

Supplementary Table S1. Primers used in this study

Primer name	Nucleotide sequence (5' to 3')	Purpose of amplification
<i>Cas1</i> -UF	ACTTGC GTTCAATACCCT	<i>Cas1</i> upstream homologous arm
<i>Cas1</i> -UR	<u>TCCCCCGGGT</u> TCCCGTTCCAAGTAAA	
<i>Cas1</i> -DF	<u>TCCCCCGGGAT</u> GGTTTAGCGGCAACAACACT	<i>Cas1</i> downstream homologous arm
<i>Cas1</i> -DR	TAACGGCGAGCAATCCTT	
<i>Cas2-3</i> -UF	CTGTGGGTTCTTGGGATT	<i>Cas2-3</i> upstream homologous arm
<i>Cas2-3</i> -UR	<u>TCCCCCGGGCT</u> GGGTGATTACAGTTTGC	
<i>Cas2-3</i> -DF	<u>TCCCCCGGGTA</u> AGGCTGCGATTCACTAT	
<i>Cas2-3</i> -DR	TTTTCTGGA ACTGGGATG	<i>Cas2-3</i> downstream homologous arm
<i>Csy1</i> -UF	TACATTTGTTGCGGGAGC	<i>Csy1</i> upstream homologous arm
<i>Csy1</i> -UR	<u>TCCCCCGGGAT</u> TCGGCAGCCAGTTTTC	
<i>Csy1</i> -DF	<u>TCCCCCGGGAT</u> CCCAATCCAGTAGCCT	<i>Csy1</i> downstream homologous arm
<i>Csy1</i> -DR	CACAACACCAACCGCAGA	
<i>Csy2</i> -UF	ACAGCATCAAACATCCCCT	<i>Csy2</i> upstream homologous arm
<i>Csy2</i> -UR	<u>TCCCCCGGGCA</u> ACACCAACCGCAGAAA	

<i>Csy2</i> -DF	<u>TCCCCGGGACTCCTCATCGCTTTGCT</u>	<i>Csy2</i> downstream homologous arm
<i>Csy2</i> -DR	AAGTGGCGTGGCTTTATC	
<i>Csy3</i> -UF	ATGCCTTATCGTTTGGAG	<i>Csy3</i> upstream homologous arm
<i>Csy3</i> -UR	<u>TCCCCGGGCTGAACCCAATGAACAAGTA</u>	
<i>Csy3</i> -DF	<u>TCCCCGGGGGGAAAAGCGTTTAGGAC</u>	<i>Csy3</i> upstream homologous arm
<i>Csy3</i> -DR	TTAGTGGAAGCCAGAGTATGT	
<i>Cas6f</i> -UF	TTTTGATACTTGGGCACG	<i>Cas6f</i> upstream homologous arm
<i>Cas6f</i> -UR	<u>TCCCCGGGAAGGCTTTGTGGAGATTG</u>	
<i>Cas6f</i> -DF	<u>TCCCCGGGAAATCAGGCAATACAAGGC</u>	<i>Cas6f</i> upstream homologous arm
<i>Cas6f</i> -DR	AGTGAGATGGTGGCAAATA	
CRISPR-UF	AGCGTATCAAACGGGCAAAG	CRISPR upstream homologous arm
CRISPR-UR	<u>TCCCCGGGTTAGTGAGATGGTGGCAAA</u>	
CRISPR-DF	<u>TCCCCGGGGCTCCAGTTATTCGTGATG</u>	CRISPR downstream homologous arm
CRISPR-DR	GCTGGCGAACAAAAGACT	
hb-Cas1-F-SacI	<u>CGAGCTCaggaggaattcaccATGGAAATACTAACAGACTTAAAAG</u>	The construction of Complementary strain of <i>cas1</i>
hb-Cas1-R-XbaI	<u>GCTCTAGATCGGCAAAAAGCATCTAAG</u>	

