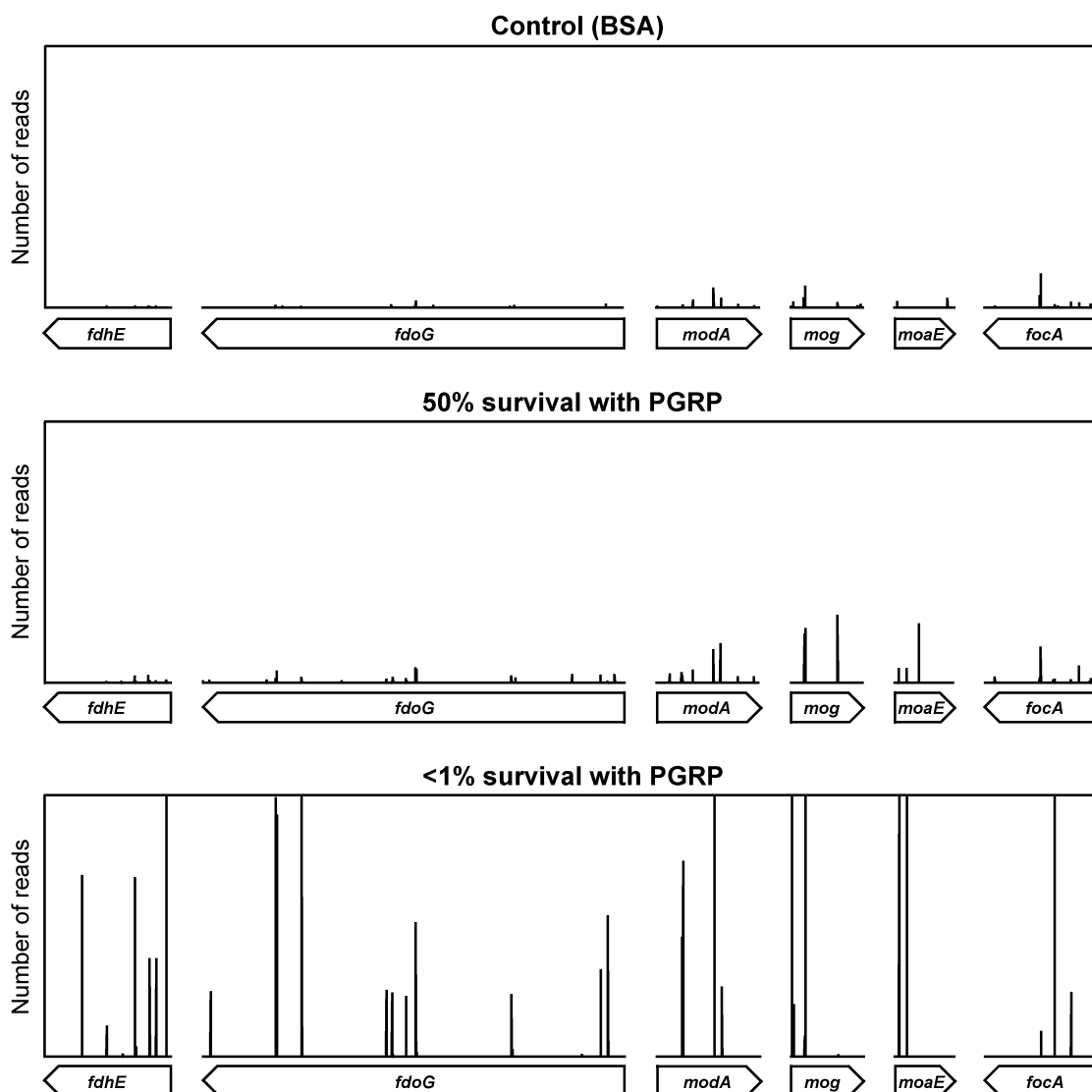


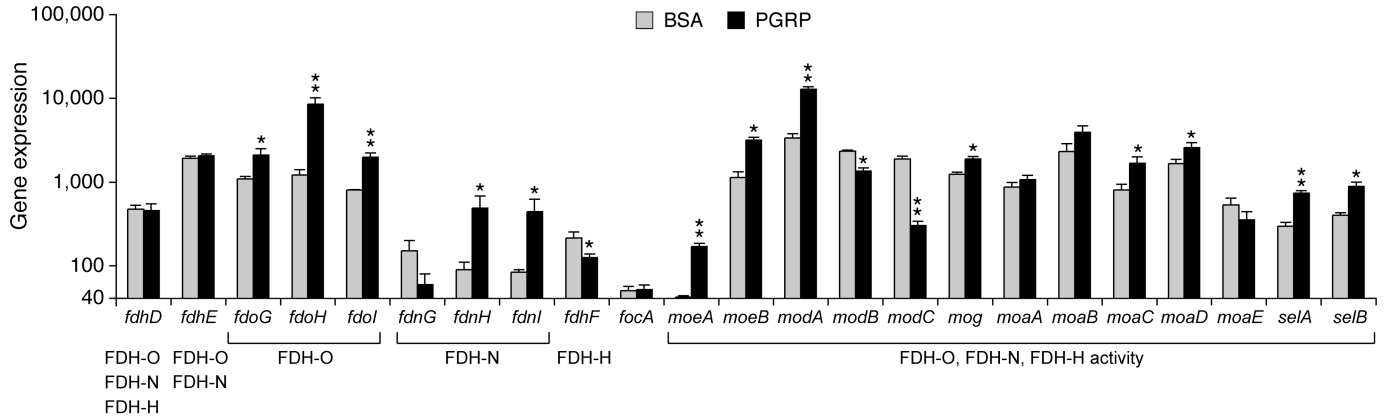
## Supplementary Information

### Formate dehydrogenase, ubiquinone, and cytochrome *bd-I* are required for peptidoglycan recognition protein-induced oxidative stress and killing in *Escherichia coli*

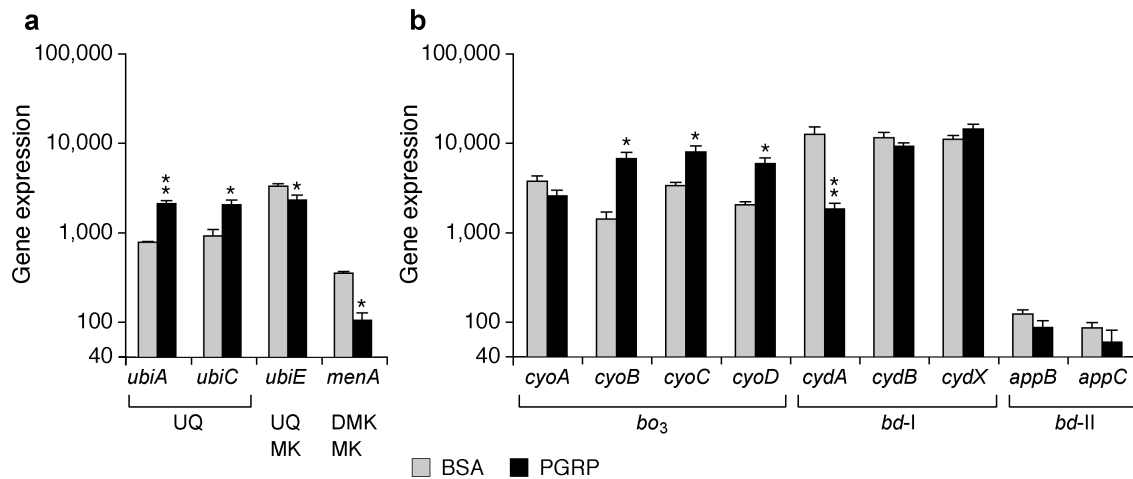
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**Supplementary Fig. S1. Tn insertion sites and the numbers of reads for FDH genes from Fig. 1 with the highest survival index in PGRP-treated cultures.** *E. coli* Tn10 insertion library was treated for 3 hrs with BSA as control or with PGRP at bactericidal (<1% survival) or sub-bactericidal (50% survival) concentration, and the numbers of reads for each Tn-insertion site in each gene was determined by Tn-seq. The height of each vertical line represents geometric mean of the number of sequencing reads at this position from 3 independent experiments (biological replicates). The complete Tn-seq results have been deposited in NCBI SRA with accession number PRJNA549505 (<https://www.ncbi.nlm.nih.gov/sra/PRJNA549505>).



