

Supporting Information for

Title. Allantoin catabolism influences the production of antibiotics in *Streptomyces coelicolor*

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Table S1. Proteomic analysis of *S. coelicolor* M145 grown in MM-GA compared to MM-GC (Student's t test; P < 0.1).

Analysis at 24 h			
Allantoin metabolism	Putative function	r^a	Pvalue
GlxR, SCO6205	putative tartronate semialdehyde reductase	10.44	0.01
Hyi, SCO6206	putative hydroxypyruvate isomerase	9.35	0.08
Glc, SCO6201	putative glyoxylate carboligase	8.88	0.04
SCO6204	putative catalase	6.32	0.03
Alc, SCO6248	allantoicase	6.23	0.00
AceB1, SCO6243	malate synthase	3.99	0.60*
Ugl, 2850	ureidoglycolate lyase	2.19	0.29
Purine metabolism			
SCO5805,	ribonucleotide reductase	5.16	0.02
CysD, SCO6098,	sulfate adenyltransferase subunit 2	0.13	0.09
Nitrogen metabolism			
GlnA, SCO2198	glutamine synthetase I	1.57	0.07
GluB, SCO5776	glutamate periplasmic binding protein	0.25	0.04
GlnII, SCO2210	glutamine synthetase	0.28	0.09
GdhA, SCO4683	NADP-specific glutamate dehydrogenase	1.20	0.63
GlnB, SCO5584	nitrogen regulatory protein pII	0.72	0.57
GluA, SCO5777	glutamate uptake system ATP-binding protein	0.22	0.21
Amino acid metabolism			
IlvC, SCO5514	acetolactate synthase small subunit	3.58	0.03
SerA, SCO5515	probable D-3-phosphoglycerate dehydrogenase	1.16	0.49
SCO4366	putative aminotransferase	2.56	0.05
Carbon metabolism			
Fba, SCO3649	putative fructose 1,6-bisphosphate aldolase	2.19	0.05
Mdh, SCO4827	malate dehydrogenase	1.64	0.06
SCO5281	putative 2-oxoglutarate dehydrogenase	0.49	0.07
Ppc, SCO3127	phosphoenolpyruvate carboxylase	0.48	0.08
Gap, SCO1947	glyceraldehyde-3-phosphate dehydrogenase	0.44	0.06
SucD, SCO4809	succinyl CoA synthetase alpha chain	0.43	0.06
CitA, SCO2736	citrate synthase	0.40	0.02
SCO3877	putative 6-phosphogluconate dehydrogenase	0.37	0.04
Idh, SCO7000	isocitrate dehydrogenase	0.27	0.05
SucC, SCO4808	succinyl-CoA synthetase beta chain	0.23	0.00
SCO2180	putative dihydrolipoamide dehydrogenase	0.40	0.03
Eno, SCO3096	enolase	1.38	0.08

Amino sugar and nucleotide sugar metabolism			
SCO0379	catalase	0.32	0.05
Transcription and DNA metabolism			
SCO4677	putative regulatory protein	5.24	0.07
Hup, SCO2950	DNA binding protein Hu (hs1)	9.45	0.07
RpoB, SCO4654	DNA-directed RNA polymerase beta chain	1.58	0.07
Protein synthesis			
SCO5624	30S ribosomal protein S2	3.19	0.01
RpsE, SCO4719	30S ribosomal protein S5	3.06	0.07
RpsH, SCO4716	30S ribosomal protein S8	2.92	0.07
RplR, SCO4718	50S ribosomal protein L18	2.47	0.07
RplE, SCO4714	50S ribosomal protein L5	2.41	0.03
FusA, SCO4661	elongation factor G	1.90	0.08
Tuf1, SCO4662	elongation factor TU-1	1.59	0.07
ProS, SCO5699	prolyl tRNA synthetase	0.69	0.05
Lipid metabolism			
FabD, SCO2387	malonyl CoA:acyl carrier protein malonyltransferase	0.30	0.06
AcpP, SCO2389	acyl carrier protein	0.12	0.04
Other proteins			
SCO6431	hypothetical protein	1.47	0.05
SCO6279	putative diaminobutyrate-pyruvate aminotransferase	4.00	0.07
SCO1080	hypothetical protein	4.08	0.07
SCO2008	putative periplasmic branched chain amino acid binding protein	6.14	0.01
SCO6009	periplasmic solute-binding protein	5.81	0.02
SCO1074	putative peptidase	0.20	0.06
SCO1480	conserved hypothetical protein	0.19	0.06
SCO1814	putative enoyl-(acyl-carrier-protein) reductase	0.14	0.09
CypH, SCO7510	peptidyl-prolyl cis-trans isomerase	0.34	0.05
SodN, SCO5254	superoxide dismutase	0.32	0.04
SCO0681	putative ferredoxin/ferredoxin-NADP reductase	0.39	0.08
DnaK, SCO3671	heat shock protein 70 (fragment)	0.46	0.09
SCO6431	hypothetical protein	1.47	0.05
SCO4762	60 kD chaperonin cpn60	1.48	0.08
SCO1355	putative secreted serine protease	1.52	0.05
SCO5178	putative sulfurylase	1.61	0.08
SCO2492	putative membrane protein	2.45	0.03

