater KF, Cerdeno-Tarraga AM, Challis GL, Thomson NR, James KD, Harris DE, Quail MA, Kieser H, Harper D, Bateman A, Brown S, Chandra G, Chen CW, Collins M, Cronin A, Fraser A, Goble A, Hidalgo J, Hornsby T, Howarth S, Huang CH, Kieser T, Larke L, Murphy L, Oliver K, O'Neil S, Rabinowitsch E, Rajandream MA, Rutherford K, Rutter S, Seeger K, Saunders D, Sharp S, Squares R, Squares S, Taylor K, Warren T, Wietzorrek A, Woodward J, Barrell BG, Parkhill J, Hopwood DA. 2002. Complete genome sequence of the model actinomycete *Streptomyces coelicolor* A3(2). *Nature* **417**:141-147.

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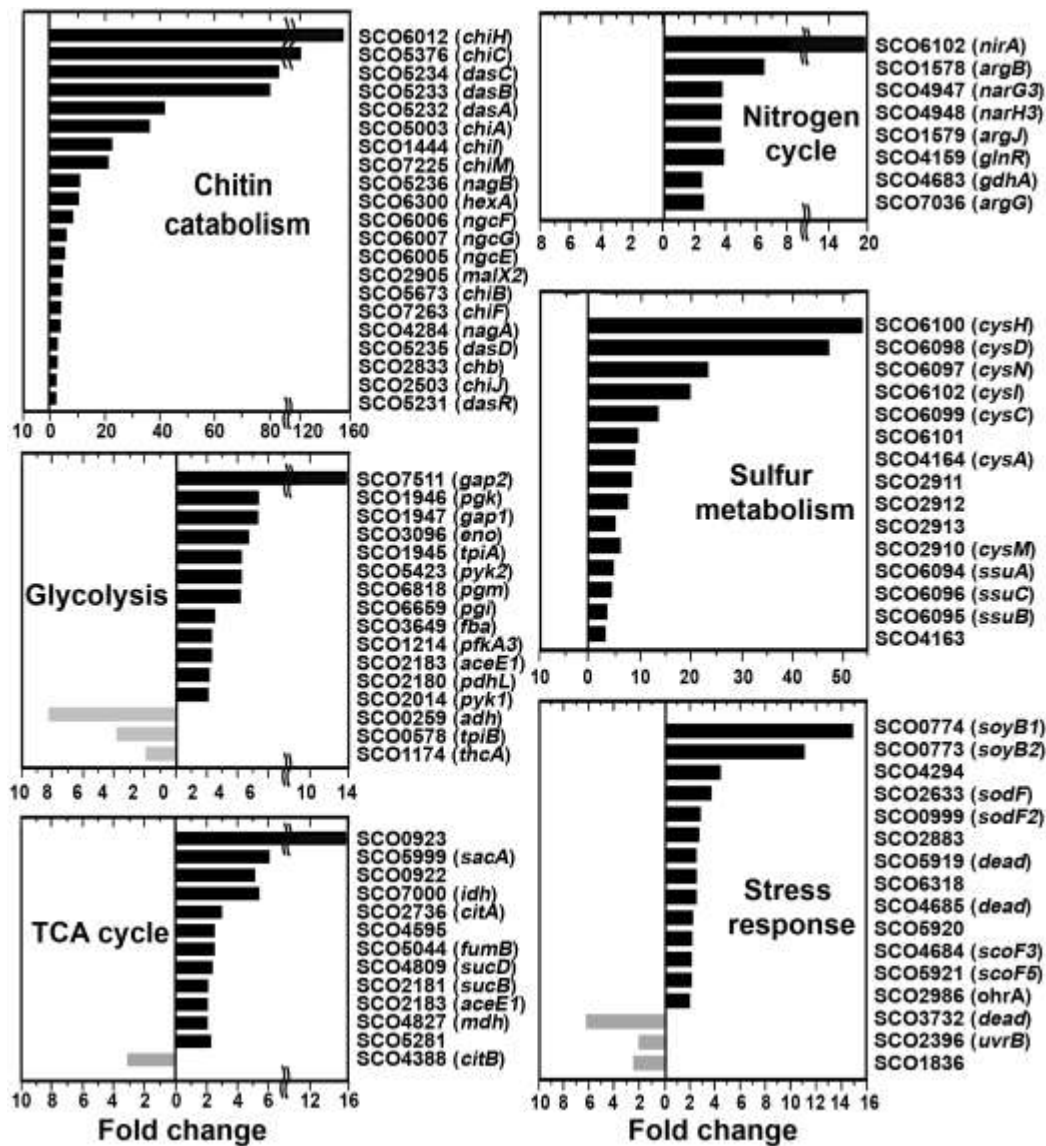