**Supplementary Fig. S2. Expression of FDH and other genes required for the assembly and activity of FDH in PGRP-treated *E. coli*.** *E. coli* MG1655 was treated with BSA or PGRP for 30 min and expression of the indicated genes was determined by whole genome expression arrays (NCBI GEO GSE44211). The results are geometric means  $\pm$  SEM of signal intensities from 3 independent experiments (biological replicates); Y axis starts at the detection limit; \*  $P < 0.05$ , \*\*  $P < 0.001$  PGRP vs BSA.



**Supplementary Fig. S3. Expression of quinone synthesis and cytochrome genes in PGRP-treated *E. coli*.** *E. coli* MG1655 was treated with BSA or PGRP for 30 min and expression of the indicated quinone synthesis genes (a) and cytochrome genes (b) was determined by whole genome expression arrays (NCBI GEO GSE44211). The results are geometric means  $\pm$  SEM of signal intensities from 3 independent experiments (biological replicates); Y axis starts at the detection limit; \*  $P < 0.05$ , \*\*  $P < 0.001$  PGRP vs BSA.

**Supplementary Table S1. Bacterial strains used in this study.**

<b><i>E. coli</i> strain</b>	<b>Relevant genotype</b>	<b>Source or reference</b>
MG1655	Parent K-12 strain for mutants, F-, $\lambda$ -, <i>ilvG</i> -, <i>rfb</i> -50, <i>rphI</i>	ATCC 700926
DRK109 ( $\Delta$ <i>ubiCA</i> )	MG1655:: <i>ubiCA</i> :: <i>kan</i> ; Km <sup>r</sup>	This Study
DRK110 ( $\Delta$ <i>ubiE</i> )	MG1655:: <i>ubiE</i> :: <i>kan</i> ; Km <sup>r</sup>	This Study
DRK111 ( $\Delta$ <i>menA</i> )	MG1655:: <i>menA</i> :: <i>kan</i> ; Km <sup>r</sup>	This Study
DRK112 ( $\Delta$ <i>appB</i> )	MG1655:: <i>appB</i> :: <i>kan</i> ; Km <sup>r</sup>	This Study
DRK113 ( $\Delta$ <i>cyoB</i> )	MG1655:: <i>cyoB</i> :: <i>kan</i> ; Km <sup>r</sup>	This Study
DRK114 ( $\Delta$ <i>cydB</i> )	MG1655:: <i>cydB</i> :: <i>kan</i> ; Km <sup>r</sup>	This Study
DRK117 ( $\Delta$ <i>cyoB</i> $\Delta$ <i>appB</i> )	MG1655:: <i>cyoB</i> :: <i>appB</i> :: <i>kan</i> ; Km <sup>r</sup>	This Study
DRK118 ( $\Delta$ <i>cyoB</i> $\Delta$ <i>cydB</i> )	MG1655:: <i>cyoB</i> :: <i>cydB</i> :: <i>kan</i> ; Km <sup>r</sup>	This Study
DRK121 ( $\Delta$ <i>fdhE</i> )	MG1655:: <i>fdhE</i> :: <i>kan</i> ; Km <sup>r</sup>	This Study
DRK122 ( $\Delta$ <i>fdnG</i> )	MG1655:: <i>fdnG</i> :: <i>kan</i> ; Km <sup>r</sup>	This Study
DRK123 ( $\Delta$ <i>fdhD</i> )	MG1655:: <i>fdhD</i> :: <i>kan</i> ; Km <sup>r</sup>	This Study
