

Figure S1. Promoter region of *OmpA* in *A. baumannii* ATCC 17978 and comparison with other Gram-negative bacteria. Based on this result, the *OmpA* promoters from six gram-negative bacteria were compared through the Multi-Align Program. The six strains of bacteria were *A. baumannii* ATCC 17978(Ab), *E. coli* MG1655(Ec), *Klebsiella aerogenes* KCTC 2190(Kaer), *Klebsiella pneumoniae* HS11286(Kpn), *Shigella flexneri* str. 301(Sfl), and *Salmonella typhimurium* str. 798(Styph). The location of the TATA boxes and TSS seemed to be conserved among the six bacteria. However, the *AbOmpA* promoter, including its TATA box and TSS, exhibited an atypical sequence, while the remaining five bacterial promoter regions showed a strong similarity.

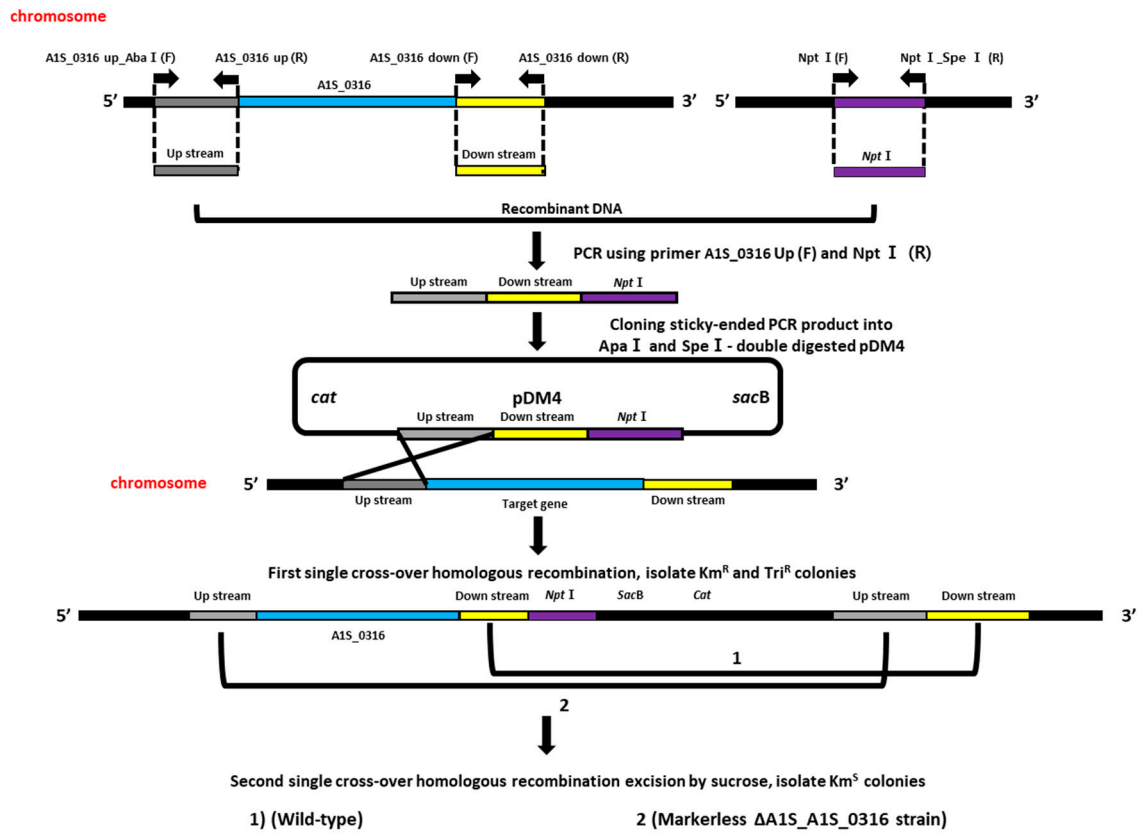


Figure S2. Schematic representation of the construction of the $\Delta A1S_0316$ mutant and its complementary strain of *A. baumannii* ATCC 17978.

