BL21(DE3) Chemically Competent Cell

Cat. No. CD601

Storage: at -70°C for six months. Do not store in liquid nitrogen.

Description

BL21(DE3) Chemically Competent Cell is specifically designed for chemical transformation of DNA. It permits a transformation efficiency of over 10^7 cfu/μg DNA (tested by pUC19 plasmid DNA).

Genotype

F- *ompT hsdS*(r_B-m_B-) *gal dcm*(DE3)

Features

- Transformation efficiency: >10^7 cfu/μg (pUC19 DNA).
- DE3 strains contains the λDE3 lysogen that carries the gene for T7 RNA polymerase.
- Suitable for T7 and T7lac such as pET, *pEASY*®.
- Suitable for high expression of non-toxic protein.
- Control plasmid I (Amp^+) is used for detection of expression function of cell. The protein size is about 25 kDa.

Procedures

- Equilibrate a water bath to 42°C.
- Warm a vial of SOC medium or LB medium to room temperature. Warm selective plates at 37°C for 30 minutes.
- Thaw a vial of 100 μl of BL21(DE3) Chemically Competent Cell on ice, aliquot 50 μl of the cells into a prechilled 1.5 ml tube, add target DNA (1 to 5 μl) into the tube. Do not mix by pipetting up and down. Incubate the cells on ice for 30 minutes.
- Heat-shock the cells for 45 seconds at 42°C without shaking. Immediately transfer the tube to ice. Incubate on ice for 2 minutes without shaking.
- Add 500 μl of prewarmed SOC medium or LB medium (without antibiotic) into the tube, mix well and shake at 37°C for 1 hour at 200 rpm.
- Spread 20 to 200 μl from each transformation vial on a prewarmed selective plate. The remaining can be stored at 2-8°C and plated the next day if needed.
- Invert the plate and incubate at 37°C overnight.
- Select colonies and analyze by restriction enzyme digestion, PCR, or sequencing.

Notes

- Higher efficiency transformation can be achieved by transforming cells immediately following thawing.
- Avoid repeated thawing.
- Gentle handling is required for the entire procedure.

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