**Table S1: Strains and plasmids used in this study**

|  |  |  |
| --- | --- | --- |
| Strain | Description or phenotype | Reference/source |
| ***E. coli*** |  |  |
| BL21 (DE3) | Lysogen of λ DE3 carrying a chromosomal copy of the T7 RNA polymerase gene under control of the *lacUV5* promoter | Novagen |
| CC118(λpir) | Host strain for pKNG101 replication ∆(ara-leu ) araD ∆lacX74 galE galK phoA20 thi-1 rpsE rpoB argE(Am) recA1 RfR(λpir) | Lab collection |
| TOP10 | F-mrcA ∆(mrr-hsdRMS-mcrBC )Φ80 lacZ∆M15 ∆lacX74 nupG recA1 araD139 ∆(ara–leu)7697 galE15 galK16 rpsL(StrR) endA1 λ- | Invitrogen |
| XL1-Blue | recA1 endA1 gyrA96 thi-1 hsdR17 supE44 relA1 lac | Stratagene |
| DH5α omnimax | [F’ proAB lacIq Z∆M15 Tn10 (TetR)] F’ proAB lacIq Z∆M15 Tn10 (TetR) ∆(ccdAB) mrcA ∆(mrr-hsdRMS-mcrBC) Φ80 lacZ∆M15 ∆(lacZYA-argF) U169 endA1 recA1 supE44 thi-1 gyrA96 relA1 tonA panD | Invitrogen |
| ***P. aeruginosa*** |  |  |
| PAO1∆*pilA*∆*fliC* | PAO1 deletion mutant for the *pilA* and *fliC* genes (PAO1∆∆) | [[1](#_ENREF_1)] |
| PAO1∆∆∆*cupB3* | PAO1∆∆ deletion mutant for the *cupB3* gene | this study |
| PAO1∆∆∆*cupB3\*R* | PAO1∆∆ deletion mutant for the *cupB3* gene | [[1](#_ENREF_1)] |
| PAO1∆∆∆*cupB5* | PAO1∆∆ deletion mutant for the *cupB5* gene | [[1](#_ENREF_1)] |
| PAO1∆∆∆*tpsB4* | PAO1∆∆ deletion mutant for the *tpsB4* gene (PA4540) | this study |
| **Plasmids** | **Description or phenotype** | **Reference/source** |
| pCR2.1 | TA cloning vector for PCR products, lacZα ColE1 f1 ori, ApR , KmR | Invitrogen |
| pCR-Blunt | Cloning vector for blunt-end DNA fragments, lacZα-ccdB pUC ori, ZeocinR , KmR | Invitrogen |
| pKNG101 | Suicide vector in *P. aeruginosa*, sacB, SmR | lab collection |
| mini-CTX1 | TcR, self-proficient integration vector with *tet*, Ω-*FRT-attP*-MCS, *ori, int,* and *oriT* | [[2](#_ENREF_2)] |
| pBBR1Mcs-4 | AmpR, broad-host-range cloning vector | [[3](#_ENREF_3)] |
| pET46 Ek/LIC | Expression vector | Novagen |
| pCR2.1-∆*cupB3* | ∆*cupB3* mutator cloned into pCR2.1 | this study |
| pCR2.1-∆*tpsB4* | ∆*tpsB4* mutator cloned into pCR2.1 | this study |
| pKNG101-∆*cupB3* | ∆*cupB3* mutator cloned into pKNG101 | this study |
| pKNG101-∆*tpsB4* | ∆*tpsB4* mutator cloned into pKNG101 | this study |
| pCR2.1-*tpsB4*mi | *tpsB4* and itspromoter cloned into pCR2.1 | this study |
| miniCTX-*tpsB4* | *tpsB4* and its promoter cloned into mini-CTX1 | this study |
| pMMB67EH-GW | Broad host range vector, IncQ, ptac, lacZα | lab collection |
| pMMB*rocS1* | *rocS1* gene cloned in pMMB67EH-GW | [[4](#_ENREF_4)] |
| pCR-Blunt-*cupB5* | *cupB5* gene and its ribosome binding site cloned into pCRblunt | this study |
| pCR-Blunt-*cupB5* ***cupB5*S69A** | ***cupB5*S69A** cloned into pCRblunt | this study |
| pCR-Blunt-*cupB5* ***cupB5*N85A,P86A** | ***cupB5*N85A,P86A** cloned into pCRblunt | this study |
| pCR-Blunt- ***cupB5*V97W,V99W** | ***cupB5*V97W,V99W** cloned into pCRblunt | this study |
| pBBR1-Mcs4-*cupB5* | *cupB5* gene and its ribosome binding site cloned into pBBR1-Mcs4 | this study |
| pBBR1-Mcs4- ***cupB5*S79A** | ***cupB5*S69A** cloned into pBBR1-Mcs4 | this study |
| pBBR1-Mcs4- ***cupB5*N85A,P86A** | ***cupB5*N85A,P86A** cloned into pBBR1-Mcs4 | this study |
| pBBR1-Mcs4- ***cupB5*V97W,V99W** | ***cupB5*V97W,V99W** cloned into pBBR1-Mcs4 | this study |
| miniCTX-***tpsB4*D61A,D149A** | ***tpsB4*D61A,D149A cloned into mini-CTX1** | this study |
| miniCTX-***tpsB4*H82W,P105A** | ***tpsB4*H82W,P105A cloned into mini-CTX1** | this study |
| miniCTX-***tpsB4*Q107A,E108A** | ***tpsB4*Q107A,E108A cloned into mini-CTX1** | this study |
| miniCTX-***tpsB4*G154W,E158W** | ***tpsB4*G154W,E158W cloned into mini-CTX1** | this study |
| miniCTX-***tpsB4*K173A,T175P** | ***tpsB4*K173A,T175P cloned into mini-CTX1** | this study |
| miniCTX-***tpsB4*ΔPOTRA1** | ***tpsB4*ΔPOTRA1 cloned into mini-CTX1** | this study |
| pB4-NT | *tpsB41-194* cloned into pET46 Ek/LIC | this study |
| pB4-P12 | *tpsB442-194* cloned into pET46 Ek/LIC | this study |
| pB4-P2 | *tpsB4122-194* cloned into pET46 Ek/LIC | this study |
| pB4-P1 | *tpsB442-121* cloned into pET46 Ek/LIC | this study |
| pB4-α1P1 | *tpsB41-121* cloned into pET46 Ek/LIC | this study |
| pA4-TPS | *tpsA41-242* cloned into pET46 Ek/LIC | this study |

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