hb-Cas2-3-F-SacI	<u>CGAGCTCaggaggaattcaccATGATGGTTATTTTTGTTTCCC</u>	The construction of Complementary strain of <i>cas2-3</i>
hb-Cas2-3-R-XbaI	<u>GCTCTAGACAAGTTCAGTCATATCCGA</u>	
hb-Csy1-F-SacI	<u>CGAGCTCaggaggaattcaccATGTTTGATCCAGCAATTAC</u>	The construction of Complementary strain of <i>csy1</i>
hb-Csy1-R-XbaI	<u>GCTCTAGACCAATGGTGAAAGGGCTG</u>	
hb-Csy2-F-SacI	<u>CGAGCTCaggaggaattcaccATGAGAAGTAACCTGATTTTATTGC</u>	The construction of Complementary strain of <i>csy2</i>
hb-Csy2-R-XbaI	<u>GCTCTAGACATCCGATGGTACCAATT</u>	
<i>hb-Csy3-F-SacI</i>	<u>CGAGCTCaggaggaattcaccATGACTAAAAAAGAACACATAGC</u>	The construction of Complementary strain of <i>csy3</i>
<i>hb-Csy3-R-XbaI</i>	<u>GCTCTAGACATTAACACTGGGGTTGTA</u>	
hb-Cas6f-F-SacI	<u>CGAGCTCaggaggaattcaccATGGATCATTATTTAGATATATCTATTCTT</u>	The construction of Complementary strain of <i>cas6f</i>
hb-Cas6f-R-XbaI	<u>GCTCTAGATAACCTATCGAACTCAAGGA</u>	
hb-CRISPR-F-SacI	<u>CGAGCTCaggaggaattcaccCTTAAAATCAATTGGTTATAAATTAGCT</u>	The construction of Complementary strain of CRISPR
hb-CRISPR-R-XbaI	<u>GCTCTAGACAATTACACTCATTTCATGG</u>	
207-F	TGATCCGGGCTTATCGACTG	Test the correct construction of Complementary strain
207-R	ACCGCTTCTGCGTTCTGATTT	
<i>Cas1-qF</i>	CTTGGGATTCCCTCACGGCTT	<i>cas1</i> gene expression detection for qPCR
<i>Cas1-qR</i>	CCTTGGCGCAGATAAAATGCC	

<i>Cas2-3-qF</i>	TGCTTCACCATGCTCGCTTA	<i>cas2-3</i> gene expression detection for qPCR
<i>Cas2-3-qR</i>	AAGCCTGCTTTGGCGATTIG	
<i>Csy1-qF</i>	ATCCCAATCCAGTAGCCTTCCAG	<i>csy1</i> gene expression detection for qPCR
<i>Csy1-qR</i>	CGTTCAGCCTCACCCAACATAGT	
<i>Csy2-qF</i>	CTGTGATGTTTTCTGCGGTTGGT	<i>csy2</i> gene expression detection for qPCR
<i>Csy2-qR</i>	CGGGCTTCTTCAATAAATGATGGT	
<i>Csy3-qF</i>	ACTAACGAAGTTGACAGTTTGGC	<i>csy3</i> gene expression detection for qPCR
<i>Csy3-qR</i>	CAGCACTAACTCTTCACTGGGAT	
<i>Cas6f-qF</i>	TACTCTGGCTTCCACTAACATCG	<i>cas6f</i> gene expression detection for qPCR
<i>Cas6f-qR</i>	CCTCACAATAACCAATCATAACCG	
<i>CRISPR-qF</i>	TCGCCGTACTCGTTCAAGTT	CRISPR expression detection for qPCR
<i>CRISPR-qR</i>	TTGTTGATTGTCGCAACGCA	
<i>dotA-qF</i>	ACGGAACGGGAAGCCAGATA	<i>dotA</i> gene expression detection for qPCR
<i>dotA-qR</i>	CGAAGGTGGAACGAAGAGGA	
<i>dotB-qF</i>	TCCGCTGTAGTAAGCCAATC	<i>dotB</i> gene expression detection for qPCR
<i>dotB-qR</i>	TCACGACATTCACCCACCAT	

