

Supplementary Materials

PhoB regulates both environmental and virulence gene expression in *Vibrio cholerae*

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Table 1. Bacterial strains and plasmids used in this study

Strain or plasmid	Relevant Genotype	Reference
<i>E. coli</i> strains		
DH5 α	F- Δ (<i>lacZYA-argF</i>) <i>U169 recA1 endA1 hsdR17 supE44 thi-1 gyrA96 relA1</i>	Laboratory strain
DH5 α λ pir	F- Δ (<i>lacZYA-argF</i>) <i>U169 recA1 endA1 hsdR17 supE44 thi-1 gyrA96 relA1</i> λ :: <i>pir</i>	Laboratory strain
SM10 λ pir	<i>thi recA thr leu tonA lacY supE RP4-2-Tc::Mu</i> λ :: <i>pir</i>	Laboratory strain
<i>V. cholerae</i> strains		
O395	classical biotype, Sm ^r	Taylor et al. (1987)
AC61	O395 <i>lacZ::res-Tet-res</i> , Sm ^r , Tc ^r	Camilli and Mekalanos (1995)
AC744	O395 Δ <i>toxR</i> , Sm ^r	Klose and Mekalanos (1998)
AC1596	AC61 <i>vieAE170A</i> , Sm ^r , Tc ^r	Tischler and Camilli (2004)
AC3475	AC61 Δ <i>pst</i> , Sm ^r , Tc ^r	This work
AC3476	AC61 Δ <i>phoB</i> , Sm ^r , Tc ^r	This work
AC3477	AC61 Δ <i>pst</i> Δ <i>phoB</i> , Sm ^r , Tc ^r	This work
AC3478	AC61 Δ <i>phoB</i> pMMBneo:: <i>phoB</i> , Sm ^r , Tc ^r , Kn ^r	This work
AC3479	AC61 Δ <i>phoB::phoB</i> rev, Sm ^r , Tc ^r	This work
AC3480	AC61 Δ <i>pst</i> Δ <i>phoB::phoB</i> rev, Sm ^r , Tc ^r	This work
AC3492	AC61 <i>pstAR454Q</i> , Sm ^r , Tc ^r	This work
AC3502	AC61 Δ <i>phoR</i> , Sm ^r , Tc ^r	This work
AC3503	AC61 Δ <i>pst</i> Δ <i>phoB</i> pMMBneo:: <i>phoB</i> , Sm ^r , Tc ^r , Kn ^r	This work
AC3509	O395 pMMBneo, Sm ^r , Kn ^r	Osorio et al. (2005)
MKW107	O395 pCTX ^{calc} Φ – Kn, Sm ^r , Kn ^r	Davis et al., (1998)
Plasmids		
pMMBneo	IncQ broad-host-range cloning vector, Kn ^r	Osorio et al., (2005)
pAC3493	pMMBneo:: <i>phoB</i> , Kn ^r	This work
pCVD442	<i>oriR6K mobRP4 sacB</i> , Amp ^r	Donnenberg and Kaper (1991)
pAC3494	pCVD442:: <i>pst</i> , Amp ^r	This work
pAC3495	pCVD442:: <i>phoB</i> , Amp ^r	This work
pAC3496	pCVD442:: <i>pstAR454Q</i> , Amp ^r	This work
pAC3499	pCVD442:: <i>phoB</i> rev, Amp ^r	This work
pAC3521	pCVD442:: <i>phoR</i> , Amp ^r	This work
pAIV71	pGEX:: <i>PhoB</i> ^{CA} , Amp ^r	This work
pAIV86	pET15B:: <i>AphB</i> , Amp ^r	This work
pWEL18	pTXB-1:: <i>AphA</i> , Amp ^r	Kovacikova et al. (2003)