DRK124 ( $\Delta$ <i>selB</i> )	MG1655:: <i>selB</i> :: <i>kan</i> ; Km <sup>r</sup>	This Study
DRK125 ( $\Delta$ <i>mog</i> )	MG1655:: <i>mog</i> :: <i>kan</i> ; Km <sup>r</sup>	This Study
DRK127 ( $\Delta$ <i>fdoG</i> )	MG1655:: <i>fdoG</i> :: <i>kan</i> ; Km <sup>r</sup>	This Study
DRK128 ( $\Delta$ <i>fdhF</i> )	MG1655:: <i>fdhF</i> :: <i>kan</i> ; Km <sup>r</sup>	This Study
DRK129 ( $\Delta$ <i>focA</i> )	MG1655:: <i>focA</i> :: <i>kan</i> ; Km <sup>r</sup>	This Study
DRK130 ( $\Delta$ <i>moeA</i> )	MG1655:: <i>moeA</i> :: <i>kan</i> ; Km <sup>r</sup>	This Study
DRK131 ( $\Delta$ <i>modA</i> )	MG1655:: <i>modA</i> :: <i>kan</i> ; Km <sup>r</sup>	This Study
DRK141 ( $\Delta$ <i>moaE</i> )	MG1655:: <i>moaE</i> :: <i>kan</i> ; Km <sup>r</sup>	This Study
DRK147 ( $\Delta$ <i>ubiCA</i> )	BW25113:: <i>ubiCA</i> :: <i>kan</i> ; Km <sup>r</sup>	This Study
DRK148 ( $\Delta$ <i>menA</i> )	BW25113:: <i>menA</i> :: <i>kan</i> ; Km <sup>r</sup>	This Study
BW25113	Parent K-12 strain for Keio collection mutants, <i>rrnB</i> , <i>DElacZ4787</i> , <i>HsdR514</i> , <i>DE(araBAD)567</i> , <i>DE(rhaBAD)568</i> , <i>rph-1</i>	Keio collection, National BioResource Project, National Institute of Genetics, Japan <sup>46</sup>
JW3866 ( $\Delta$ <i>fdhD</i> )	BW25113 F-, $\Delta$ ( <i>araD-araB</i> )567, <i>AlacZ4787</i> :: <i>rrnB</i> -3), $\lambda$ -, <i>rph-1</i> , <i>AfdhD758</i> :: <i>kan</i> , $\Delta$ ( <i>rhaD-rhaB</i> )568, <i>hsdR514</i>	
JW3862 ( $\Delta$ <i>fdhE</i> )	BW25113 F-, $\Delta$ ( <i>araD-araB</i> )567, <i>AlacZ4787</i> :: <i>rrnB</i> -3), $\lambda$ -, <i>rph-1</i> , <i>AfdhE754</i> :: <i>kan</i> , $\Delta$ ( <i>rhaD-rhaB</i> )568, <i>hsdR514</i>	
JW4040 ( $\Delta$ <i>fdhF</i> )	BW25113 F-, $\Delta$ ( <i>araD-araB</i> )567, <i>AlacZ4787</i> :: <i>rrnB</i> -3), $\lambda$ -, <i>rph-1</i> , $\Delta$ ( <i>rhaD-rhaB</i> )568, <i>AfdhF774</i> :: <i>kan</i> , <i>hsdR514</i>	
JW3865 ( $\Delta$ <i>fdoG</i> )	BW25113 F-, $\Delta$ ( <i>araD-araB</i> )567, <i>AlacZ4787</i> :: <i>rrnB</i> -3), $\lambda$ -, <i>rph-1</i> , <i>AfdoG757</i> :: <i>kan</i> , $\Delta$ ( <i>rhaD-rhaB</i> )568, <i>hsdR514</i>	
JW1470 ( $\Delta$ <i>fdnG</i> )	BW25113 F-, $\Delta$ ( <i>araD-araB</i> )567, <i>AlacZ4787</i> :: <i>rrnB</i> -3), $\lambda$ -, <i>AfdnG767</i> :: <i>kan</i> , <i>rph-1</i> , $\Delta$ ( <i>rhaD-rhaB</i> )568, <i>hsdR514</i>	