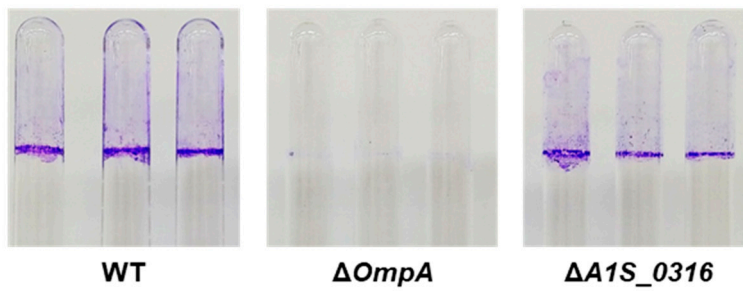
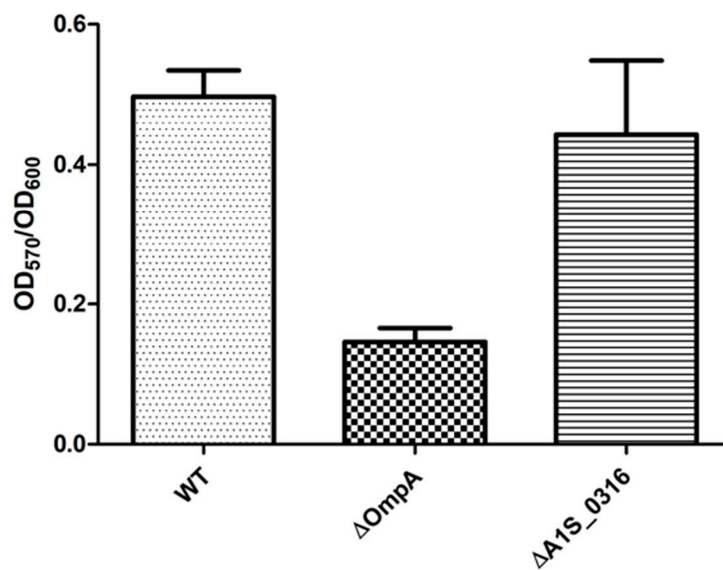
A**B**

Figure S3. Qualitative assay showing biofilm formation on abiotic (glass) surface. Biofilms formed by *A. baumannii* ATCC 17978, $\Delta OmpA$ and $\Delta A1S_0316$ strains. Biofilms were allowed to form for 24 h at 37 °C in LB media without salt. (A) Determination of biofilm synthesis was performed using crystal violet staining. (B) Biofilm values (OD₅₇₀) were normalized by growth levels (OD₆₀₀) to compensate for differences in growth rate. Error bars represent standard deviations.

Table S1. Bacteria Strains, plasmids used in this study

Strain or plasmid	Relevant characteristic(s)	Reference or source
Bacterial strains		
<i>A. baumannii</i>		
ATCC 17978	Prototype strain	[1]
ΔA1S_0316	ATCC 17978 with ΔA1S_0316	This study
ΔOmpA	ATCC 17978 with ΔOmpA	Laboratory collection
<i>E. coli</i>		
SY327 λ <i>pir</i>	<i>supE44 ΔlacU169 (φ80 lacZΔM15) hsdR17 recA1 endA1 gyrA96 thi-1 relA1 λpir</i> (phage lysogen); plasmid replication	Laboratory collection
SM10 λ <i>pir</i>	<i>thi thr leu tonA lacY supE recA::RP4-2-Tc::Mu Km λpir</i> π-requiring plasmids; conjugal donor	[2]
DH5α	<i>fhuA2 lac(del)U169 phoA glnV44 Φ80' lacZ(del)M15 gyrA96 recA1 relA1 endA1 thi-1 hsdR17</i>	Laboratory collection [3]
BL21 star (DE3)	<i>F- ompT hsdS_B (r_B⁻m_B⁻) gal dcm rne131</i> (DE3)	Laboratory collection
Plasmids		
pOH4	pHKD01 with <i>ompA</i> coding region with <i>nptI</i> ; Km ^R	[4]
pB4	derived from pET21a, HisTag-MBP, TEV cleavable	Laboratory collection
pB4::A1S_0316	pB4 with A1S_0316	This study