Table 2. Primers used in this study

Primer Name	Primer Sequence*
phoBF1	5' <u>GGTCTAGAG</u> TCGTTTCGACCGCTAT TTC 3'
phoBF2	5' GTCTAGAAGGGCCTAAGAGGGTGTAGAACAAATC 3'
phoBR1	5' CCTCTTAGGCCCTTCTAGACATAATTGATTAACCTTTG 3'
phoBR2	5' <u>GGTCTAGAT</u> CATCACACCATGGGCTTTA 3'
pstF1	5' <u>GGTCTAGAC</u> AGTTGGTCGGTTGGAATCT 3'
pstF2	5' CAGACAGCCAAAGCCGAGTTTGTAAGCGG 3'
pstR1	5' AACTCGGCTTTGGCTGTCTGACCATGCTT 3'
pstR2	5' <u>GGTCTAGACT</u> GCAAGAGTGGGTGAAACA 3'
phoRF1	5' <u>GGTCTAGAC</u> ATGGCGTTGGTGAAAAAC 3'
phoRF2	5' GGTTGAACGTATGAGCCGCATGCTAAGTC 3'
phoRR1	5' TGCGGCTCATAACGTTCAACCATACTGATTTGTTCT 3'
phoRR2	5' <u>GGTCTAGAC</u> CCCGACAGAATATTGGCTTG 3'
phoBF	5' <u>GCCCTAGG</u> TTTAGGATACATTTTTATGTCTAGAAGGATTCTGGTTGTTG 3'
phoBR	5' <u>GGGCATG</u> CTTAGGCTTTGGTTGAAAAACG 3'
D10AR1	5' CATTTACGAATCGGTGCTTCGGCTTCAACAACCAGAATCCTTCTA 3'
D10AF2	5' TAGAAGGATTCTGGTTGTTGAAGCCGAAGCACCGATTCTGTAAGT 3'
D53ER1	5' GCAACATCCATTCAAGCAGGACGAGATCAGGAA 3'
D53EF2	5' CGTCCTGCTTGAATGGATGTTGCCTGGTGGTAG 3'
tcpPF	5' AAAATAATGATGTGAAAAATCAGCTT 3'
tcpPR	5' CACCAAAGGTTATCGGGAAA 3'
tcpPF1	5' GTACCGAGTTCGACCGTTTT 3'
tcpPR2	5' GCGAACATGAGTTTTTGTCTGACA 3'
Mut1R1	5' GAACTCGCCTGCATAATATTTAAATTTGTTAAAAAAAATAAAAT 3'
Mut1F2	5' AATATTATGCAGGCGAGTTCTCATTATCAACTGC 3'
Mut2R1	5' TGATACTGCGCACTCGTGTGCATAATATTTAAATTTGTTAAAAAAAATAAAAT 3'
Mut2F2	5' ATGCACACGAGTGCAGTATCAACTGCAAAATTAGATTGCA 3'
R454QF1	5' GCTTGCCTGTTCAAAGTGGT 3'
R454QF2	5' GATTGATTTTGGCGATCGCCCAAGCAGCCGGTGAAGTGGCTCC 3'
R454QR1	5' GGAGCCACTTCACCGGCTGCTTGGGCGATCGCCAAAATCAATC 3'
R454QR2	5' CATCATCTAACCGCGGCTA 3'
tcpAqF	5' CGAAGTGATCATCGTTCTAGGC 3'
tcpAqR	5' TCTGTGTCAGTGCAACTTGG 3'
toxTqF	5' GCTGTCTTTTCTGAAGTGGT 3'
toxTqR	5' TTCTACTTTTCGAGAAGAACCCTGA 3'
toxRqF	5' GTATTACTGCTCACTAACCAGAGC 3'
toxRqR	5' CCAGTTTGAAAGATCAGGGTGG 3'
tcpPqF	5' CTCTGTGAATATCATCTGCCC 3'
tcpPqR	5' GCCTGAGTTAGACTTTCAGAGC 3'
rpoBqF	5' CTGTCTCAAGCCGGTTACAA 3'
rpoBqR	5' TTTCTACCAGTGCAGAGATGC 3'
F NdeI ntPhoB	5' TCACTGTG <u>CATAT</u> GTCTAGAAGGATTCTGGTTGTTG 3'
R ctPhoB st BamHI	5' GAGCGAGT <u>CGGATC</u> CTTAGGCTTTGGTTGAAAAACGATAC 3'
F PhoB T201C	5' CATCAACCTCATCAAGCACATGAAGCGCGAAGAGATG 3'
R PhoB T201C	5' CATCTCTTCGCGCTTCATGTGCTTGATGAGGTTGATG 3'
F NdeI ntAphB	5' TCACTGTG <u>CATAT</u> GCAACATAATGTGTCAGAAACGATGAAAC 3'
R ctAphB st HindIII	5' GAGCGAGT <u>CAAGCT</u> TTTATTGCAGGTGGTAGCCAATCAC 3'
F 6FAM tcpPH -175	5' GATCGGAATTCCTGTAACGAATATTGCTTTCCG 3'
F 6FAM rcpPH -41	5' CAACTGCAAAATTAGATTGCAAATAA 3'

*Restriction sites are underlined

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