## Supplementary File 3. Primers and oligos used in this study.

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| **Primers** | **Sequences** | **Function** |
| p*rcsC* F | gctctagagcagaagatgaacatgttgttggt | Plasmid construction |
| p*rcsC* R | GGAATTCATTAGCCAAGTGGATAAAGAACTGt |
| p*rcsD* F | CGGGATCCGCCGCCTAAACGGTGATCg |
| p*rcsD* R | GGAATTCCGACAACGTTTACCCATTCAATT |
| pBAD*rcsD-hpt*573bp F | catgccatggtaTtgttaaattcatttggtgctaact |
| pBAD*rcsD-hpt*573bp R | ggaattcttatgggttaccttcctgcaaca |
| pBAD*rcsD-hpt*462bp F | catgccatggtattgctcgcggcggatg |
| p*rcsD*C-term F | CGGGATCCGCGGGTATTTCTGATGAAGAGAT |
| p*rcsD*N-term R | GGAATTCCTGGTTTTTAGTTGCCTCTCATAGAG |
| *rcsC* deletion F | Tcggttatcggtagtatattctgttgttaacacagcctgtctggcgattccggggatccgtcgacc | Gene deletion by Recombineering |
| *rcsC* deletion R | atttttgccccgctcgactctcaagcccagttttgctctgcaatacgtgtaggctggagctgcttcg |
| *rcsD* deletion F | gcgccagagccgtcacgcgctaaccaagggaatcggcattatcgtt attccggggatccgtcgacc |
| *rcsD*2530bp down deletion R | tgtacggtttgcgaaagtgatgttaaatcactttgctgaagttcagtgtaggctggagctgcttcg |
| *rcsD*1898bp up deletion F | tatgtaaccaattatgtaaaaaattaaacggccaattagagattc attccggggatccgtcgacc |
| *rcsF* deletion F | TTGCTTCAATAGCACAACTTAAATGAATAAGGCTGAGGAATTTTTattccggggatccgtcgacc |
| *rcsF* deletion R | ttagatgaaacgttcagtgctgaaccctgacacaccgcttgttga gtgtaggctggagctgcttcg |
| *igaA* deletion F | tttcatagcgttgacgtaaatttaaagcgctctgatggggatggg attccggggatccgtcgacc |
| *igaA* deletion R | cgtgcatctgctggcatgagcgatgatcaaccggagatgggtgaggtgtaggctggagctgcttcg |
| *rcsD* fused 3xFlag F | gattataaagatcatgacatcgactacaaggatgacgatgacaagtgaccAtgaacaaccttaatgtaattattgctgatgac | Overlap PCR for mutagenesis |
| *rcsD* fused 3xFlag R | gtcgatgtcatgatctttataatcaccgtcatggtctttgtagtCtgggttaccttcctgcaacagtctg |
| *rcsD* fused 3xFlag and His6 F | ATGACGATGACAAGCATCATCATCATCATCACTGACCATGAACA ACCTTAATGTAATTATTGCTg |
| *rcsD* fused 3xFlag and His6 R | aggttgttcaTggTCAGTGATGATGATGATGATGCTTGTCATCGTCATCCTTGTAGTCg |
| *rcsC* (H489A) F | gcgGAATTGCGCACACCGCTGTATGGCATTATCGG |
| *rcsC* (H489A) R | AGCGGTGTGCGCAATTCCGCACTAACGGTAGCCAGAAACATGGATTTTG |
| *rcsC* (D885A) F | GCGGTGAACATGCCAAATATGGATGGTTACCGTTT |
| *rcsC* (D885A) R | ATATTTGGCATGTTCACCGCGGTCAATACCATATCTACGGTATTAGTATTTAACGC |