JW0887 ( $\Delta$ <i>focA</i> )	BW25113 F-, $\Delta$ ( <i>araD-araB</i> )567, <i>AlacZ4787</i> :: <i>rrnB</i> -3), $\lambda$ -, <i>AfocA728</i> :: <i>kan</i> , <i>rph-1</i> , $\Delta$ ( <i>rhaD-rhaB</i> )568, <i>hsdR514</i>	
JW0811 ( $\Delta$ <i>moeA</i> )	BW25113 F-, $\Delta$ ( <i>araD-araB</i> )567, <i>AlacZ4787</i> :: <i>rrnB</i> -3), $\lambda$ -, <i>AmoeA727</i> :: <i>kan</i> , <i>rph-1</i> , $\Delta$ ( <i>rhaD-rhaB</i> )568, <i>hsdR514</i>	
JW0746 ( $\Delta$ <i>modA</i> )	BW25113 F-, $\Delta$ ( <i>araD-araB</i> )567, <i>AlacZ4787</i> :: <i>rrnB</i> -3), <i>AmoA735</i> :: <i>kan</i> , $\lambda$ -, <i>rph-1</i> , $\Delta$ ( <i>rhaD-rhaB</i> )568, <i>hsdR514</i>	
JW0008 ( $\Delta$ <i>mog</i> )	BW25113 F-, <i>Amog</i> -729:: <i>kan</i> , $\Delta$ ( <i>araD-araB</i> )567, <i>AlacZ4787</i> (::rrnB-3), $\lambda$ -, <i>rph-1</i> , $\Delta$ ( <i>rhaD-rhaB</i> )568, <i>hsdR514</i>	
JW0768 ( $\Delta$ <i>moaE</i> )	BW25113 F-, $\Delta$ ( <i>araD-araB</i> )567, <i>AlacZ4787</i> :: <i>rrnB</i> -3), $\lambda$ -, <i>AmoaE757</i> :: <i>kan</i> , <i>rph-1</i> , $\Delta$ ( <i>rhaD-rhaB</i> )568, <i>hsdR514</i>	
JW3563 ( $\Delta$ <i>selB</i> )	BW25113 F-, $\Delta$ ( <i>araD-araB</i> )567, <i>AlacZ4787</i> :: <i>rrnB</i> -3), $\lambda$ -, <i>AselB774</i> :: <i>kan</i> , <i>rph-1</i> , $\Delta$ ( <i>rhaD-rhaB</i> )568, <i>hsdR514</i>	
JW5581 ( $\Delta$ <i>ubiE</i> )	BW25113 F-, $\Delta$ ( <i>araD-araB</i> )567, <i>AlacZ4787</i> :: <i>rrnB</i> -3), $\lambda$ -, <i>rph-1</i> , <i>AubiE778</i> :: <i>kan</i> , $\Delta$ ( <i>rhaD-rhaB</i> )568, <i>hsdR514</i>	
JW0421 ( $\Delta$ <i>cyoB</i> )	BW25113 F-, $\Delta$ ( <i>araD-araB</i> )567, <i>AlacZ4787</i> :: <i>rrnB</i> -3), <i>AcyoB788</i> :: <i>kan</i> , $\lambda$ -, <i>rph-1</i> , $\Delta$ ( <i>rhaD-rhaB</i> )568, <i>hsdR514</i>	
JW0723 ( $\Delta$ <i>cydB</i> )	BW25113 F-, $\Delta$ ( <i>araD-araB</i> )567, <i>AlacZ4787</i> :: <i>rrnB</i> -3), <i>AcydB782</i> :: <i>kan</i> , $\lambda$ -, <i>rph-1</i> , $\Delta$ ( <i>rhaD-rhaB</i> )568, <i>hsdR514</i>	
JW0961 ( $\Delta$ <i>appB</i> )	BW25113 F-, $\Delta$ ( <i>araD-araB</i> )567, <i>AlacZ4787</i> :: <i>rrnB</i> -3), $\lambda$ -, <i>AappB722</i> :: <i>kan</i> , <i>rph-1</i> , $\Delta$ ( <i>rhaD-rhaB</i> )568, <i>hsdR514</i>	

**Supplementary Table S2. Primers used in this study.**

<b>Primers for Tn-seq</b>	
olj363	GTGTGGGCACTCGACATATGACAAG
olj376	GTGACTGGAGTTCAGACGTGTGCTCTTCCGATCTGGGGGGGGGGGGGGGGGG
