

SUPPORTING INFORMATION

Genetic Analysis of *Agrobacterium tumefaciens* Unipolar Polysaccharide Production Reveals Complex Integrated Control of the Motile-to-Sessile Switch

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Table S1. Genes decreased in the $\Delta visR$ mutant as determined by DNA microarray expression profiling ($\log_2 FC < -0.6$, $P < 0.05$)

Gene ID	Gene	Product	$\log_2 FC$	P value
Atu0574	<i>flgE</i>	flagellar hook protein FlgE	-5.86	<0.01
Atu0573	-	transcriptional regulator	-5.12	<0.01
Atu0555	<i>flgB</i>	flagellar basal body rod protein FlgB	-5.10	<0.01
Atu0562	<i>fliN</i>	flagellar motor switch protein FliN	-4.87	<0.01
Atu0525	<i>agrR</i>	LuxR family transcriptional regulator	-4.48	<0.01
Atu0543	<i>flaB</i>	hypothetical protein	-4.44	<0.01
Atu0561	<i>fliM</i>	flagellar motor switch protein	-4.39	<0.01
Atu0575	<i>flgK</i>	flagellar hook-associated protein FlgK	-4.35	<0.01
Atu0551	<i>flgA</i>	flagellar basal body P-ring biosynthesis protein FlgA	-4.12	<0.01
Atu0560	<i>motA</i>	flagellar motor protein MotA	-3.96	<0.01
Atu0552	<i>flgG</i>	flagellar basal body rod protein FlgG	-3.92	<0.01
Atu0520	<i>cheY</i>	chemotaxis receiver protein	-3.83	<0.01
Atu0554	<i>flgC</i>	flagellar basal body rod protein FlgC	-3.73	<0.01
Atu0576	<i>flgL</i>	flagellar hook-associated protein FlgL	-3.55	<0.01
Atu0571	<i>motD</i>	chemotaxis MotD protein	-3.40	<0.01
Atu0515	-	hypothetical protein	-3.39	<0.01
Atu0563	<i>fliG</i>	flagellar motor switch protein G	-3.37	<0.01
Atu0844	-	hypothetical protein	-3.30	0.01
Atu0519	<i>cheB</i>	chemotaxis-specific methyl-esterase	-3.26	<0.01
Atu0569	<i>motB</i>	flagellar motor protein MotB	-3.25	<0.01
Atu0568	-	hypothetical protein	-3.19	<0.01
Atu0557	<i>fliI</i>	flagellum-specific ATP synthase	-3.17	<0.01
Atu0549	-	hypothetical protein	-3.14	0.01
Atu0585	-	hypothetical protein	-3.09	0.01
Atu0558	<i>flgF</i>	flagellar basal body rod protein FlgF	-3.07	<0.01
Atu0545	<i>flaA</i>	flagella associated protein	-3.07	<0.01
Atu0516	<i>cheY</i>	chemotaxis receiver protein	-3.04	<0.01
Atu0892	-	hypothetical protein	-3.01	<0.01
Atu0542	<i>fla</i>	flagellin	-2.84	<0.01
Atu0514	-	methyl-accepting chemotaxis protein	-2.81	<0.01
Atu0566	-	hypothetical protein	-2.78	<0.01
Atu4345	-	hypothetical protein	-2.78	<0.01
Atu0584	-	hypothetical protein	-2.68	<0.01
Atu0517	<i>cheA</i>	chemotaxis protein histidine kinase	-2.68	<0.01
Atu0577	<i>flaF</i>	flagellar biosynthesis regulatory protein	-2.61	<0.01
Gene ID	Gene	Product	$\log_2 FC$	P value

Atu4054	<i>exoL</i>	succinoglycan biosynthesis protein	-2.60	<0.01
Atu0553	<i>fliE</i>	flagellar hook-basal body protein FliE	-2.53	0.01
Atu4736	-	methyl-accepting chemotaxis protein	-2.52	<0.01
Atu3977	<i>ina</i>	ice nucleation-like protein	-2.48	<0.01
Atu0579	<i>flgD</i>	flagellar basal body rod modification protein	-2.46	<0.01
Atu4050	<i>exoN</i>	UTP-glucose-1-phosphate uridylyltransferase	-2.45	0.01
Atu0565	-	hypothetical protein	-2.43	<0.01
Atu0521	<i>cheD</i>	chemoreceptor glutamine deamidase CheD	-2.42	<0.01
Atu0567	<i>flaD</i>	flagellin protein FlaD	-2.40	0.02
Atu4049	<i>exoP</i>	exopolysaccharide polymerization/transport protein	-2.39	<0.01
Atu0522	-	hypothetical protein	-2.39	<0.01
Atu0700	-	two component sensor kinase	-2.35	<0.01
Atu0518	<i>cheR</i>	chemotaxis methyltransferase	-2.33	<0.01
Atu1559	-	hypothetical protein	-2.27	<0.01
Atu1233	-	hypothetical protein	-2.26	0.04
Atu3725	-	methyl-accepting chemotaxis protein	-2.25	<0.01
Atu0556	-	hypothetical protein	-2.20	<0.01
Atu2173	<i>mcpA</i>	methyl-accepting chemotaxis protein	-2.13	<0.01
Atu0653	-	hypothetical protein	-2.12	<0.01
Atu3327	<i>exoY</i>	succinoglycan exopolysaccharide synthesis protein	-2.10	<0.01
Atu0578	<i>flbT</i>	flagellar biosynthesis repressor FlbT	-2.09	<0.01
Atu5089	-	hypothetical protein	-2.09	<0.01
Atu4344	<i>clpB</i>	ATP-dependent Clp protease, ATP-binding subunit	-2.08	<0.01
Atu0373	-	methyl-accepting chemotaxis protein	-2.06	<0.01
Atu4052	<i>exoM</i>	UDP-hexose transferase	-2.00	<0.01
Atu2075	<i>cheW</i>	chemotaxis protein	-1.95	0.01
Atu4347	-	hypothetical protein	-1.95	<0.01
Atu4348	<i>vgrG</i>	VgrG protein	-1.92	<0.01
Atu4349	-	hypothetical protein	-1.91	0.01
Atu4346	-	hypothetical protein	-1.90	0.03
Atu4056	<i>exoH</i>	succinoglycan biosynthesis protein	-1.86	0.04
Atu4341	<i>impC</i>	hypothetical protein	-1.85	<0.01
Atu4343	<i>impA</i>	hypothetical protein	-1.79	0.02
Atu4340	<i>impD</i>	hypothetical protein	-1.78	0.02
Atu0547	<i>fliL</i>	flagellar protein	-1.59	0.02
Atu3642	<i>vgrG</i>	vgrG protein	-1.59	0.01
Atu3564	<i>exsH</i>	endo-1,3-1,4-beta-glycanase	-1.58	<0.01
Atu1601	<i>hemN</i>	coproporphyrinogen III oxidase	-1.57	0.05
Gene ID	Gene	Product	log₂ FC	P value

