

Supplemental materials

Table S1 Genetic information of PA0847 mutants *

PA ORF	PA0847	PA0847	The PA ORF number in the reference annotation table corresponding to the location of the genome / transposon junction.
Position in ORF	1026(2208)	988(2208)	The position of the hit relative to the ORF = base of insertion / total length of ORF
Frame	-3	2	The frame of the insertion. Either + or (-) (depending on whether the transposon is inserted parallel or antiparallel to the ORF) and the frame position. +2 positions will produce an in-frame insertion, which is required to express a fusion. A + or (-) only, with no number, means that the original round of sequencing did not find the exact junction, and frame is not known.
Genome Position	926189	926227	The position in the reference genome that corresponds to the first base of the chromat after the last transposon base. Only the top hit is given. For two identical matches, the first hit is given.
Transposon	ISphoA/hah	ISphoA/hah	The transposon that is in this mutant.
Transposon Direction	F	R	The absolute direction of the transposon in the genome (forward is oriented parallel to increasing base #).
F Primer Name	F1	F2	Name of forward primer for verification of presence of transposon by PCR
F Primer Seq	AACCGTG GCCTCGA AGTAG	AACCGTG GCCTCGA AGTAG	Sequence of forward primer for verification of presence of transposon by PCR
F Primer Position	925604	925604	Position in genome of forward primer for verification of presence of transposon by PCR
F Primer Tm	60.262	60.262	Tm of forward primer for verification of presence of transposon by PCR
Insert - F	585	623	Number of bases between forward primer and insert location
R Primer Name	R1	R2	Name of reverse primer for verification of presence of transposon by PCR

R Primer Seq	GAACAGG AAAACCT CGACCA	AAAACCT CGACCAC GAGATG	Sequence of reverse primer for verification of presence of transposon by PCR
R Primer Position	926710	926703	Position in genome of reverse primer for verification of presence of transposon by PCR
R Primer Tm	60.088	60.111	Tm of reverse primer for verification of presence of transposon by PCR
R - Insert	521	476	Number of bases between reverse primer and insert location
WT PCR Len	1106	1099	Length of PCR product of forward and reverse primers in wild-type MPAO1

Table S1 Genetic information of PA0847 mutants, which were requested from Manoil Lab. This table is adopted from <http://www.gs.washington.edu/labs/manoil/libraryindex.htm>.

Literature: **Comprehensive transposon mutant library of *Pseudomonas aeruginosa***. Jacobs MA, Alwood A, Thaipisuttikul I, Spencer D, Haugen E, Ernst S, Will O, Kaul R, Raymond C, Levy R, Chun-Rong L, Guenther D, Bovee D, Olson MV, Manoil C. Proc Natl Acad Sci U S A. 2003 Nov 25;100(24):14339-44.

Compounds	Working concentration (mM or %)
Arginine	4.8
Isoleucine	17.6
Leucine	25.6
Valine	17.6
Phenylalanine	8
CaCl ₂	4
MgCl ₂	4
MnCl ₂	2
NaNO ₃	10
CuSO ₄	1
SDS	0.01%
Sucrose	6%
H ₂ O ₂	10
NaCl	4%
Raffinose	0.1
DTT	4
FeCl ₂	2
Spermidine	0.1