| *rcsC* (T913A) F | GCGGCGAATGCTTTGGCTGAGGGAAAACAAC |
| *rcsC* (T913A) R | TCAGCCAAAGCATTCGCCGCTACACCAATAATCGGGAAATTGTGATTCAA |
| *rcsD* (H844A) F | GCGCGATTAAAAGGCGTATTTGCCATGCTGAACC |
| *rcsD* (H844A) R | AATACGCCTTTTAATCGCGCTACGGTTTGCGAAAGTGATGTTAAATCACTT |
| *rcsB* (D56Q) F | AATTACTGAACTCTCTATGCCAGGGGATAAGTATGGTGATGGCATCAC |
| *rcsB* (D56Q) R | AGAGTTCAGTAATTAGCACGTTGGCATCAAGTTTGGACAAATTGTTAATAAGC |
| *rcsB* fused 3xFlag F | GATTATAAAGATCATGACATCGACTACAAGGATGACGATGACAAGtgaGCATGCCTGCAGGTCGACTCTAGA |
| *rcsB* fused 3xFlag R | GTCGATGTCATGATCTTTATAATCACCGTCATGGTCTTTGTAGTCAAGCTTGGCGTAATCATGGTCATAGCTGTTTCCT |
| *rcsD (*TTG-462**🡪**TTA) F | ttactcgcggcggatgaaacggggtttca |
| *rcsD* (TTG-462**🡪**TTA) R | Gtttcatccgccgcgagtaataatgtgtaattatcataatgttgcggattatccgttattgta |
| *rcsD (*TTG-573**🡪**CTT) F | cttcTAAATTCATTTGGTGCTAACTGCATTCTCACCGATGAAC |
| *rcsD (*TTG-573**🡪**CTT) R | GCACCAAATGAatttagaagTATTGTAATGATCGAGCGTACT |
| i*gaA* (C413S) F | CAGGGTAATGGCATGagtTATGTCCCCCCCAACATTCAAAACACACG |
| *igaA* (C413S) R | GTTGGGGGGGACATAactCATGCCATTACCCTGAGCCTTCAGCATATCG |
| *rcsF* (C125S) F | tatcaacaagcggtgAgtcagggttcagcactgaacgtttcatctaaatga g |
| *rcsF* (C125S) R | cagtgctgaaccctgacTcaccgcttgttgatagcagccagggac |
| *rcsD*pe (ATT-312**🡪**GGT) F | |  | | --- | | aaaggttgagtacGGtacagaagatgatataaacctctatgagag | |  | |
| *rcsD*pe (ATT-312**🡪**GGT) R | |  | | --- | | tatcatcttctgtaCCgtactcaaccttttgttgcgattgttc | |
| *rcsD*pe (inserting stop codon before frameshift) F | ctgacatgattttttTatgtaaccaattatgtaaaaaattaaacggccaattagagattcgca |
| *rcsD*pe (inserting stop codon before frameshift) R | ataattggttacataAAAAAAtcaTGTCAGCCCCGAACCATGATTGTATCTGTCG |
| Modified pBAD’ F | TTTTTTGGGCTAACAGGAGGttggatcccGGGGGTTCTCATCATCATCATCATCATGGTATGGC | Overlap PCR for ATG deletion in pBAD/Myc-His A |
| Modified pBAD’ R | TGATGATGATGAGAACCCCCgggatccaaCCTCCTGTTAGCCCAAAAAACGGGTATGGAGAAACA |
| *rcsD* (H844) crRNA F | TgcaaaccgtacatcgattaaaaggcGT | Cas12a mediated  genome editing |
| *rcsD* (H844) crRNA R | gccttttaatcgatgtacggtttgcATC |
| *rcsD* (H844A) oligo | tgatttaacatcactttcgcaaaccgtaGCGcgattaaaaggcgtatttgccatgctga |
| *rcsB* (D56) crRNA F | TctgacctctctatgccaggggataaGT |
| *rcsB* (D56) crRNA R | ttatcccctggcatagagaggtcagATC |