<i>dotC</i> -qF	CCCAATGTCACTCTACTCCC	<i>dotC</i> gene expression detection for qPCR
<i>dotC</i> -qR	CATACCGCCGAAATCTTCTT	
<i>dotD</i> -qF	GCTGCGCTGGAACAATGAAA	<i>dotD</i> gene expression detection for qPCR
<i>dotD</i> -qR	GACCAATCGACACTGGCTCT	
<i>icmV</i> -qF	GTGGTTATGATCGCATTGGT	<i>icmV</i> gene expression detection for qPCR
<i>icmV</i> -qR	AACCCTTGCCGTGACCATTC	
<i>icmW</i> -qF	TAGGGCAAGAACTTGATGAT	<i>icmW</i> gene expression detection for qPCR
<i>icmW</i> -qR	GCTACCAGGATGGACTGTAT	
<i>icmX</i> -qF	TACCAATCCAGCACCCTAC	<i>icmX</i> gene expression detection for qPCR
<i>icmX</i> -qR	TGACTTGAGCACTTGCGTAT	
<i>icmS</i> -qF	CTTATGACGAGTCTGAGGGA	<i>icmS</i> gene expression detection for qPCR
<i>icmS</i> -qR	GGAGTATAATCACGGCTTGG	
<i>icmK</i> -qF	GCAACTGCGAATGCTACAAC	<i>icmK</i> gene expression detection for qPCR
<i>icmK</i> -qR	GCCCGCATTTGCTGTTTGAT	
<i>icmB</i> -qF	ACACGCTCCTCTGCACAATA	<i>icmB</i> gene expression detection for qPCR
<i>icmB</i> -qR	TTTATGACCCTGCGGAAGCA	

<i>icmH</i> -qF	AGCAATACCCGTCCTCACTT	<i>icmH</i> gene expression detection for qPCR
<i>icmH</i> -qR	CCTCTACTGGAGGTAAAGAA	
<i>icmF</i> -qF	AACTCCATATCGCTGCCTAC	<i>icmF</i> gene expression detection for qPCR
<i>icmF</i> -qR	TCATTGGCTAAGGAGGGAAG	
<i>lvgA</i> -qF	TCCATTAGCGATAGTGGAGC	<i>lvgA</i> gene expression detection for qPCR
<i>lvgA</i> -qR	CCAGGTATCAGTTCGGGTTC	
<i>flaA</i> -qF	CCACGGCAACAGGAACAGAAGTA	<i>flaA</i> gene expression detection for qPCR
<i>flaA</i> -qR	ATCCCGGCATCGTTGATAGCCG	
<i>pilA</i> -qF	TGAAACGGCTATTACCAACAACG	<i>pilA</i> gene expression detection for qPCR
<i>pilA</i> -qR	CTATGGTACCACCACCTGCTGA	
<i>pilB</i> -qR	TCGTATTACAGCACGTATCAAGG	<i>pilB</i> gene expression detection for qPCR
<i>pilB</i> -qF	TACCAGAATCAAGAACCCGCATA	
<i>pilC</i> -qR	AGAGAAAGCAGCAATTGCTAGA	<i>pilC</i> gene expression detection for qPCR
<i>pilC</i> -qF	TGCTGTCCTGTTGCAACTTC	
<i>pilD</i> -qR	CCTCACTTTAGGGCTCTTATGGA	<i>pilD</i> gene expression detection for qPCR
<i>pilD</i> -qF	TCCCATACCTACTTTACCTGTCA	

<u>pilQ</u> -qR	TGCTGGAACACCAAGGAATGTT	<i>pilQ</i> gene expression detection for qPCR
<u>pilQ</u> -qF	AGGAGGCAACATCAAGTGGAGCG	
<u>pilP</u> -qR	GATCAATAGGCTTGAATGGATTTTCGC	<i>pilP</i> gene expression detection for qPCR
<u>pilP</u> -qF	TGGTTCTATTACTTAGTGCCTGCTC	
<u>pilO</u> -qR	TGCATATAGGATCCGACAATCGTA	<i>pilO</i> gene expression detection for qPCR
<u>pilO</u> -qF	ATGAACGCTTTGGGACTATGCTA	
<u>pilN</u> -qR	GGTTCTGTATCCAGACATTAGCCTC	<i>pilN</i> gene expression detection for qPCR
<u>pilN</u> -qF	CTGCAGTCAACAAGAACTTTAATGG	
<u>pilM</u> -qR	TGCACTACTTTACTGATAACAGCCG	<i>pilM</i> gene expression detection for qPCR
<u>pilM</u> -qF	AGAAATACCTGCCTCTGCCTTAG	
<u>gyrB</u> -qF	AATCCCCTGCAGCAAAAATC	Reference gene <i>gyrB</i> for qPCR
<u>gyrB</u> -qR	TGGTAAACCGCAATATCCA	