Atu0550	<i>flgI</i>	flagellar basal body P-ring protein	-1.56	0.01
Atu0646	<i>mcpA</i>	methyl-accepting chemotaxis protein A	-1.50	0.03
Atu4337	<i>impG</i>	hypothetical protein	-1.48	0.04
Atu5091	<i>rcdB</i>	Curdlan synthesis protein	-1.47	<0.01
Atu5442	-	methyl-accepting chemotaxis protein	-1.46	0.03
Atu1724	-	hypothetical protein	-1.43	0.02
Atu3975	<i>glf</i>	UDP-galactopyranose mutase	-1.34	0.05
Atu4329	<i>bexD</i>	polysaccharide export protein	-1.33	0.01
Atu0548	<i>flgH</i>	flagellar basal body L-ring protein	-1.32	0.01
Atu4856	-	nucleotidyltransferase	-1.30	0.01
Atu3974	<i>ina</i>	ice nucleation-like protein	-1.27	0.03
Atu0580	<i>fliQ</i>	flagellar biosynthesis protein FliQ	-1.27	0.03
Atu4338	<i>impF</i>	hypothetical protein	-1.26	0.01
Atu3091	-	hypothetical protein	-1.25	0.05
Atu3640	-	hypothetical protein	-1.24	0.02
Atu3321	-	hypothetical protein	-1.23	0.02
Atu4733	-	hypothetical protein	-1.20	0.01
Atu4331	<i>impM</i>	serine/threonine phosphoprotein phosphatase	-1.15	0.02
Atu0544	-	hypothetical protein	-1.15	0.02
Atu4732	-	fimbrial chaperone	-1.10	0.03
Atu3639	-	hypothetical protein	-1.09	0.03
Atu0572	-	hypothetical protein	-1.08	0.02
Atu0916	<i>ros</i>	transcriptional regulator	-1.07	0.02
Atu4335	<i>impl</i>	putative forkhead-type phosphopeptide-binding protein	-1.02	0.02
Atu2617	<i>cheW</i>	chemotaxis protein	-1.01	0.02
Atu0984	<i>pepN</i>	aminopeptidase N	-0.96	0.04
Atu0523	<i>fliF</i>	flagellar MS-ring protein	-0.93	0.03
Atu4058	<i>exoW</i>	succinoglycan biosynthesis protein	-0.85	0.04

Table S2. Genes increased in the $\Delta visR$ mutant as determined by DNA microarray expression profiling ($\log_2 FC > 0.6$, $P < 0.05$)

Gene ID	Gene	Product	$\log_2 FC$	P value
Atu3318	-	LuxR family transcriptional regulator	2.70	<0.01
Atu3384	-	hypothetical protein	2.10	0.01
Atu4454	-	RpiR family transcriptional regulator	1.86	0.02
Atu1691	-	GGDEF family protein (DGC2)	1.67	0.04
Atu3419	-	hippurate hydrolase	1.64	0.02
Atu3426	-	putative sulfonate monooxygenase	1.55	0.03
Atu2707	<i>rzcA</i>	rhizobiocin	1.54	0.02
Atu0204	-	hypothetical protein	1.45	0.02
Atu3094	<i>mcpA</i>	methyl-accepting chemotaxis protein	1.23	0.02
Atu3315	<i>galE</i>	UDP-glucose 4-epimerase	1.14	0.03
Atu0203	-	hypothetical protein	1.13	0.02
Atu3842	-	hypothetical protein	1.01	0.04
Atu0681	-	hypothetical protein	0.99	0.03
Atu3644	-	hypothetical protein	0.95	0.04
Atu2179	-	GGDEF family protein (DgcC)	0.91	0.60*
Atu0824	-	hypothetical protein	0.87	0.05

* Although the P value of *dgcC* is larger than 0.05, β gal assay confirmed that *dgcC* was upregulated in the $\Delta visR$ mutant (Fig. 8B).