Table S2 Working concentration of compound

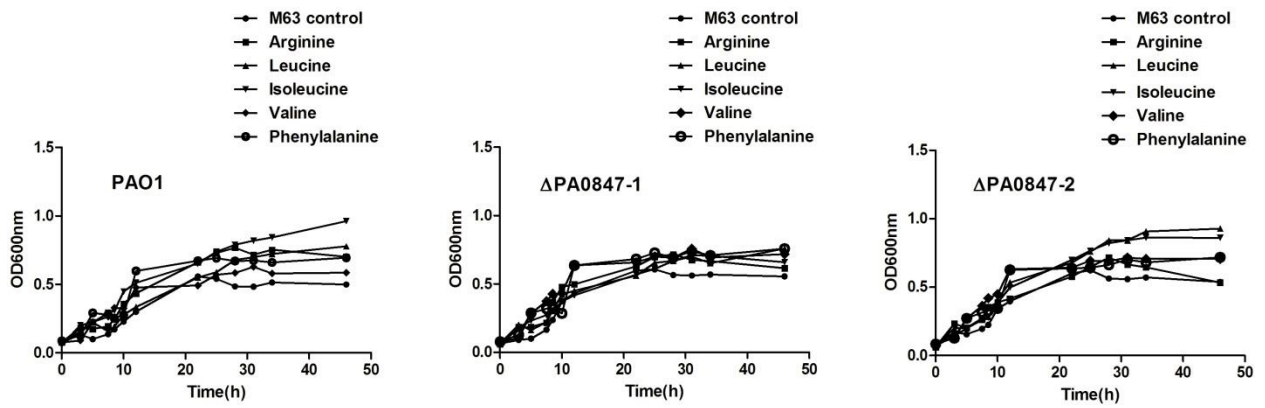


Figure S1 Growth curves of wild type and PA0847 mutants of *Pseudomonas aeruginosa* PAO1 in the presence/absence of different amino acids.