FIGURE S1. Differential upregulation of genes in *S. coelicolor* A3(2) grown in soil with (■) and without (■) chitin. Only genes relevant to the discussion are shown. Supplementary Dataset S1 presents the complete sets of differentially expressed genes. ⌘ indicates genes that were highly expressed on a different scale compared with the other genes.

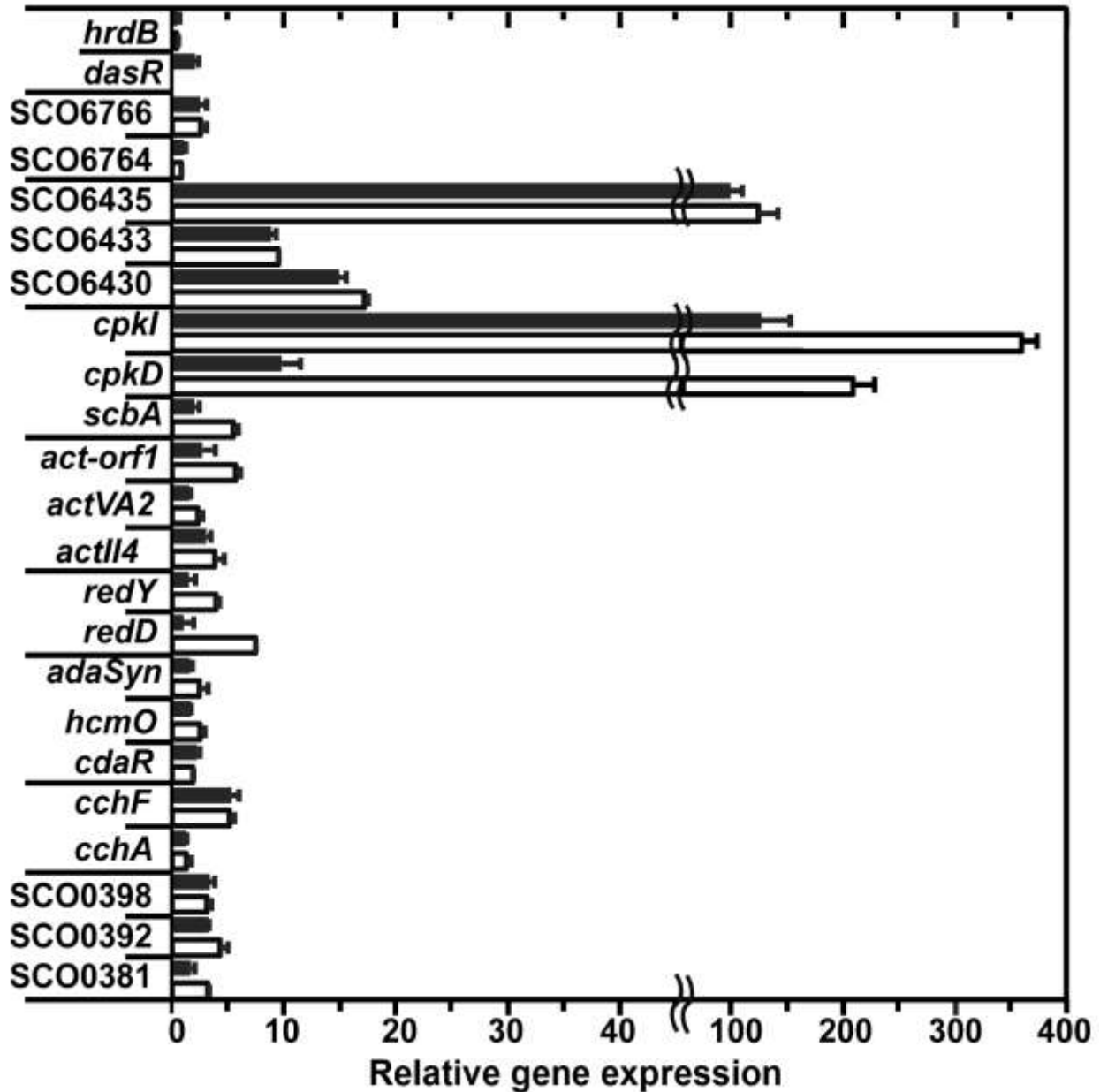


FIGURE S2. Validation of microarray data by qRT-PCR for genes involved in biosynthesis of secondary metabolites. The effects of chitin on gene expression in the wild type and *dasR*-disrupted mutant of *S. coelicolor* A3(2) were investigated in soil at various time points after spore inoculation. The relative gene expression levels in the wild type (■) and mutant (□) grown in chitin-amended soil were calculated taking the expression level in the control (the wild type grown in soil without chitin) as unity (1). In the figure, only the maximum induction levels for each gene during the growth phase are indicated.