| *rcsB* (D56Q) oligo | caaacttgatgccaacgtgctaattactcagctctctatgccaggggataagtatggtg |
| *rcsD-hpt* (RBS)crRNA F | TtgtaatgtactcaaccttttgttgcGT |
| *rcsD-hpt* (RBS)crRNA R | gcaacaaaaggttgagtacattacaATC |
| *rcsD-hpt (*RBS\*) Oligo | tttatatcatcttctgtaatgtactcaacTTTGTTttgcgattgttcagataaaattcg |
| *rcsD*C-term (ATT-312) crRNA F | Tag tacattacagaagatgatataaaGT |
| *rcsD*C-term (ATT-312) crRNA R | tttatatcatcttctgtaatgtactATC |
| *rcsD*C-term (ATT-312 **🡪**GGT) oligo | tgaacaatcgcaacaaaaggttgagtacggtacagaagatgatataaacctctatga |
| *rcsD* (ATT-312**🡪**ATG) oligo | tgaacaatcgcaacaaaaggttgagtacATGacagaagatgatataaacctctatga |
| *rcsD*pe (7T) crRNA F | |  | | --- | | TggggctgacatttttttatgtaaccGT | |  | |
| *rcsD*pe (7T) crRNA R | ggttacataaaaaaatgtcagccccATC |
| *rcsD*pe (inserting stop codon before frameshift) oligo | cagatacaatcatggttcggggctgacatgatttttttatgtaaccaattatgtaaaaa |
| *rcsD*-*lacZ* frameshift test F | AtgaCcatggcgggtatttctgatgaag | Construction of pMal-*rcsD*pe /*rcsD*pstb-*lacZ* and pMal-*rcsDpe*-stop-*lacZ* |
| *rcsD*-*lacZ* frameshift test F | gtacagatcttctactttactgcgaatctctaattg |
| *rcsD*pe-stop-*lacZ* | gtacagatcttatgctactttactgcgaatctctaattg |
| 16sRNA F | TGGGAGTGGGTTGCAAAAGA | qPCR |
| 16sRNA R | GTTACGACTTCACCCCAGTCA |
| *ybtT* qPCR F | CGTTCAGAATGCAGATGCGG |
| *ybtT* qPCR R | GAAGCATCCGTATCGCCTGA |
| *osmY* qPCR F | ACGCTTTTCACGCCGTTTAC |
| *osmY* qPCR R | GGTGTCGTACAGCTATCGGG |
| *glnH* qPCR F | TCAGCAGTACGGTGTTGCAT |
| *glnH* qPCR R | TTCAGCGTAAGTGCCGTCTT |
| *tus* qPCR F | ACTGATGCTGACAGAACGCA |
| *tus* qPCR R | CGTGAGCAATGGAGGGAACT |
| *hmsT* qPCR F | TGCTATCATCGTCGCCCAAG |
| *hmsT* qPCR R | ATTTCTGCCCGTGTCGTTCT |
| *igaA* qPCR F | CCATCCGGCGACGATTTTTC |
| *igaA* qPCR R | CATTGTCACTCTGGGTCGCT |
| *lsrF* qPCR F | ATTGGCATCCCACAGCAGAA |
| *lsrF* qPCR R | TAGCCATGGTCAAAGGCCAG |
| *irp2* qPCR F | TGGTAGCGATCTTCAGGGGA |
| *irp2* qPCR R | ACCGACTTCTTCCAGCAAGG |
| *pgm*-test F (*hmsS*) | CGCCCCTGATTTTTACGG | PCR for testing the loss of *pgm* locus |
| *pgm*-test R (*hmsS*) | CATCCCTGGCGTAAATGG |
| *pgm*-test F (upstream of *pgm* region) | ctttagattgtatgtaggcaagaga |
| *pgm*-test R (downstream of *pgm* region ) | TGAGGCAGGTAGGTATCG |
| *pgm*-test F (*ybtT*) as qPCR | CGTTCAGAATGCAGATGCGG |
| *pgm*-test R (*ybtT*) as qPCR | GAAGCATCCGTATCGCCTGA |