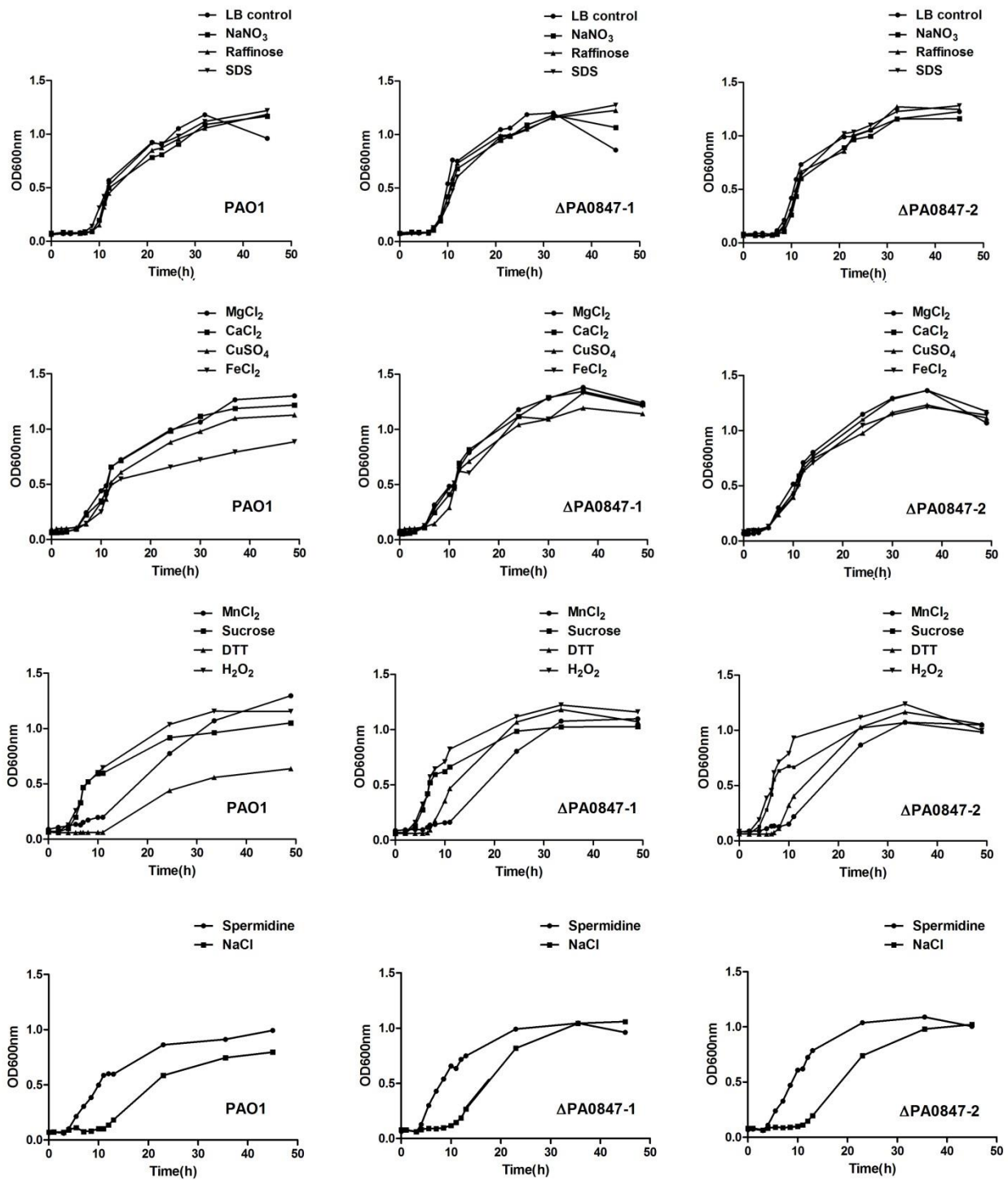


Figure S2 Growth curves of wild type and PA0847 mutants of *Pseudomonas aeruginosa* PAO1 in the presence/absence of different external signals

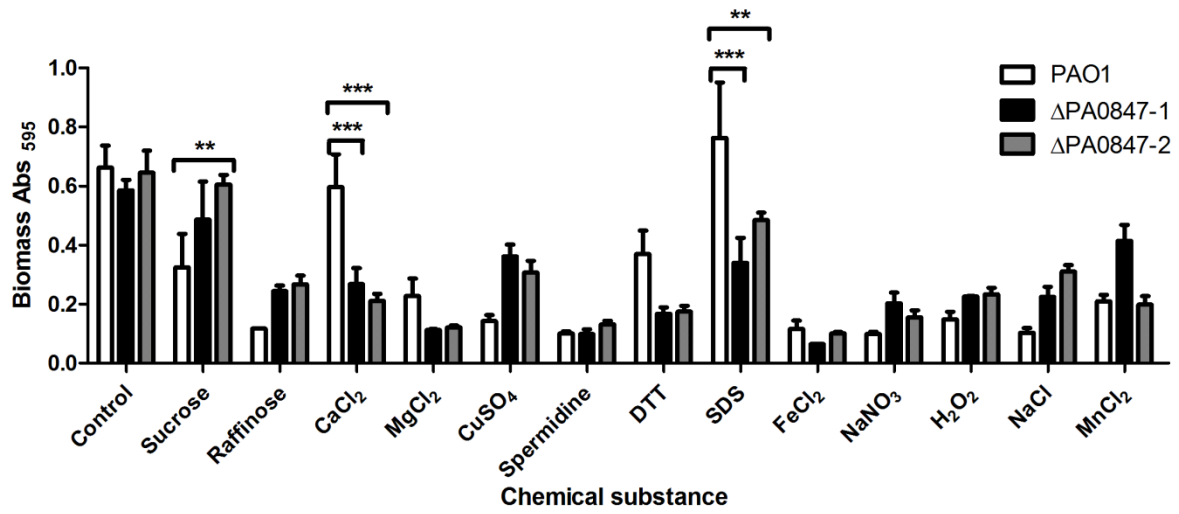


Figure S3 Biofilm formation of *P. aeruginosa* PAO1 and PA0847 mutants in response to signals in the extracellular environment.