Table S3
Strains and plasmids

Strain/plasmid	Relevant features	Reference
<i>E. coli</i>		
DH5 α / λ pir	λ pir ; cloning strain	(Chiang & Rubin, 2002)
TOP10 F'	Cloning strain	Invitrogen
S17-1/ λ pir	λ pir ; Tra ⁺ , cloning host	(Kalogeraki & Winans, 1997)
S17-1/ λ pir (pFD1)	<i>Himar1</i> conjugal donor	(Lampe <i>et al.</i> , 1999)
<i>A. tumefaciens</i>		
C58	Nopaline type strain; pTiC58; pAtC58	(Watson <i>et al.</i> , 1975)
JX100	Δ crdS (Δ Atu3055-3057)	(Xu <i>et al.</i> , 2012)
JX101	Δ chvAB (Δ Atu2728-2730)	(Xu <i>et al.</i> , 2012)
JX102	Δ cel (Δ Atu3302-8187)	(Xu <i>et al.</i> , 2012)
PMM26	Δ upp (Δ Atu1235-1240)	(Xu <i>et al.</i> , 2012)
MLL2 A	Δ exoA (Δ Atu4053)	(Tomlinson <i>et al.</i> , 2010)
JX103	Δ crdS Δ exoA	(Xu <i>et al.</i> , 2012)
JX104	Δ chvAB Δ upp	This study
JX105	Δ cel Δ upp	This study
JX106	Δ crdS Δ exoA Δ upp	This study
JX107	Δ crdS Δ upp	This study
JX108	Δ crdS Δ cel Δ exoA	(Xu <i>et al.</i> , 2012)
JX109	Δ crdS Δ cel Δ exoA Δ upp	This study
JX110	Δ crdS Δ cel Δ exoA Δ chvAB (EPS ⁻ UPP ⁺)	(Xu <i>et al.</i> , 2012)
JX111	Δ crdS Δ cel Δ exoA Δ chvAB Δ upp	This study
JX117	Δ visR (Δ Atu0525)	This study
JX119	Δ visN (Δ Atu0524)	This study
JX120	EPS ⁻ UPP ⁺ Δ visR	This study
JX118	EPS ⁻ UPP ⁺ Δ visN	This study
JX134	Δ visR Δ cel	This study
JX131	Δ visR Δ upp	This study
JX135	Δ visR Δ cel Δ upp	This study
JX125	Δ dgcA (Δ Atu1257)	This study
JX187	Δ dgcB (Δ Atu1691)	This study
YW010	Δ dgcC (Δ Atu2179)	This study
JX127	EPS ⁻ UPP ⁺ Δ dgcA	This study
JX128	EPS ⁻ UPP ⁺ Δ visR Δ dgcA	This study
JX126	Δ visR Δ dgcA	This study
JX191	Δ visR Δ dgcB	This study
JX178	Δ visR Δ dgcC	This study
JX779	Δ visR Δ dgcA Δ dgcB	This study
JX188	Δ visR Δ dgcB Δ dgcC	This study
JX192	Δ visR Δ dgcA Δ dgcB Δ dgcC	This study
JX001	EPS ⁻ UPP ⁺ Δ visR <i>uppE</i> (Atu1236)::Mn	This study
JX138	Δ Atu3495	This study
JX153	EPS ⁻ UPP ⁺ Δ Atu3495	This study
Plasmids		
pGEM-T easy	PCR cloning vector; Ap ^R	Promega

pNPTS138 pSRKGm	ColE1 suicide plasmid; <i>sacB</i> ; Km ^R Broad host range <i>P_{lac}</i> expression vector; <i>lacI</i> Q; Gm ^R	gift of M. Alley (Khan <i>et al.</i> , 2008)
pSRKKm	Broad host range <i>P_{lac}</i> expression vector; <i>lacI</i> Q; Km ^R	(Khan <i>et al.</i> , 2008)
pRA301	Broad host range; promoterless <i>lacZ</i> ; Spc ^R	(Akakura & Winans, 2002)
pJX106	pNPTS138 carrying <i>visR</i> SOE deletion fragment; Km ^R	This study
pJX108	pNPTS138 carrying <i>visN</i> SOE deletion fragment; Km ^R	This study
pJX102	pNPTS138 carrying <i>cel</i> SOE deletion fragment; Km ^R	This study
pJX123	pNPTS138 carrying <i>dgcA</i> SOE deletion fragment; Km ^R	This study
pJX802	pNPTS138 carrying <i>dgcB</i> SOE deletion fragment; Km ^R	This study
pYW010	pNPTS138 carrying <i>dgcC</i> SOE deletion fragment; Km ^R	This study
pJX133	pNPTS138 carrying <i>Atu3495</i> SOE deletion fragment; Km ^R	This study
pJX109	pSRKGm carrying <i>P_{lac}-visR</i> ; Gm ^R	This study
pJX124	pSRKGm carrying <i>P_{lac}-visN</i> ; Gm ^R	This study
pJX136	pSRKGm carrying <i>P_{lac}-visNR</i> ; Gm ^R	This study
pJX125	pSRKGm carrying <i>P_{lac}-dgcA</i> ; Gm ^R	This study
pJX520	pSRKGm carrying <i>P_{lac}-dgcB</i> ; Gm ^R	This study
pYW007	pSRKGm carrying <i>P_{lac}-dgcC</i> ; Km ^R	This study
pJW110	pSRKKm carrying <i>P_{lac}-pleD</i> ; Km ^R	This study
pJX522	pSRKGm carrying <i>P_{lac}-dgcA*</i> ; Gm ^R	This study
pJX521	pSRKGm carrying <i>P_{lac}-dgcB*</i> ; Gm ^R	This study
pJX611	pSRKGm carrying <i>P_{lac}-dgcC*</i> ; Gm ^R	This study
pJX612	pSRKGm carrying <i>P_{lac}-pleD*</i> ; Gm ^R	This study
pJW168	pSRKGm carrying <i>P_{lac}-Atu3495</i> ; Gm ^R	This study
pJX613	pRA301 carrying <i>PdgcA-lacZ</i> ; Spc ^R	This study
pJX162	pRA301 carrying <i>PdgcB-lacZ</i> ; Spc ^R	This study
pJX163	pRA301 carrying <i>PdgcC-lacZ</i> ; Spc ^R	This study
pJX158	pRA301 carrying <i>PAtu3318-lacZ</i> ; Spc ^R	This study
pJX160	pRA301 carrying <i>P-flgE (Atu0574)- lacZ</i> ; Spc ^R	This study
pJX159	pRA301 carrying <i>P-rem (Atu0573)- lacZ</i> ; Spc ^R	This study
pJX166	pRA301 carrying <i>P-motA (Atu0560)- lacZ</i> ; Spc ^R	This study