olj385	AATGATACGGCGACCACCGAGATCTACACTCTTTGGGGGCCAAAATCATTAGGGGATTTCATCAG
BSA bar code	CAAGCAGAAGACGGCATAACGAGATATCACGGTGACTGGAGTTCAGACGTGTGCTCTTCCGATCT
PGRP 50% survival bar code	CAAGCAGAAGACGGCATAACGAGATCGATGTGTGACTGGAGTTCAGACGTGTGCTCTTCCGATCT
PGRP <1% survival bar code	CAAGCAGAAGACGGCATAACGAGATTTAGGCGTGACTGGAGTTCAGACGTGTGCTCTTCCGATCT
olj386	ACACTCTTTGGGGGCCAAAATCATTAGGGGATTTCATCAG
<b>Primers for extensions for gene deletions</b>	
<i>ubiCA</i>	ubiCA (H1) 5'-CTTTTCTGCGATACAATGCCTTTACGTTATGTAACGGAGAGTTCGGCATG-3' ubiCA (H2) 5'-CCGGATGATCATCCGGCTTTTTTACATCATCAGAAATGCCAGTAACTCAT-3'
<i>ubiE</i>	ubiE (H1) 5'-CAATCTGTTACTTCTGGAACAATTTTTGATGAGCAGGCATTGAGATG-3' ubiE (H2) 5'-AAGGTTTTAAAGGCATTTCCGGTCTCCTGTCAGAATTATAACCACGATG-3'
<i>menA</i>	menA (H1) 5'-TCATTGTTTGTGAGGGCTGAAAGGCCCATTTTTATTGGCGCGTATTATG-3' menA (H2) 5'-ATCATCAATTGTAAATTGATATTTGTCAGTTATGCTGCCACTGGCTTAG-3'
<i>appB</i>	appB (H1) 5'-ATGCAGAGTGAACAACCGACGCAGCAACAGGGGTAAAGGAGAAAATCATG-3' appB (H2) 5'-CAAAGTAAATACCACATTGTTTCTGCTCCTTAGTACAACCTCGTTTTTCGTT-3'
<i>cyoB</i>	cyoB (H1) 5'-TGAGCCACGCGGAATCCGCCATTAAGGGGTTGAGGAAGAATAAAGATG-3' cyoB (H2) 5'-GCGCGTGGGCAGTCGCGTGCCTCAAAGTATCAGTTGCCATTTTTTCAGCCC-3'
<i>cydB</i>	cydB (H1) 5'-AGTCTTCCACGACTACTCAGCCGGCAGCTAAGACAGGAGTCGTCAAATG-3' cydB (H2) 5'-TGCGAAATACCACATTTTTAGCTCCTTACTTAGTACAGAGAGTGGGTGTT-3'
<i>fdhE</i>	fdhE (H1) 5'-CCGCGCTGGTACCGTGAGGTCCGCAAGACAACGGAAAAGAAAGCTGAATG-3' fdhE (H2) 5'-GACCTGCGCCACCCGAAAACGTCAGAGATTACTCTCCTTCACCCGGAAA-3'
<i>fdnG</i>	fdnG (H1) 5'-CTGTTAGTTTCTCGCGCAGTAATACCCCTGAAAAAAGAGGAAAGCAATG-3' fdnG (H2) 5'-TGCGTTTTCCATAGCCATCTGTTTCGCCCCCTTACGCCTTCTCGATGTTGAC-3'
<i>fdhD</i>	fdhD (H1) 5'-AGAATACCATAATGTTGGTGTGTGTGTTCTTATCTGGTTAAGAGAAAAGTG-3' fdhD (H2) 5'-TTTAATATATTTTCGCAGGACTTATTTTTAATTAATTGCTCAAACGCTGCGG-3'
<i>selB</i>	selB (H1) 5'-TTACGCTGCCTTGAAGATGAGCAACGGTTTTTGGAGATGTTGTTGAAATG-3' selB (H2) 5'-TTTATTTTTAAACTTAATTAATCATTTTCCTTATTTTTCCGGAAATAATAA-3'
<i>mog</i>	mog (H1) 5'-CAGTGTATCATTCTGTTTAAACGAGACTGTTTAAACGGAAAAATCTTGATG-3' mog (H2) 5'-TTGAGATCCCCCGCTCGGGGGGATTTTTTTATTCGCTAACGTCGCGTCT-3'
<i>fdoG</i>	fdoG (H1) 5'-AAGCGAATGTAAATGGGACGTGACAAATGTCGAAACAAGGAGCAATCCATG-3' fdoG (H2) 5'-TGAGATTGATAAGCCATATGTCGTCCTCGTTACACTTTTCCACATTCAC-3'
<i>fdhF</i>	fdhF (H1) 5'-GTAGTCGAGAGCGCGTATGCGTGATTTGATTAACCTGGAGCGAGACCGATG-3' fdhF (H2) 5'-TCCGAAAGGAGGCTGTAGAAAGGACGGTATTACGCCAGTGCCGCTTCGCG-3'
<i>focA</i>	focA (H1) 5'-ATGCTTTGTTAGTATCTCGTCGCCGACTTAATAAAGAGAGAGTTAGTGTG-3' focA (H2) 5'-TTTTTATTTACTGCGTACTTCGACAACCATTAATGGTGGTTCGTTTTACG-3'