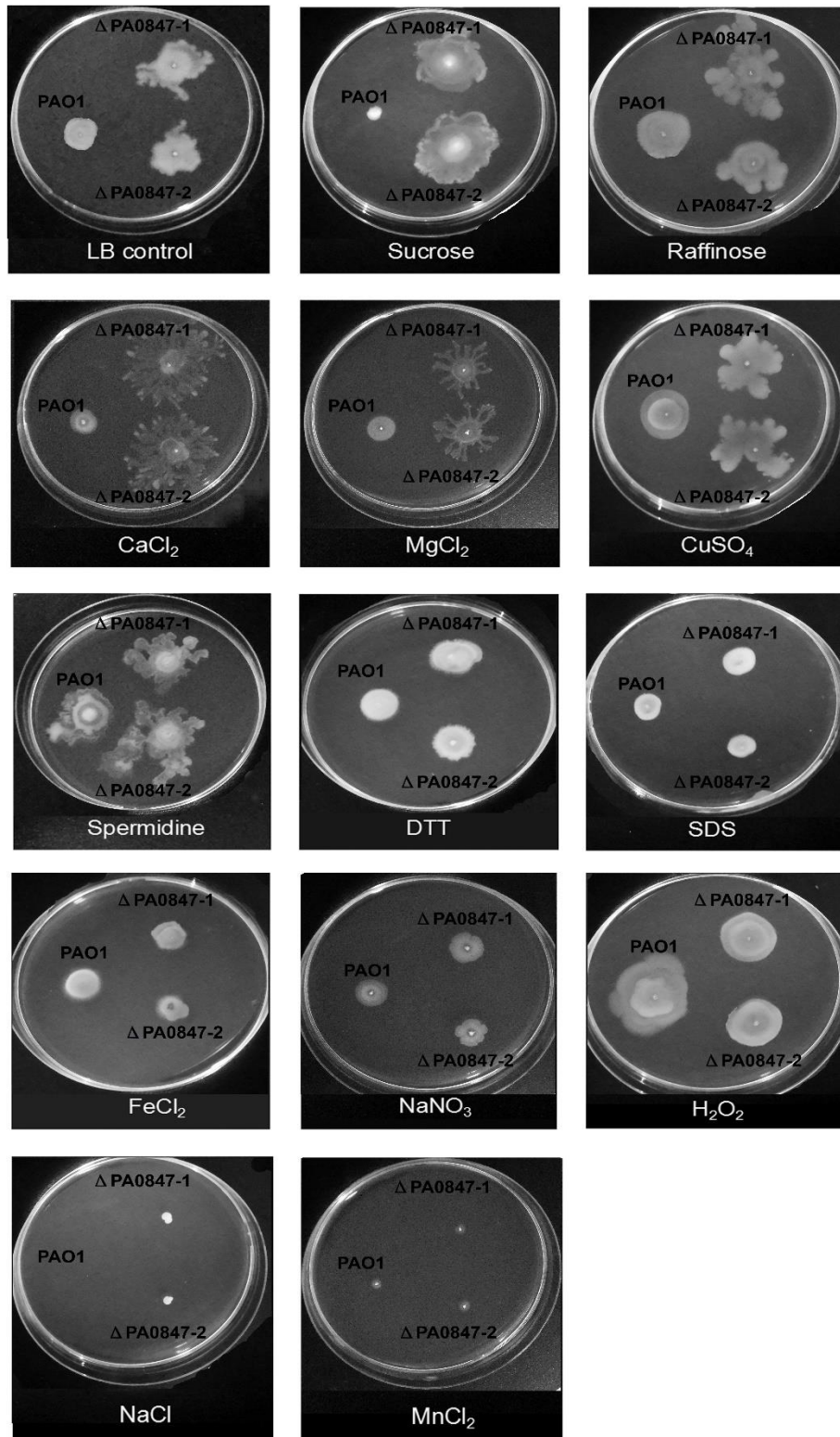


Figure S4 Swimming motility of *P. aeruginosa* PAO1 and PA0847 mutants in response to signals in the extracellular environment.

