

Table S1: Primers used in this study

Primer	Sequence	Description
prRPE-39	5'-CCTCTCGGTGGGCGGAGCTGGAAAGACCTC-3'	Quikchange for S49A point mutant
prRPE-39rc	5'-GAGGTCTTTCCAGCTCCGCCACCAGAGAGG-3'	Reverse complement for S49A
prRPE-41	5'-GTATACTGTCCCGGGCCCAAGAGAAAGAGTAAAGG-3'	Quikchange for Y74A point mutant
prRPE-41rc	5'-CCTTTACTCTTTCTCTTGGCGCCCCGGGACAGTATAC-3'	Reverse complement Y74A
prRPE-56	5'-GGAAGAGGCAGGGGGCGGAGCCCTACCTTATG-3'	Quikchange for D99A point mutant
prRPE-56rc	5'-CATAAGGTAGGGCTCCGCCCTGCCTCTTCC-3'	Reverse complement D99A
prRPE-57	5'-CCATAAGGTAGGGCTCATTCCCTGCCTTCCCAG-3'	Quikchange for D99N point mutant
prRPE-57rc	5'-CTGGGAAGAGGCAGGGAATGAGCCCTACCTTATGG-3'	Reverse complement D99N
prRPE-60	5'-GGGAAGAGGCAGGGGAAGAGCCCTACCTTATGG-3'	Quikchange for D99E point mutant
prRPE-60rc	5'-CCATAAGGTAGGGCTCTTCCCCTGCCTTCCC-3'	Reverse complement D99E
prRPE-42	5'-GAGGCAGGGACGCGCCCTACCTTATG-3'	Quikchange for E100A point mutant
prRPE-42rc	5'-CATAAGGTAGGGCGCGTCCCCTGCCTC-3'	Reverse complement E100A
prRPE-53	5'-GGGAAGAGGCAGGGGACCAGCCCTACCTTATG-3'	Quikchange for E100Q point mutant
prRPE-53rc	5'-CATAAGGTAGGGCTGGTCCCCTGCCTTCCC-3'	Reverse complement E100Q
prRPE-61	5'-GAGGCAGGGACGACCCTACCTTATGGC-3'	Quikchange for E100D point mutant
prRPE-61rc	5'-GCCATAAGGTAGGGTTCGTCCCCCTGCCTC-3'	Reverse complement E100D
prRPE-51	5'-CTCCCGAGGTTTTTATTCTTAATGACGGATTC AACACAGG-3'	Quikchange for D138N point mutant
prRPE-51rc	5'-CCTGTGTTGAAATCCGTCATTAAGAATAAAAACTCGGGAG-3'	Reverse complement D138N
prRPE-52	5'-CCCAGGTTTTTTATTCTTGACAATGGATTTCAACACAGGAACTCC-3'	Quikchange for D139N point mutant
prRPE-52rc	5'-GGAGTTTTCTGTGTTGAAATCCATTGTCAAGAATAAAAACTCGGG-3'	Reverse complement D139N
prRPE-45	5'-GAATTTATGTCCTTCCCCGCTCACTACGATTACTCGG-3'	Quikchange for D260A point mutant
prRPE-45rc	5'-CCGAGTAATCGTAGTGAGCGGGGAAGGACATAAATTC-3'	Reverse complement D260A
prRPE-46	5'-GAATTTATGTCCTTCCCCGATGCCTACGATTACTCGGATTTCCAC-3'	Quikchange for H261A point mutant
prRPE-46rc	5'-GTGAAATCCGAGTAATCGTAGGCATCGGGGAAGGACATAAATTC-3'	Reverse complement H261A
prT7F	5'-TAATACGACTCACTATAGGG-3'	Construct Sequencing
prT7R	5'-GCTAGTTATTGCTCAGCGG-3'	Construct Sequencing

Table S2: Strains and plasmids used in this study

<i>E. coli</i> Strains	Relevant Genotype	Source
C41(DE3)	F ⁻ <i>ompThsds_B (r_B⁻ m_B⁻)gal dcm</i> (DE3) Δ (<i>srl-recA</i>)306::Tn10	(1)
Plasmids	Relevant Genotype	
pET21(b)	Expression vector, T7lac promoter, AmpR	Novagen
pRPE7	pET21(b) harboring <i>A. aeolicus lpxK</i>	(2)
pRPE53	pET21(b) harboring S49A point mutant of <i>A. aeolicus lpxK</i>	This work
pRPE36	pET21(b) harboring K51A point mutant of <i>A. aeolicus lpxK</i>	(2)
pRPE37	pET21(b) harboring T52A point mutant of <i>A. aeolicus lpxK</i>	(2)
pRPE38	pET21(b) harboring S53A point mutant of <i>A. aeolicus lpxK</i>	(2)
pRPE55	pET21(b) harboring Y74A point mutant of <i>A. aeolicus lpxK</i>	This work
pRPE70	pET21(b) harboring D99A point mutant of <i>A. aeolicus lpxK</i>	This work
pRPE71	pET21(b) harboring D99N point mutant of <i>A. aeolicus lpxK</i>	This work
pRPE74	pET21(b) harboring D99E point mutant of <i>A. aeolicus lpxK</i>	This work
pRPE56	pET21(b) harboring E100A point mutant of <i>A. aeolicus lpxK</i>	This work
pRPE69	pET21(b) harboring E100Q point mutant of <i>A. aeolicus lpxK</i>	This work
pRPE75	pET21(b) harboring E100D point mutant of <i>A. aeolicus lpxK</i>	This work
pRPE39	pET21(b) harboring D138A point mutant of <i>A. aeolicus lpxK</i>	(2)
pRPE67	pET21(b) harboring D138N point mutant of <i>A. aeolicus lpxK</i>	This work
pRPE40	pET21(b) harboring D139A point mutant of <i>A. aeolicus lpxK</i>	(2)
pRPE68	pET21(b) harboring D139N point mutant of <i>A. aeolicus lpxK</i>	This work
pRPE59	pET21(b) harboring D260A point mutant of <i>A. aeolicus lpxK</i>	This work
pRPE60	pET21(b) harboring H261A point mutant of <i>A. aeolicus lpxK</i>	This work

¹Miroux B, Walker JE (1996) *J Mol Biol* 260:289-298

²Emptage RP, et al. (2012) *Proc Natl Acad Sci USA* 109:12956-61