^a All *A. tumefaciens* strains are C58 derivatives

Table S4. Oligonucleotides

Primer	Sequence
<i>visR</i> -P1	(Spe I) ggtacc actagt AAATGGAGAAGTGCCTTTTCGCTGC
<i>visR</i> -P2	<i>aagctt</i> ggtaccgaattcCTGTACCATCCGTCCGACCAT
<i>visR</i> -P3	<i>gaattc</i> ggtaccaagcttTACATCCATTGATGGCGGACG
<i>visR</i> -P4	(Sph I) aagctt gcatgc ATTTACGGCATCTGTCAGCTTGC
<i>visN</i> -P1	(Spe I) ggtacc actagt AAATTCCGCCGGCATCATCAACTC
<i>visN</i> -P2	<i>aagctt</i> ggtaccgaattcCTTACCCATGCCCTCCAGAGGCTT
<i>visN</i> -P3	<i>gaattc</i> ggtaccaagcttGTATAATGGTTCGGACGGATGGTAC
<i>visN</i> -P4	(Sph I) aagctt gcatgc GCCAGCATGTCCACCATGATTTCA
<i>dgcA</i> -P1	(Spe I) actagt TTATCGACATGCCGCGAAACGAAC
<i>dgcA</i> -P2	<i>aagctt</i> ggtaccgaattcTGAGGATGTCACCGTACAGCG
<i>dgcA</i> -P3	<i>gaattc</i> ggtaccaagcttGCTCGTCTTTGCATCCAGCAT
<i>dgcA</i> -P4	(Sph I) gcatgc GCCATGGCGGTATCAACTGGATTT
<i>dgcB</i> -P1	(Spe I) agagct actagt TATTGGCCATTCTGATGGCCTGA
<i>dgcB</i> -P2	<i>aagctt</i> ggtaccgaattcCATCAAGAACTCCAGTGAGTCTG
<i>dgcB</i> -P3	<i>gaattc</i> ggtaccaagcttTGAAAATTAGAGGGCAGCAGCCGT
<i>dgcB</i> -P4	(Pst I) tata ctgcag AGCTTCGAGATGGAT
<i>dgcC</i> -P1	(Spe I) actagt TGCCTTACTACACGACGATG
<i>dgcC</i> -P2	<i>aagctt</i> ggtaccgaattcTAAGAAGCGGAAAAGCCTGC
<i>dgcC</i> -P3	<i>gaattc</i> ggtaccaagcttCAGCATAATATGCCCATACG
<i>dgcC</i> -P4	(Sph I) gcatgc AAGCTCTACGTCACCAATGC
Atu3495-P1	(Spe I) actagt ACGACATCCAGAGATCCTTGCGAA
Atu3495-P2	<i>aagctt</i> ggtaccgaattcTGAAACGGGCCTGTTTCGCGA
Atu3495-P3	<i>gaattc</i> ggtaccaagcttCACCGCCACAAGTTTTCGCAA
Atu3495-P4	(Sph I) gcatgc CAAACCGCAGGGCAAACCGATATT
Com- <i>visR</i> -P1	(Nde I) aagctt catatg GTCGGACGGATGGTACAG
Com- <i>visR</i> -P2	(Spe I) ggtacc actagt CGGGCGTTTGACCGGCTTAA
Com- <i>visN</i> -P1	(Nde I) catatg GGTAAGCCCTTGCCGCGC
Com- <i>visN</i> -P2	(Spe I) actagt GTACCATCCGTCCGACCATTATAC
Com- <i>dgcA</i> -P1	(Nde I) catatg CTGGATGCAAAGACGAGC
Com- <i>dgcA</i> -P2	(Sac I) gagctc CGCTGTACGGTGACATCCTCA
Com- <i>dgcB</i> -P1	(Nde I) ggcagc catatg AACACGGCATCCACG
Com- <i>dgcB</i> -P2	(Spe I) tctaga actagt ACGGCTGCTGCCCTCTAATTT
Com- <i>dgcC</i> -P1	(Nde I) ggcagc catatg CTGGGTGAACTCGACCTTGCA
Com- <i>dgcC</i> -P2	(Spe I) tctaga actagt CCACTGCAGATGCAGCGAAAGA
Com- <i>pleD</i> -P1	(Nde I) ggcagc catATG ACGGCGAGAGTTCTG
Com- <i>pleD</i> -P2	(Spe I) tctaga actagt AACAGTCGTGGTTAATTG
Com3495-P1	(Nde I) cataTG CGAAAACCTTGTGGCGGTGATCATC

Com3495-P2	(Xba I) <i>ggtctaga</i> TCAGACCGAACTCCAAGCCACCAGT
<i>dgcA</i> *-P1	GTCCGGATGGGCGGCGcGGcATTTGCGCTGGTGGCC
<i>dgcA</i> *-P2	GGCCACCAGCGCAAATgCCgCGCCGCCCATCCGGAC
<i>dgcB</i> *-P1	GCCCCAAGCGGCGGCGcGGcATTGCGCCATCATTCTC
<i>dgcB</i> *-P2	GAGAATGATGGCGAATgCCgCGCCGCCGCTTCGGGC
<i>dgcC</i> *-P1	GCGCGTTACGGCGGCGcGGcATTCATCATTTTCCTG
<i>dgcC</i> *-P2	CAGGAAAATGATGAATgCCgCGCCGCCGTAACGCGC
<i>pleD</i> *-P1	TGCCGTTATGGCGGAGcGGcATTGTCGTCGTCATG
<i>pleD</i> *-P2	CATGACGACGACGAATgCCgCTCCGCCATAACGGCA
<i>p-motA</i> -p1	(Kpn I) gagctc <i>ggtacc</i> TCCAGCGACTTTGCAAGCTCG
<i>p-motA</i> -p2	(Sph I) ctgcag <i>gcatgc</i> CATTGATCGACCACGCCGAA
<i>p-rem</i> -p1	(Kpn I) gagctc <i>ggtacc</i> AAATGTTGATCCGCGCCGCA
<i>p-rem</i> -p2	(Sph I) ctgcag <i>gcatgc</i> CATTGTCGTCGTCGTCGCA
<i>p-flgE</i> -p1	(Kpn I) gagctc <i>ggtacc</i> CGTCGATGATGTCGTCGCA
<i>p-flgE</i> -p2	(Sph I) ctgcag <i>gcatgc</i> CATTCAAAAACCCTCGTGGTC
<i>p-dgcA</i> -p1	(Kpn I) gagctc <i>ggtacc</i> TGTCCGTCCTTTGTCAACTCTG
<i>p-dgcA</i> -p2	(Sph I) ctgcag <i>gcatgc</i> CATTGCAAATCCTATCTGCGCC
<i>p-dgcB</i> -p1	(Kpn I) gagctc <i>ggtacc</i> TATAAGGCTGAAGCCCTGACA
<i>p-dgcB</i> -p2	(Sph I) ctgcag <i>gcatgc</i> CATCAAGAACTCCAGTGAGT
<i>p-dgcC</i> -p1	(Kpn I) gagctc <i>ggtacc</i> GTCACCAATGCCGTCAAGCAC
<i>p-dgcC</i> -p2	(Sph I) ctgcag <i>gcatgc</i> CATAATATGCCCATACGG
<i>p-3318</i> -p1	(Kpn I) gagctc <i>ggtacc</i> CGCCAAATATATATGTCGCTTGG
<i>p-3318</i> -p2	(Sph I) ctgcag <i>gcatgc</i> CATTGACGCCTCCCTAAAGCG
MarTDL2	GACACGGGCCTCGANGNNCNTNGG
MarRSeq	CGGGTATCGCTCTTGAAGGGA
MarLSeq	GGGAATCATTTGAAGGTTGGT
MarTDR1	CAACCGTGCGGGGNTNCNNGNCNG

