Product Manual

Product name: BL21(DE3) ΔserB Chemically Competent *E. coli* Cells for Phosphoserine Incorporation

Catalog# BLPB-201**, Size: 10 X 0.1 ml/tube**

Product details:
BL21(*λ*DE3) ΔserB is an engineered strain for specific co-translational O-phospho-L-serine incorporation in a protein by an *Escherichia coli*. To prevent possible enzymatic dephosphorylation of O-phospho-L-serine (Sep) in vivo, the gene encoding phosphoserine phosphatase (serB), which catalyzes the last step in serine biosynthesis, was deleted from Escherichia coli strain BL21(DE3).

BL21(*λ*DE3) ΔserB chemically competent cells are supplied as a pack of 10 convenient 100 µl/tube aliquots.

The transformation efficiency is ≥ $1x10^6$ cfu/µg pBR322 plasmid DNA.

E. coli genotype: *fhuA2 [lon] ompT gal (λDE3) [dcm] ΔhsdS ΔserB*

[λDE3 = λ sBamHIo ΔEcoRI-B int::(lacI::PlacUV5::T7 gene1) i21 Δnin5]

Suggested Transformation Procedure for Optimal Results:

1. Remove cells from -80°C and let thaw on ice.
2. Gently mix cells by lightly flicking tube. Aliquot ~50-100 µl of cells into chilled, 17 x 100 mm polypropylene tube(s). Unused cells may be refrozen, but a drop in efficiency may result. For optimal recovery, refreeze cells in a dry ice/ ethanol bath prior to -80°C stora ge.
3. Add DNA solution (≤5 µl per 50 µl cells) to cell suspension and gently swirl tube(s) for a few seconds to mix.
4. Incubate on ice for 30 minutes.
5. Place tube(s) in 42°C water bath for ~30 to 45 seconds without shaking. For 50 µl aliquots, 30 seconds is recommended for maximum efficiency.
6. Place tube(s) on ice for ~2 minutes.
7. Add 200-250 µl SOC to transformation reaction(s).
   SOC medium: 2% Tryptone, 0.5% Yeast Extract, 0.4% glucose, 10 mM NaCl, 2.5 mM KCl, 10 mM $MgCl_2$ & 10 mM $MgSO_4$. Other media can be used to grow transformed cells, including standard LB or TB broths. However, SOC is the optimal choice for recovery of the cells and for obtaining maximum transformation efficiencies.
8. Shake tube(s) ~200 rpm for 1 hour at 37°C.
9. Plate by spreading 100-200 µl of cell transformation mixture on LB agar plates containing appropriate antibiotic and incubate overnight at 37°C.

Storage is recommended at -80°C.

Usage: This product is intended for LABORATORY RESEARCH USE ONLY. Not for diagnostic or therapeutic use.

Important Note: Purchase of BL21(*λ*DE3) ΔserB cells includes research use limited license for use only at site of purchase. This license prohibits the purchaser from selling, assigning or transferring this product to any third party without the express written consent of Amid Biosciences, LLC. Please review this license before purchasing this product.

References

Park HS, et al. Expanding the genetic code of Escherichia coli with phosphoserine. Science. 2011 Aug 26;333(6046):1154. 10.1126/science.1207203