pDM4	Suicide vector; <i>oriR6K</i> <i>sacBR</i> , Cm ^R	
pDM4::ΔA1S_0316	pDM4 with ΔA1S_0316:: <i>nptI</i> ; , Cm ^R , Km ^R	This study
pAra::AbH-NS	derived from pBAD, AbH-NS <i>oriAb</i> cloned from pWH1266, Tc ^R	Laboratory collection
pWH1266	pWH1277 cloned into pBR322, Amp ^R , Tc ^R	[5]
pWH1266::A1S_0316	pWH1266 with AbCRISPR-array	This study
pSA508	Promoterless, Amp ^R	[6]
pSA508::AbOmpAp	pSA508 with AbOmpAp	This study

Table S2. Identified proteins that bind to *AbOmpAp*

Order	Size (kDa)	Proteins	Locus tag
①	154.9	DNA-directed RNA polymerase subunit beta	A1S_0288
②	61.1	30S ribosomal protein S1	A1S_1572
③	37.2	RNA polymerase alpha subunit	A1S_3056
	30.7	protein chain elongation factor EF-Tu	A1S_0279
④	29.3	Putative transcriptional regulator	A1S_0316
	24.9	minC activating cell division inhibitor a membrane ATPase	A1S_0880
	19.0	putative two-component response regulator (citB)	A1S_3304
	14.2	putative universal stress protein A (UspA)	A1S_2692
⑤	12.5	putative DNA binding protein	A1S_0268
⑥	9.3	DNA-binding protein HU-beta	A1S_1637

Table S3. Oligonucleotides used in this study.

Primers	Primer sequence (5'→3')
A1S_0316 SpeI UF	GTT GGG CCC TGC TGA GCA AAT TGG AAA AC
A1S_0316 UR	AAT AAG ACA TCA TGT AAA ATA ATA TTT TTG ATA ATT TAA AGT TTT CTT ACA C
A1S_0316 DF	TTA TCA AAA ATA TTA TTT TAC ATG ATG TCT TAT TCG TAT CCT T
A1S_0316 DR	CGA GGC AGA CTT ATA CAC AAT CAT TGC GGT TG
A1S_0316 NF	TGA TTG TGT ATA AGT CTG CCT CGT GAA GAA GGT G
A1S_0316 ApaI NR	GTT GGG CCC GAT CCG TCG ACC TGC AGG
A1S_0316 PstI F	TTC CAA TGC ATT GGC TGC AGG AGC TTA TTC CTC TGA GGG AG
A1S_0316 EcoRI R	GGA ATT CGA AAG AAG AGA CAT CGA GAA AGC
A1S_0316 LIC F	GGG CGG CGG TGG TGG CGG CAT GGC AAT TTC AAG TTT TGG C
A1S_0316 LIC R	GTT CTT CTC CTT TGC GCC CTA GAT CTC GCC CTT TTC TGA A
AbOmpAp EcoRI F	CCG GAA TTC AAC ACA AAA TTA AAT AAG GGT TAT CAG
AbOmpAp PstI R	GGT CTG CAG GAT AAC AAT TGT TGT TCA AGC TCA
AbOmpAp biotin F	CGA GTG TTA TAG TGA GCT CA
AbOmpAp biotin R	CAG CTA ATG GAG CAG CAA CA
AbOmpAp EMSA F	GGC AAG CGA AAC AAA GAA GTT G
AbOmpAp EMSA R	AAT GGA GCA GCA ACA AGC ATA G
16s rRNA-sense	GCA CAA GCG GTG GAG CAT
16s rRNA-antisense	CGA AGG CAC CAA TCC ATC TC
OmpA-sense	TTG CAC TTG CTA CTA TGC TTG TTG
OmpA-antisense	TGG CTG TCT TGG AAA GTG TAA CC

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