Upper case sequence anneals to target sequences; lower case are engineered into oligonucleotides but do not anneal with target sequences in genome

Italicized sequences are complementary among paired oligonucleotides

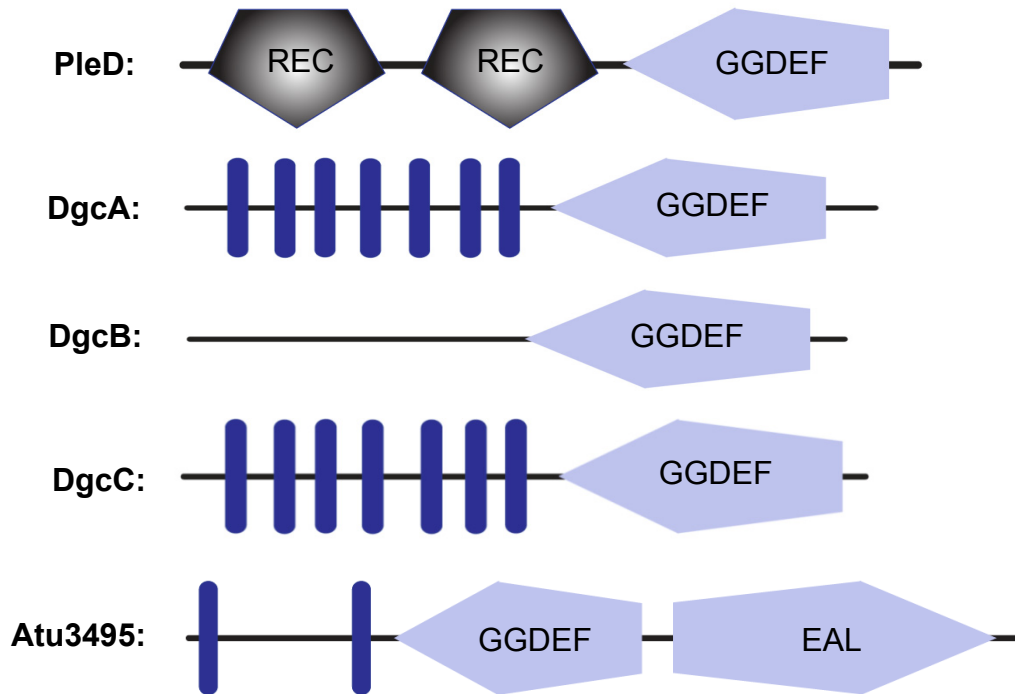
Engineered restriction endonuclease cleavage sites are indicated in parentheses and highlighted in yellow

Supplemental References

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Xu et al. – Supplemental Figure 1