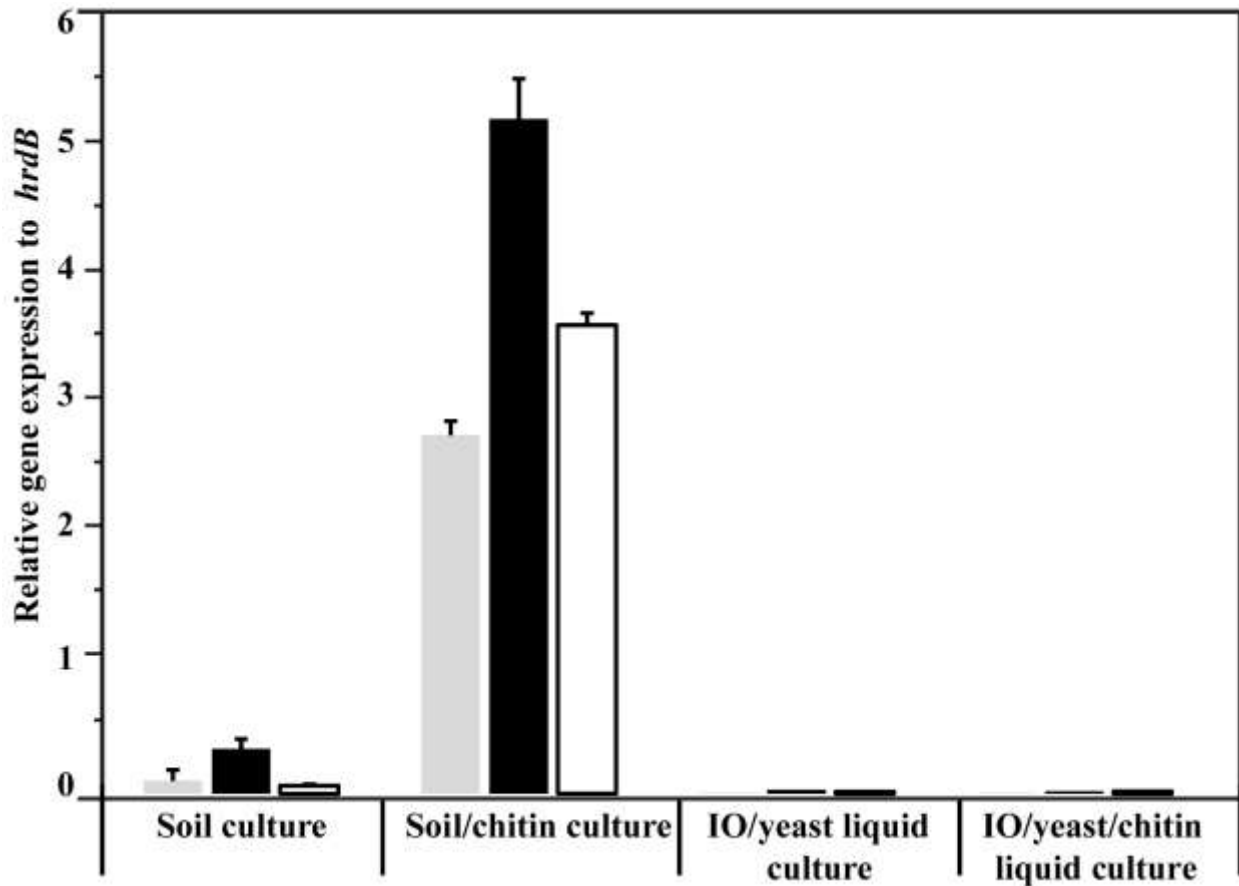


FIGURE S3. Comparison of expression of putative NRPS gene cluster SCO6429-40 in soil and liquid cultures in the presence and absence of chitin. The transcription levels of three genes, SCO6430 (■), SCO6433 (■), and SCO6435 (□), in *S. coelicolor* A3(2) grown in soil and liquid cultures were investigated using qRT-PCR. These genes