All strains are derivatives of BL21Gold(DE3) [genotype F- ompT hsdS(rB– mB– ) dcm+ Tetr gal λ(DE3) endA Hte]

The strains lack the coding sequence of one or more abundant outer membrane proteins (OmpA, OmpC, OmpF, LamB)

|  |  |  |  |  |  |
| --- | --- | --- | --- | --- | --- |
| Knock-outs | Strain | | Genes knocked out | | Genotype |
| Single knock-outs | |  | |  |  |
|  | BL21ΔA | | *ompA* | | F– *ompT* *gal* *dcm* *lon* *hsdSB*(*rB*–*mB*–) λ(DE3 [*lacI* *lacUV5*-*T7p07* *ind1* *sam7* *nin5*]) [*malB*+]K-12(λS) Δ*ompA* |
|  | BL21ΔB | | *lamB* | | F– *ompT* *gal* *dcm* *lon* *hsdSB*(*rB*–*mB*–) λ(DE3 [*lacI* *lacUV5*-*T7p07* *ind1* *sam7* *nin5*]) [*malB*+]K-12(λS) Δ*lamB* |
|  | BL21ΔC | | *ompC* | | F– *ompT* *gal* *dcm* *lon* *hsdSB*(*rB*–*mB*–) λ(DE3 [*lacI* *lacUV5*-*T7p07* *ind1* *sam7* *nin5*]) [*malB*+]K-12(λS) Δ*ompC* |
|  | BL21ΔF | | *ompF* | | F– *ompT* *gal* *dcm* *lon* *hsdSB*(*rB*–*mB*–) λ(DE3 [*lacI* *lacUV5*-*T7p07* *ind1* *sam7* *nin5*]) [*malB*+]K-12(λS) Δ*ompF* |
| Double knock-outs |  | |  | |  |
|  | BL21ΔAB | | *ompA*, *lamB* | | F– *ompT* *gal* *dcm* *lon* *hsdSB*(*rB*–*mB*–) λ(DE3 [*lacI* *lacUV5*-*T7p07* *ind1* *sam7* *nin5*]) [*malB*+]K-12(λS) Δ*ompA* Δ*lamB* |
|  | BL21ΔAC | | *ompA*, *ompC* | | F– *ompT* *gal* *dcm* *lon* *hsdSB*(*rB*–*mB*–) λ(DE3 [*lacI* *lacUV5*-*T7p07* *ind1* *sam7* *nin5*]) [*malB*+]K-12(λS) Δ*ompA* Δ*ompC* |
|  | BL21ΔAF | | *ompA*, *ompF* | | F– *ompT* *gal* *dcm* *lon* *hsdSB*(*rB*–*mB*–) λ(DE3 [*lacI* *lacUV5*-*T7p07* *ind1* *sam7* *nin5*]) [*malB*+]K-12(λS) Δ*ompA* Δ*ompF* |
|  | BL21ΔBC | | *lamB*, *ompC* | | F– *ompT* *gal* *dcm* *lon* *hsdSB*(*rB*–*mB*–) λ(DE3 [*lacI* *lacUV5*-*T7p07* *ind1* *sam7* *nin5*]) [*malB*+]K-12(λS) Δ*ompC* Δ*lamB* |
|  | BL21ΔBF | | *lamB*, *ompF* | | F– *ompT* *gal* *dcm* *lon* *hsdSB*(*rB*–*mB*–) λ(DE3 [*lacI* *lacUV5*-*T7p07* *ind1* *sam7* *nin5*]) [*malB*+]K-12(λS) Δ*ompF* Δ*lamB* |
|  | BL21ΔCF | | *ompC*, *ompF* | | F– *ompT* *gal* *dcm* *lon* *hsdSB*(*rB*–*mB*–) λ(DE3 [*lacI* *lacUV5*-*T7p07* *ind1* *sam7* *nin5*]) [*malB*+]K-12(λS) Δ*ompC* Δ*ompF* |
| Triple knock-outs |  | |  | |  |
|  | BL21ΔABC | | *ompA*, *lamB*, *ompC* | | F– *ompT* *gal* *dcm* *lon* *hsdSB*(*rB*–*mB*–) λ(DE3 [*lacI* *lacUV5*-*T7p07* *ind1* *sam7* *nin5*]) [*malB*+]K-12(λS) Δ*ompA* Δ*ompC* Δ*lamB* |
|  | BL21ΔABF | | *ompA*, *lamB*, *ompF* | | F– *ompT* *gal* *dcm* *lon* *hsdSB*(*rB*–*mB*–) λ(DE3 [*lacI* *lacUV5*-*T7p07* *ind1* *sam7* *nin5*]) [*malB*+]K-12(λS) Δ*ompA* Δ*ompF* Δ*lamB* |
|  | BL21ΔACF | | *ompA*, *ompC*, *ompF* | | F– *ompT* *gal* *dcm* *lon* *hsdSB*(*rB*–*mB*–) λ(DE3 [*lacI* *lacUV5*-*T7p07* *ind1* *sam7* *nin5*]) [*malB*+]K-12(λS) Δ*ompA* Δ*ompC* Δ*ompF* |
|  | BL21ΔBCF | | *lamB*, *ompC*, *ompF* | | F– *ompT* *gal* *dcm* *lon* *hsdSB*(*rB*–*mB*–) λ(DE3 [*lacI* *lacUV5*-*T7p07* *ind1* *sam7* *nin5*]) [*malB*+]K-12(λS) Δ*lamB* Δ*ompC* Δ*ompF* |
| Quadruple knock out |  | |  | |  |
|  | BL21ΔABCF | | *ompA*, *lamB*, *ompC*, *ompF* | | F– *ompT* *gal* *dcm* *lon* *hsdSB*(*rB*–*mB*–) λ(DE3 [*lacI* *lacUV5*-*T7p07* *ind1* *sam7* *nin5*]) [*malB*+]K-12(λS) Δ*ompA* Δ*ompC* Δ*ompF* Δ*lamB* |

Primers

*ompA* locus: 648 + 649, expected size ~130 bp

*ompC* locus: 650 +651, expected size ~130 kb

*ompF* locus: 652 + 653, expected size ~130 bp

*lamB* locus: 646 + 647, expected size ~150 bp

Primer sequences:

646: AAA AGA AAA GCA ATGACT CAG GAG ATA GAA TG

647: GGT TTT GCT ATT ACCACC AGA TTT CCA TCT G

648: ATTTTGGATGATAACGAGGCGCAAAAAATG

649: GAACTTAAGCCTGCGGCTGAGTTAC

650: GCA AAT AAA GGC ATA TAA CAG AGG GTT AAT AAC ATG

651: ATA TCA ATC GAG ATT AGA ACT GGT AAA CCA GAC C

652: aggtgtcataaaaaaaaccatgagggtaataaataatg

653: gaggtgtgctattagaactggtaaacgatacc