A



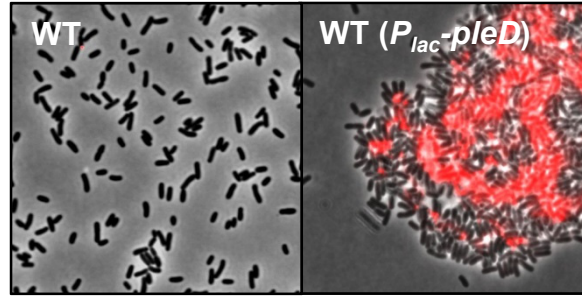
B

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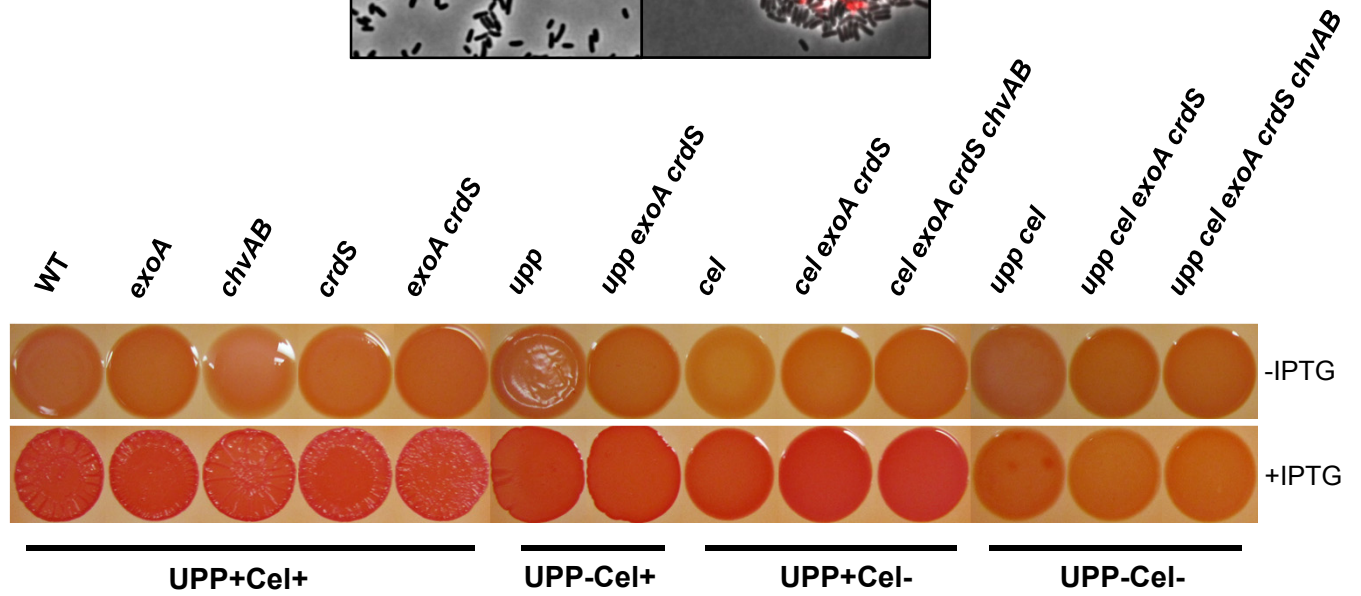
PleD: STVRGADLACRYGGEEFVVVMPDTPIELAASVAERLRAIVEDKPFYVRSIDRELSITASL
DgcA: RVIGERGLFVRMGGEEFALVAEVESPSSTVTLAEAIRSNLRLSKITAR--NEAIKVTTSV
DgcB: ANVRKDG FVARSGGEEFAIILDGNTPEEVMVMCERIRLSLESTPFRNSRSGADYGTVTIS
DgcC: GSIRGGDVLARYGGEEFIIFLPGAGVDKACMIGERIRDAVAINEVDCG--GLRVGVTISI
Atu3495: AIGGDYDMVCRLGGDEFVAVSDDLNEDAARRLSTKLIDQISR TYQLGE---QEVKIGTCI
      .. * **:** .. . . : : : .
    