<i>moeA</i>	moeA (H1) 5'-TCTGACATAATAGGCAAATTCGATTTTGCCTCCGCAGGAGTGTTCATG-3' moeA (H2) 5'-CAGCATCTCCTGATCGCTGAGTTCCGCCATTACAGGCCTCCGAACAACGC-3'
<i>modA</i>	modA (H1) 5'-TAGTCGTTATATTGTCGCCTACATAACGTTACATTAAGGGGTTACCAATG-3' modA (H2) 5'-TGCCTGCCATTCTGGATCGGTGAGTATCATTACTTGATTGTAATCCGTA-3'
<i>moaE</i>	moaE (H1) 5'-TGACGGCGACGAAGTAGCTTTCTTCCCGCCGGTAACCGGAGGTTAAGATG-3' moaE (H2) 5'-CAGCGGTGCCTTATCCGGCTAACAAAAAACTACCAGCGTTTTGCCGCCTG-3'
<b>Primers for verification of deletions by PCR and sequencing</b>	
Kanamycin-specific primers	Kan F 5'-GTG CCC TGA ATG AAC TGC AGG ACG A-3' Kan R 5'-GTG CCC AGT CAT AGC CGA ATA GCC T-3'
<b>Gene-specific primers</b>	
<i>ubiCA</i>	ubiCA F 5'-GGG CAT CGG TAA AGC GTA AGG-3' ubiCA R 5'-GCA TCA GGC AAC CCA GAA GAA A-3'
<i>ubiE</i>	ubiE F 5'-CCA TTG GGA GTA GTT AAG CCG-3' ubiE R 5'-ACA GGA AGG TGT TGA GCA GAC-3'

<i>menA</i>	menA F 5'-TGG ATG CGT TGG TGG CAG ATG A-3' menA R 5'-GGC TTA ACA TTC AGT TGC TGC G-3'
<i>appB</i>	appB F 5'-GCG ATA CAG GAC ATC TTA CCG A-3' appB R 5'-TTG CCT GAT GCG ATG CTA ATG C-3'
<i>cyoB</i>	cyoB F 5'-GTG AAA CCA GAC TTG TTT GCC G-3' cyoB R 5'-TTC ACC AGA ACG GCA TAG GTA GC-3'
<i>cydB</i>	cydB F 5'-CTC AAT GGT GCT GAT TTG CGG C-3' cydB R 5'-GGT CCC AAA ACA TAC ATC CTG C-3'
<i>fdhE</i>	fdhE F 5'-GGT GGA AGG ATG GGT TAC CAG CG-3' fdhE R 5'-CCT TAT TGA TGT GGT GGT GGG T-3'
<i>fdnG</i>	fdnG F 5'-CTG TCA GGC AGT GGT GCG TTT TT-3' fdnG R 5'-TAC AGC CGA TAC AGG TGG AAA CG-3'
<i>fdhD</i>	fdhD F 5'-TTA CAT TCG CTT GCT GCG TGT GC-3' fdhD R 5'-CGA GGG ACA TGA TGG CGA CTA TA-3'
<i>selB</i>	selB F 5'-TAG CCA CCT TGA GTC ATT AGC CGC-3' selB R 5'-GAA AGT GTA AAA AAG GGC GGG CAG-3'
<i>mog</i>	mog F 5'-ATC ACT GAG TCC GAG TTC CTG TG-3' mog R 5'-GCA ATC TAT CTG GCG ATG GGT GA-3'
<i>fdoG</i>	fdoG F 5'-GTT ACA AAC CCT TCC TGG ATG GAG-3' fdoG R 5'-TGA GTT TCG CCA CTT CTT CCT GG-3'
<i>fdhF</i>	fdhF F 5'-ACT GAC GAC AGC CTG TTT TTC G-3' fdhF R 5'-TCG TTG GCG TGG GCA AAG AAT GT-3'
<i>focA</i>	focA F 5'-GCC TAT AAG CCA GGC GAG ATA TGA-3' focA R 5'-CTT CAT TCT GCC AGT CAC CTT TGG-3'
<i>moeA</i>	moeA F 5'-TTC GTC CAT TAA GGG TGG TTC GC-3' moeA R 5'-CGA GCA GCG TCA GGT TAC CGA CA-3'
<i>modA</i>	modA F 5'-TTC CCT GAA TGC CCG CTT AGT CG-3' modA R 5'-CAA CGC ACC AGT AAC CAG GCA AA-3'
<i>moaE</i>	moaE F 5'-GCA AAT TAC TGG CTG CCG TCA AC-3' moaE R 5'-GCA GCG TAA ACG TAT GTA CTG AGC-3'