were only induced in soil cultures and the addition of chitin enhanced their expression.

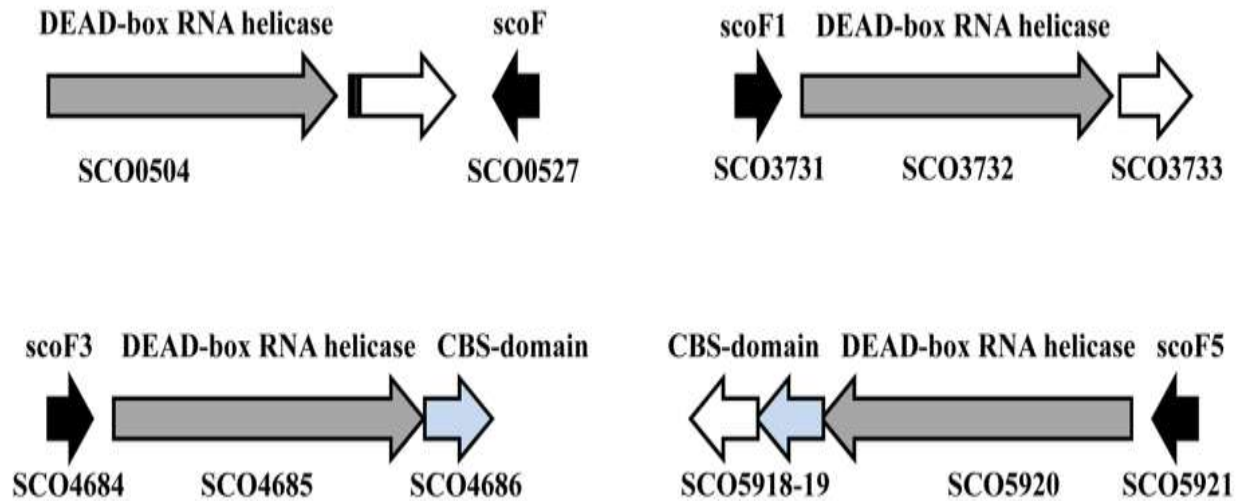


FIGURE S4. Organization of the genes for DEAD-box RNA helicases (gray arrows) and cold shock proteins (black arrows) in the genome of *S. coelicolor* A3(2). Expression of all genes indicated was dramatically reduced in the *dasR* mutant.