```

Xu et al. – Supplemental Figure 2

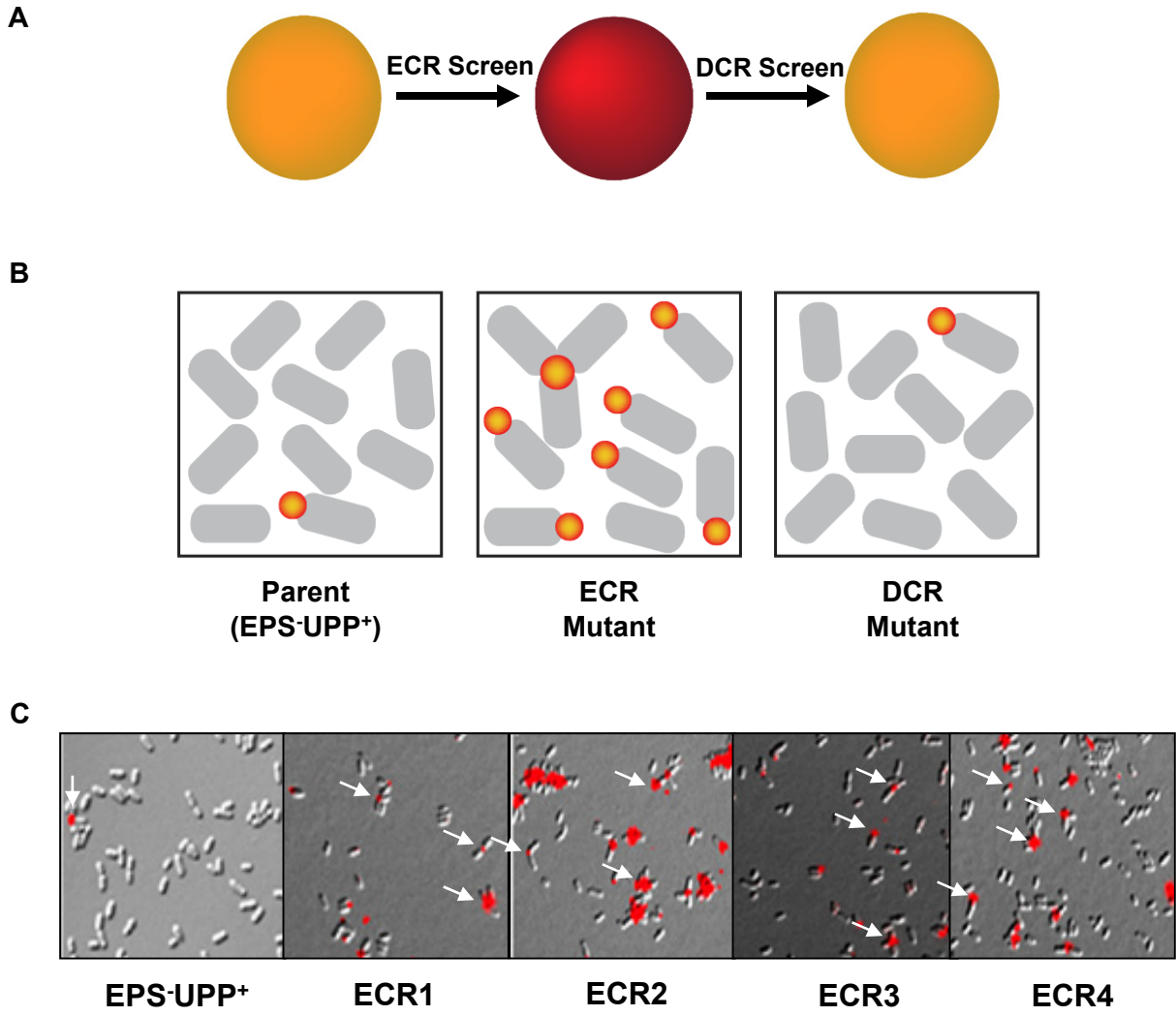
A



B

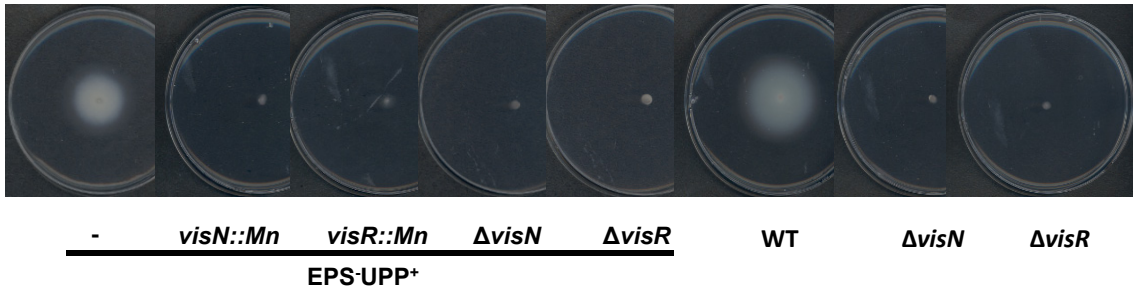


Xu et al. – Supplemental Figure 3

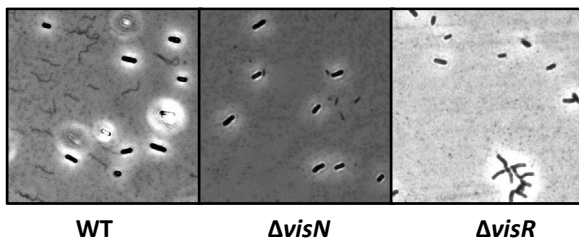


Xu et al. – Supplemental Figure 4

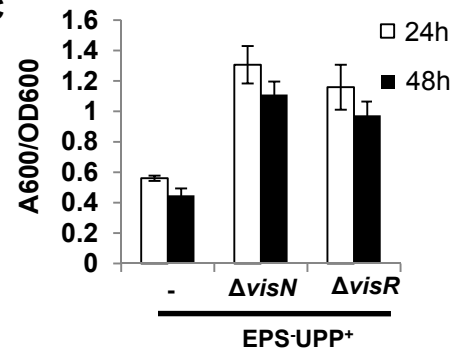
A