Note. ^aUnderlined sequences are restriction site adapters. The sequence in lowercase represents Ribosome Bind Site (RBS).

Supplementary Table S2. The bacterial strains and plasmids used in gene editing studies

Strains / Plasmids	Genotypes and relevant information
<i>L. pneumophila</i>	
WX48	Isolated strain possessing a Type I-F CRISPR-Cas system
ATCC 33152	<i>L. pneumophila</i> reference strain Philadelphia-1
ΔCas1	WX48 <i>cas1</i> ::Km ^r
ΔCas2-3	WX48 <i>cas2-3</i> ::Km ^r
ΔCsy1	WX48 <i>csy1</i> ::Km ^r
ΔCsy2	WX48 <i>csy2</i> ::Km ^r
ΔCsy3	WX48 <i>csy3</i> ::Km ^r
ΔCas6f	WX48 <i>cas6f</i> ::Km ^r
ΔCRISPR	WX48 CRISPR::Km ^r
Hb-Cas1	WX48 (pMMB207 <i>cas1</i>)
Hb-Cas2-3	WX48 (pMMB207 <i>cas2-3</i>)
Hb-Csy1	WX48 (pMMB207 <i>csy1</i>)
Hb-Csy2	WX48 (pMMB207 <i>csy2</i>)
Hb-Csy3	WX48 (pMMB207 <i>csy3</i>)
Hb-Cas6f	WX48 (pMMB207 <i>cas6f</i>)
Hb-CRISPR	WX48 (pMMB207CRISPR)
<i>E. coli</i>	
DH5a	<i>supE44</i> Δ <i>lacU169</i> (φ80 <i>lacZ</i> Δ M15) <i>hsdR17</i> <i>recA1</i> <i>endA1</i> <i>gyrA96</i> <i>thi-1</i> <i>relA1</i>
Plasmids	

pGEM-T Easy	Amp ^r
pLAW344	<i>oriT</i> (RK2) <i>oriR</i> (ColE1) <i>sacB</i> Cm ^r Ap ^r
pUT-mini-Tn5 Km	Km ^r
pMMB207	RSF1010 derivative, <i>IncQ</i> <i>lacIq</i> Cm ^r Ptac <i>oriT</i>
pMMB207-Cas/CRISPR	pMMB207 carrying <i>cas1</i> or <i>cas2-3</i> or <i>csy1</i> or <i>csy2</i> or <i>csy3</i> or <i>cas6f</i> or CRISPR
pT-Cas/CRISPR	pGEM-T Easy carrying <i>cas1</i> or <i>cas2-3</i> or <i>csy1</i> or <i>csy2</i> or <i>csy3</i> or <i>cas6f</i> or CRISPR
pT-Cas/CRISPR-Km	pT-Cas/CRISPR carrying <i>cas1::Km^r</i> or <i>cas2-3::Km^r</i> or <i>csy1::Km^r</i> or <i>csy2::Km^r</i> or <i>csy3::Km^r</i> or <i>cas6f::Km^r</i> or CRISPR::Km ^r
pLAW344-Cas/CRISPR-Km	pLAW344 carrying <i>cas1::Km^r</i> or <i>cas2-3::Km^r</i> or <i>csy1::Km^r</i> or <i>csy2::Km^r</i> or <i>csy3::Km^r</i> or <i>cas6f::Km^r</i> or CRISPR::Km ^r

Note. Ap^r, ampicillin resistance. Km^r, kanamycin resistance. Cm^r, chloramphenicol resistance.