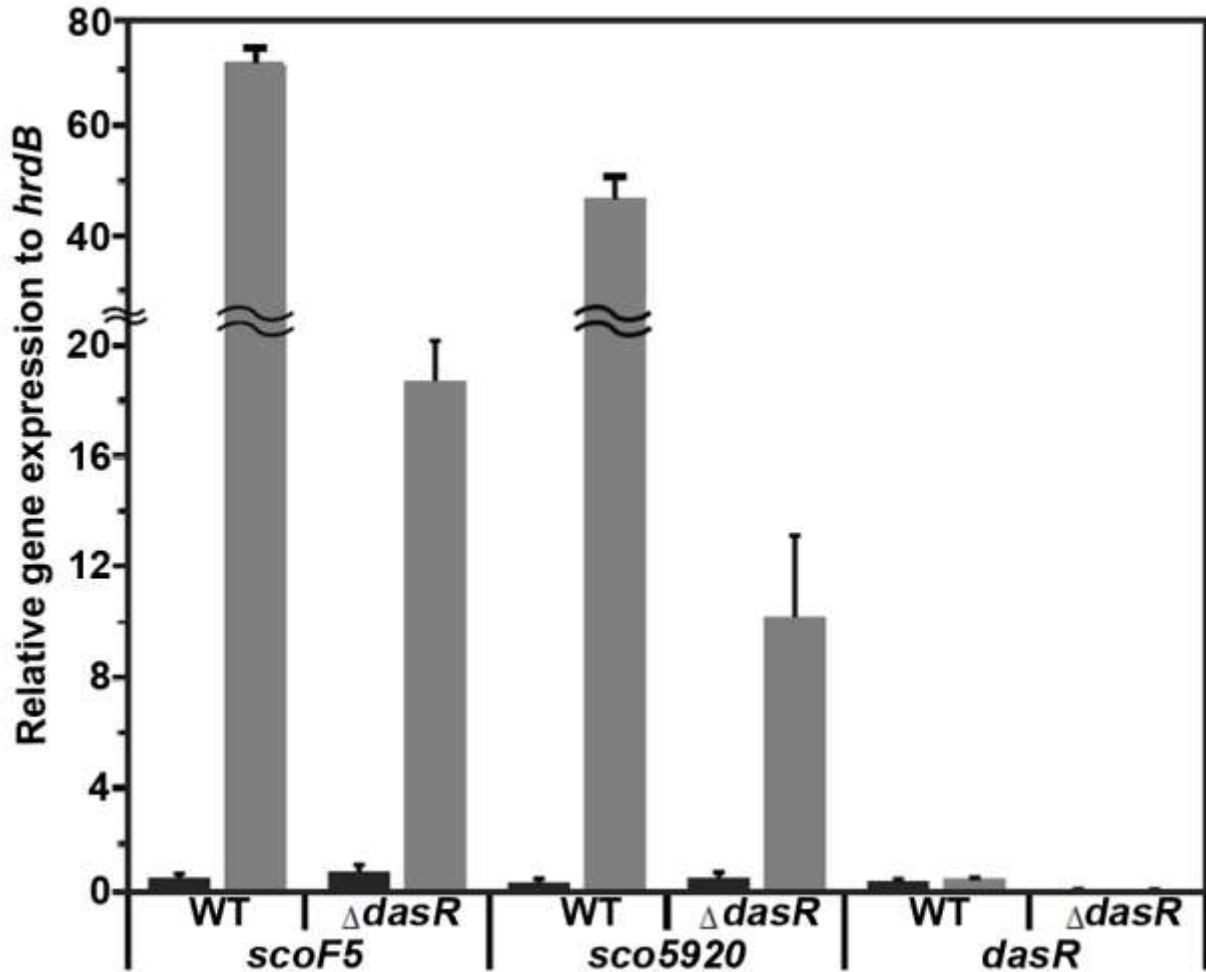


FIGURE S5. Effects of temperature downshifts on gene expression of a cold shock protein (*scoF5*) and a DEAD box RNA helicase (SCO5920). The transcriptional responses to a temperature downshift from 30°C (■) to 10°C (■) of the wild type and YU1 mutant of *S. coelicolor* A3(2) grown in a minimal medium were investigated using qRT-PCR. A temperature downshift triggers induction of the cold shock protein (*scoF5*) and the DEAD box RNA helicase (SCO5920) in the wild type and YU1 mutant, but to different levels. The induction of these proteins was reduced in the YU1 mutant. ≡ shows genes that were highly expressed on a different scale compared with the other genes. The transcriptional analysis of genes involved in the cold-stress response of *S. coelicolor* A3(2) was performed on NMP liquid medium according to a previously reported method (Kormanec & Sevcikova, (2000) *Mol Gen Genet* **264**:251-256).