SCO5477	putative oligopeptide-binding lipoprotein	2.47	0.01
SCO1630	putative integral membrane protein	2.53	0.06
AtpD, SCO5373	ATP synthase beta chain	4.33	0.08
SCO4761	10 kD chaperonin cpn10	0.09	0.06
SCO0498	putative peptide monooxygenase	0.35	0.04
SCO2785	hypothetical protein	0.54	0.07
Analysis at 46 h			
Allantoin metabolism			
GlxR, SCO6205	putative tartronate semialdehyde reductase	35.91	0.01
Hyi, SCO6206	putative hydroxypyruvate isomerase	3.49	0.07
AceB1, SCO6243	malate synthase	1.56	0.10
SCO6204	putative catalase	3.15	0.21
Purine metabolism			
SCO4068	phosphoribosylamine-glycine ligase	0.92	0.06
Nitrogen metabolism			
GluB, SCO5776	glutamate periplasmic binding protein	0.30	0.01
GlnA, SCO2198	glutamine synthetase I	0.22	0.05
GdhA, SCO4683	NADP-specific glutamate dehydrogenase	0.98	0.74
GlnII, SCO2210	glutamine synthetase	0.82	0.60
GlnB, SCO5584	nitrogen regulatory protein pII	0.42	0.26
GluA, SCO5777	glutamate uptake system ATP-binding protein	0.59	0.45
Amino acid metabolism			
GlyA1, SCO4837	serine hydroxymethyltransferase	1.28	0.60
SerA, SCO5515	probable D-3-phosphoglycerate dehydrogenase	2.58	0.07
IlvC, SCO5514	acetolactate synthase small subunit	1.76	0.10
MsdA, SCO2726	methylmalonic acid semialdehyde dehydrogenase	2.69	0.07
CysA, SCD66.01, SCO4164	putative thiosulfate sulfurtransferase (fragment)	6.25	0.00
Carbon metabolism			
SCO5281	putative 2-oxoglutarate dehydrogenase	1.81	0.07
Mdh, SCO4827	malate dehydrogenase	2.68	0.02
CitA, SCO2736	citrate synthase	0.52	0.06
Glk, SCO2126	glucokinase	0.14	0.05
SCO2180	putative dihydrolipoamide dehydrogenase	0.38	0.01
Amino sugar and nucleotide sugar metabolism			
SCO0379	catalase	2.07	0.01

