BL21 Combo Pack

Shipping: On Dry Ice Catalog numbers

Batch No.: See vial BIO-85035

BIOLINE
A Meridian Life Science® Company

Store at –80°C

Storage and stability: BL21 Chemically Competent Cells are shipped on Dry/Blue Ice and stored at –80°C.

Expiry: When stored under the recommended conditions and handled correctly, full activity of the cells is retained until the expiry date on the outer box label.

Product Specifications:

Product Efficiency	Pack Size	Control Vector
Combo Pack	10^7 cfu/µg of pUC19	1.5ml (15 x 100µl) pUC19 (100pg/µl)

Genotype:

BL21 F⁻ *omp*T hsdS_B($r_B^-m_B^-$) *gal dcm*
BL21(DE3) F⁻ *omp*T hsdS_B($r_B^-m_B^-$) *gal dcm* (DE3)
BL21(DE3)pLysS F⁻ *omp*T hsdS_B($r_B^-m_B^-$) *gal dcm* (DE3) pLysS (Cam^R)
BL21(DE3)pLysE F⁻ *omp*T hsdS_B($r_B^-m_B^-$) *gal dcm* (DE3) pLysE (Cam^R)

Safety precautions: This product is for R&D use only, not for human use, or any other use. Please refer to the material safety data sheet for information regarding hazards and safe handling practice.

Notes: Research Use Only.

Features

- Contains, BL21(DE3), BL21 (DE3) pLysS and BL21 (DE3) pLysE competent cells
- High-level protein expression
- Protease deficient
- Transformation efficiency: ≥1 x 10^7 cfu/µg of pUC19
- IPTG inducibility helps to minimize toxic effects of some recombinant proteins

Applications

- Non-T7 promotor protein expression (BL21)
- T7 promotor expression (BL21 (DE3))
- Regulation of basal T7 promotor expression (BL21), (DE3) pLysS, BL21 (DE3) pLysE)

Description

BL21 and its λDE3 lysogenic derivatives are all-purpose *E. coli* host strains for high-level expression of a variety of recombinant proteins. All strains are deficient in both *lon* and *omp*T proteases resulting in a higher level of intact recombinant proteins. BL21 competent cells are an ideal host for optimal expression of proteins from vectors utilizing *E. coli* promotors (this strain lacks a source of T7 RNA polymerase).

Suggested Transformation Procedure for Optimal Results:

1. Remove cells from –80°C and let thaw on wet ice.
2. Gently mix cells by lightly flicking tube. Aliquot ~50-100µl of cells into chilled, 17 x 100mm polypropylene tube(s), e.g., Falcon 2059. Unused cells may be refrozen, but a small drop in efficiency may result. For optimal recovery, refreeze cells in a dry ice/ ethanol bath prior to -80°C storage.
3. Add DNA solution (≤5µl per 50µl cells) to cell suspension and gently swirl tube(s) for a few seconds to mix. If a control is desired, repeat this step with 2µl of the provided Control Vector (pUC19) in a separate tube.
4. Incubate on ice for 30 minutes.
5. Place tube(s) in 42°C water bath for ~30 to 45 seconds without shaking. For 50µl aliquots in Falcon 2059 tubes, 30 seconds is recommended for maximum efficiency.
6. Replace tube(s) on ice for ~2 minutes.
7. Dilute transformation reaction(s) to 1ml by addition of 900-950µl SOC. SOC Medium: 2% Tryptone, 0.5% Yeast Extract, 0.4% glucose, 10mM NaCl, 2.5mM KCl, 10mM $MgCl_2$ & 10mM $MgSO_4$.
8. Shake tube(s) ~200 rpm for 60 minutes at 37°C.
9. Plate by spreading 5-200µl of cell transformation mixture on LB agar plates containing appropriate antibiotic and incubate overnight at 37°C.

When performing the pUC19 control transformation, plate 5µl of the transformation mixture on a LB agar plate containing 100µg/ml ampicillin. To facilitate cell spreading, place a pool of SOC (100µl) onto surface of plate prior to addition of transformation mixture.

Transformation Efficiency Calculation for Control Vector

$$\text{Transformation Efficiency (cfu/µg pUC19 DNA)} = \frac{\text{\# colonies (colony forming units)}}{\text{pg pUC19 transformed}} \times \frac{10^6\ \text{pg}}{\text{µg}} \times \frac{\text{Final volume (µl) of transformation mix}}{\text{Volume plated (µl)}}$$

For example:
If 40 colonies were obtained after transforming 20pg of pUC19 and plating 5µl of the final 1ml transformation mixture, the calculated transformation efficiency would be:

$$\frac{40\text{cfu}}{20\text{pg pUC19}} \times \frac{10^6\ \text{pg}}{\text{µg}} \times \frac{1000\text{µl}}{5\text{µl}} = 4 \times 10^8\ \text{cfu/µg pUC19}$$

Competent Cells Supplied:

BL21 Combo Pack contains five aliquots of each of the following:
BL21 (DE3) **Green**
BL21 (DE3) pLysS **Purple**
BL21 (DE3) pLysE **Orange**
and sufficient control plasmid

Associated Products:

Product Name	Pack Size	Cat No
T4 DNA Ligase	500 Units	BIO-27026
Quick-Stick Ligase	50 Reactions	BIO-27027
IPTG	5g	BIO-37036
X-GAL	1g	BIO-37035

Product Citations:

1 Debdip Ghosh, G. & Berg, J. M. *J. Am. Chem. Soc.* **132(11)**, 3973–3979 (2010).
2. Diekmann, S. *et al. Eur. J. Human Gene.* **18**, 985-92 (2010).
3. Jensen, K. K. *Euro. J. Pharmacol.* doi:10.1016/j.ejphar.2010.09.080 (2010).
4. Markham, A. P. *et al. Biochem.* **48(43)**, 10353-61 (2009).
5. Nayeem, N., *et al. Mol. Pharmacol.* **76(3)**, 534-542 (2009).
6. Allerston, C., *et al. Mol. Gen. Metab.* **98(2)**, 198-202 (2009).

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