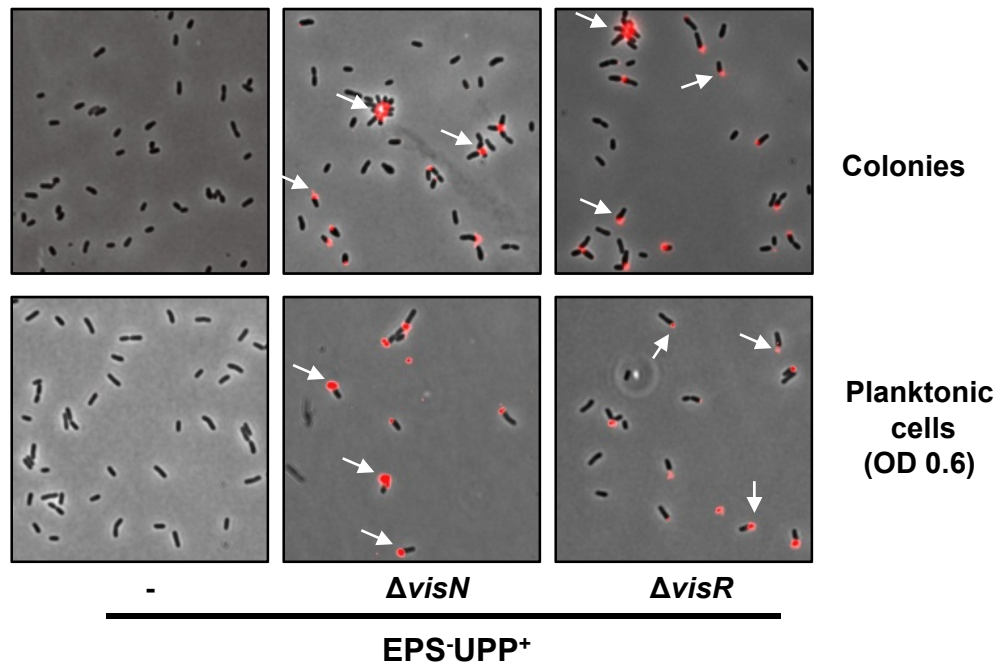
B



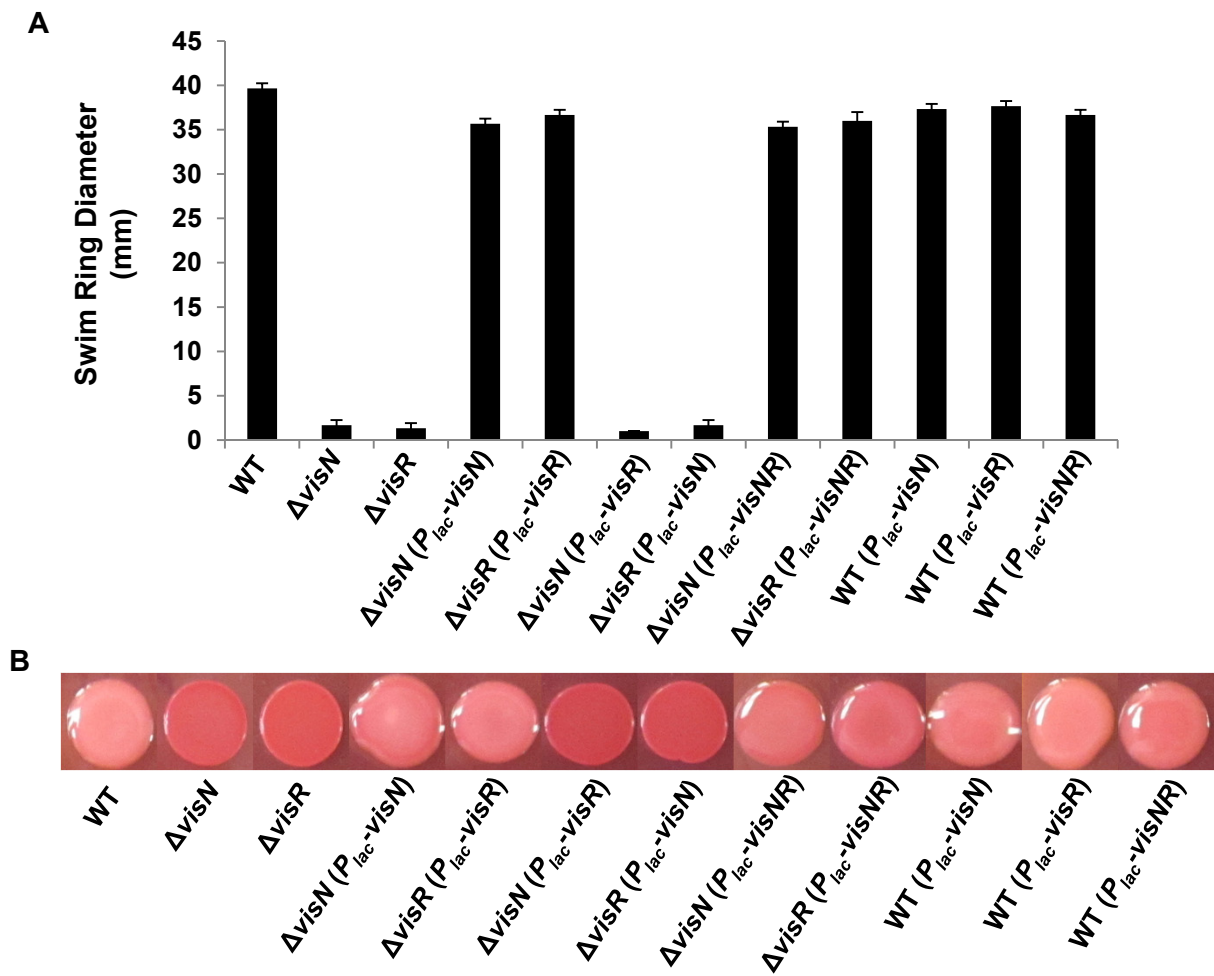
C



D

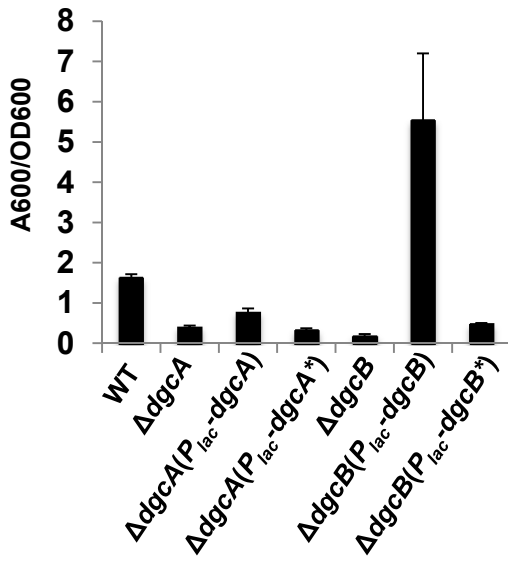


Xu et al. – Supplemental Figure 5

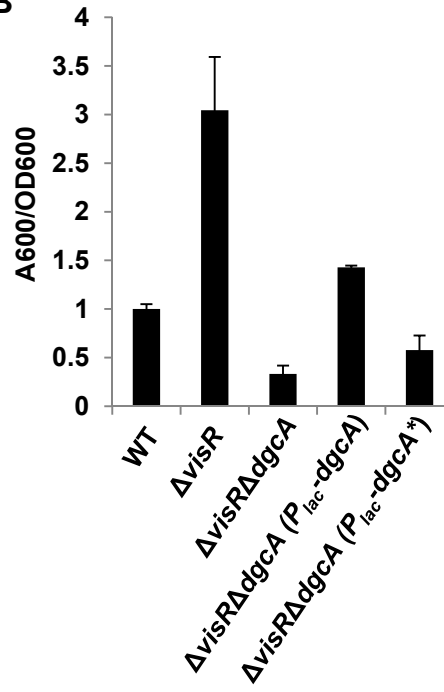


Xu et al. – Supplemental Figure 6

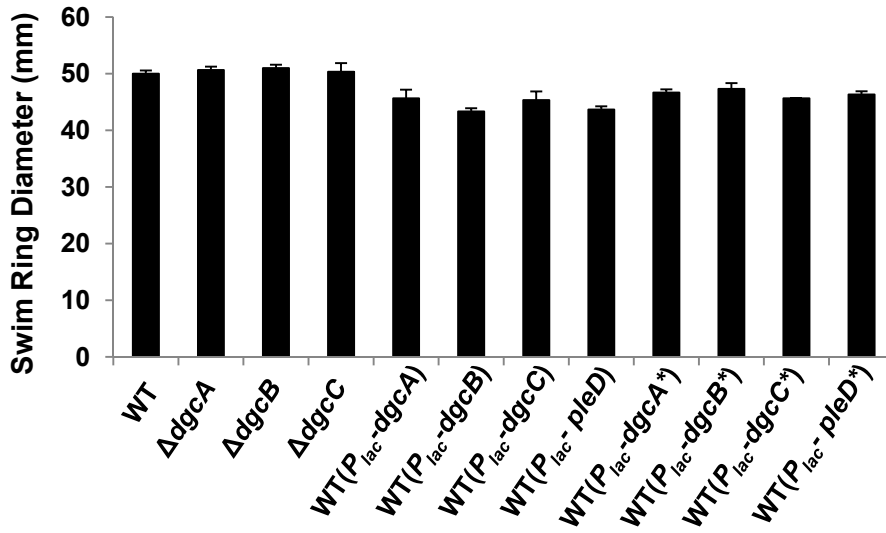
A



B



C



D

