

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/ μ g DNA (tested by pUC19 plasmid DNA).

Genotype

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

Features

- Transformation efficiency: $>10^7$ cfu/ μ g (pUC19 DNA).
- DE3 strains contains the λ DE3 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^r) 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 μ l of BL21(DE3) Chemically Competent Cell on ice, aliquot 50 μ l of the cells into a prechilled 1.5 ml tube, add target DNA (1 to 5 μ l) 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 μ l 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 μ l 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.

For research use only, not for clinical diagnosis.

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