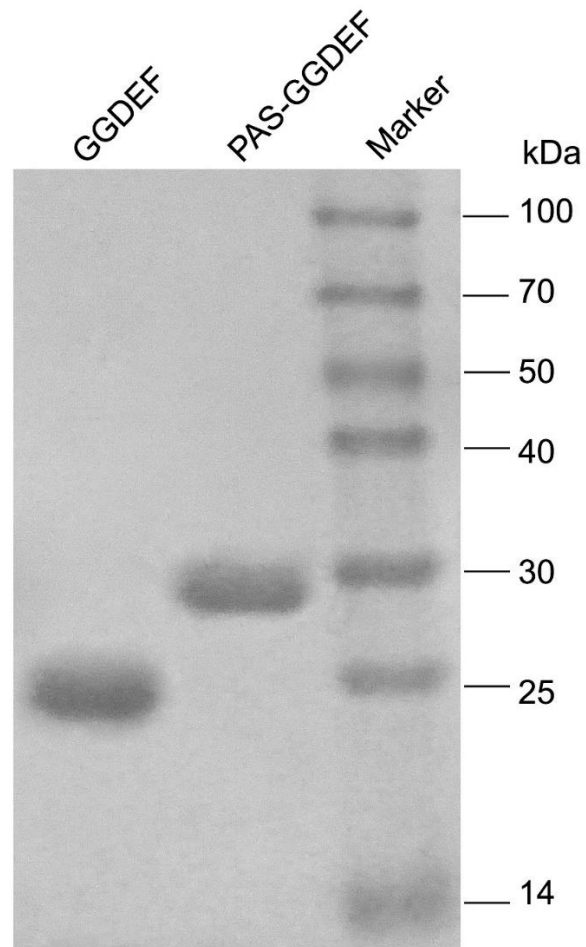


Figure S5 SDS PAGE of purified His-tagged GGDEF and His-tagged PAS-GGDEF domains. Their theoretical MWs are 19.4kDa and 33.7 kDa, respectively.

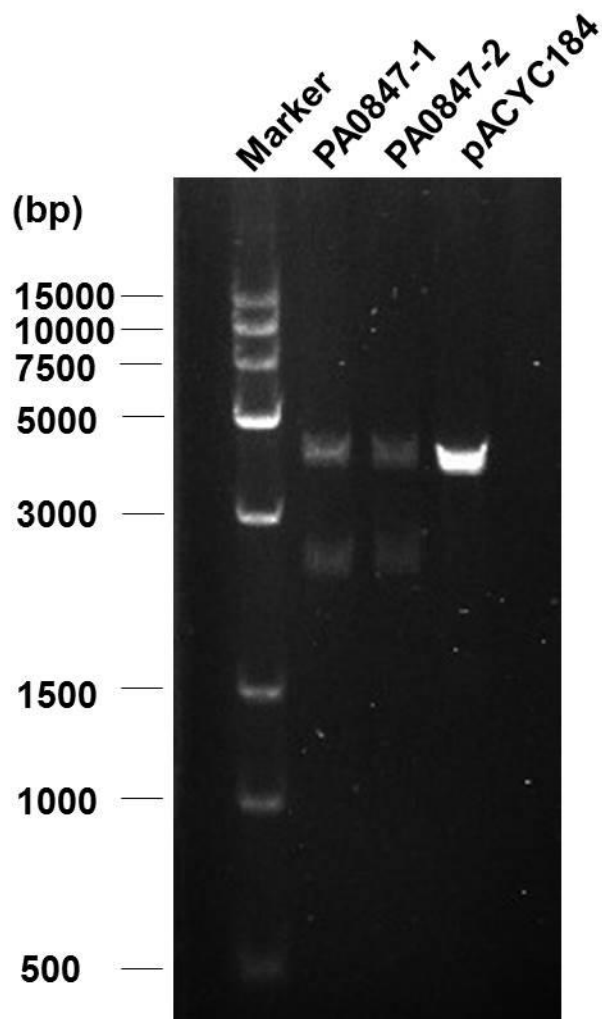


Figure S6 Agarose gel analysis of two constructs harboring full-length PA0847 with its native promoter and RBS sequence. PA0847-1 and -2 represented two constructs with 50 bp difference at the 5' end, digested with BamH I and Hind III. The length of the insert and the pACYC184 vector are 2200 bp and 4244 bp, respectively.