SCO3122	putative UDP-N-acetylglucosamine pyrophosphorylase	6.41	0.07
SCO4881	putative polysaccharide biosynthesis related protein	0.93	0.07
Transcription and DNA metabolism			
DnaN, SCO3878	DNA polymerase III, beta chain	0.27	0.04
SCO4729	DNA-directed RNA polymerase alpha chain	0.32	0.08
Protein synthesis			
RpsC, SCO4708	30S ribosomal protein S3	0.38	0.05
SCO1998	30S ribosomal protein S1	0.37	0.06
SCO3909	putative 50S ribosomal protein L9	4.42	0.08
Lipid metabolism			
SCO1814	putative enoyl-(acyl-carrier-protein) reductase	0.23	0.04
Other proteins			
SCO4761	10 kD chaperonin cpn10	6.79	0.04
SCO2396	hypothetical protein	4.06	0.06
SCO2008	putative periplasmic branched chain amino acid binding protein	0.34	0.05
SCO2011	putative branched chain amino acid transport ATP-binding protein	0.18	0.07
SCO3542	integral membrane protein with kinase activity	6.32	0.07
SCO5389	hypothetical protein	5.94	0.06
SCO4277	hypothetical protein	5.53	0.03
GlgX2, SCO5456	putative glycosyl hydrolase	3.97	0.07
SCO2780	putative secreted protein	3.41	0.07
SCO4880	putative transferase	2.14	0.05
Arc, SCO1648	AAA ATPase	2.12	0.06
SCO5178	putative sulfurylase	2.07	0.07
SCO1089	hypothetical protein	0.73	0.06
SCO4228	putative phosphate transport system regulatory protein (fragment)	0.65	0.07
SCO1849	cobalamin biosynthesis protein	0.59	0.07
SCO1925	conserved hypothetical protein	0.48	0.06
SCO3767	hypothetical protein	0.45	0.06
PstS, SCO4142	phosphate-binding protein precursor	0.36	0.01
SCO5477	putative oligopeptide-binding lipoprotein	0.32	0.02
BldKB, SCO5113	BldKB, putative ABC transport system lipoprotein	0.25	0.01
SCO1647	hypothetical protein	0.23	0.07
SCO1565	putative glycerophosphoryl diester phosphodiesterase (fragment)	0.19	0.07
SodF2, SCO0999	superoxide dismutase	0.16	0.09

SCO5520	delta-1-pyrroline-5-carboxylate dehydrogenase	0.14	0.03
SCO1230	putative secreted tripeptidylaminopeptidase	0.05	0.03
SCO6431	hypothetical protein	0.12	0.08
ActIII, SCO5086	ketoacyl reductase	1.33	0.08
SCO0494	putative iron-siderophore binding lipoprotein	0.44	0.08
SCO5074	hypothetical protein	0.19	0.03

^a: Ratio of protein detected in MM-GA/MM-GC

*: Validated by RT-PCR (Table S2).

Table S2. Transcriptional analysis of *aceB1* (24 h) and *actII-ORF4* (46 h) gene expression in *S. coelicolor* M145 strain grown with (MM-GA) or without (MM-GC) allantoin. The threshold cycle (CT) value of the target gene was normalized to the housekeeping *hrdB* gene. Results are the averages from three independent experiments SD.

r^a			
<i>actII-ORF4</i>		<i>aceB1</i>	
M145 MM-GA	M145 MM-GC	M145 MM-GA	M145 MM-GC
0.014 ± 0.007	1	11.41 ± 2	1

^a: Values represent mean difference between MM-GA and MM-GC.

Table S3. Metabolomic analysis of *S. coelicolor* M145 grown in MM-GC or MM-GA at exponential (24 h) and stationary phase (46 h).

Metabolite	24 h		46 h	
	r^a	P value	r^a	P value
alanine	2.27	0.04	46.10	0.01
glycine	2.66	0.10	16.25	0.10
valine	0.44	0.05	0,0	0.10
urea	83.55	0.01	453.00	0.05
phosphate	1.20	0.08	0.25	0.03
fumaric acid	0.17	0.10	47.52	0.04
aspartate	0.36	0.08	97.91	0.01
citramalic	1.29	0.05	6.70	0.02
glutamine	1.07	0.09	0.36	0.06
glucose oxime	1.29	0.08	1.11	0.01
glucopiranosose	0.45	0.12	7.58	0.01

turanose	0.04	0.03	0.36	0.10
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^a: Values represent mean differences between growth in MM-GA and MM-GC.

Figure S1. Determination of urease activity assayed in cell extracts of *S. coelicolor* M145 and UreB⁻ mutant strain grown in presence of allantoin. Values represent the average of experiments performed in triplicate SD. Different letters denote statistical differences applying an ANOVA test using Sigma Stat 3.1 (P < 0.05).

