

PRODUCT INFORMATION

Product Name : Competent Cell BL21

Code No. : DS240

Size : 100 μ l \times 10

Competency : $> 5 \times 10^7$ cfu/ μ g (pUC19)

Supplied product : SOC medium, 1 ml \times 10

This product is for research use only

Description :

E. coli BL21 strain is a derivative of *E. coli* B strain and lacks both the *lon* protease and the *ompT* membrane protease. The lack of these proteases reduces degradation of proteins when the strain is used as a host for protein expression. *E. coli* BL21 strain has a mutation of DNA cytosine methylase (*dcm*). The mutation blocks a methylation of internal cytosine residues on plasmid DNAs, which is used for cleavage of the plasmids by some restriction enzymes.

Genotype of *E. coli* strain BL21 : F^- *ompT hsdS*($r_B^- m_B^-$) *gal dcm*

Composition of SOC medium supplied :

20 g/L	tryptone
5 g/L	yeast extract
0.5 g/L	NaCl
0.186 g/L	KCl
2.4 g/L	MgSO ₄ , anhydrous
4 g/L	glucose

Storage condition :

Stable at -80°C with little or no loss in transformation efficiency for 12 months from the date of receipt. Competent cells are sensitive to variation in temperature. Must be stored at -80°C. Upon receipt, store the Competent Cell BL21 in a freezer at -80°C directly from the dry ice shipping box and store SOC medium at room temperature or -80°C.

Handling of competent cells :

- Competent cells are sensitive to mechanical shock. Excessive mixing should be avoided.
- After thawing competent cells on ice, cells tend to lose transformation efficiency gradually. Transformation should be started immediately after the cells have thawed on ice.
- Use of refrozen competent cells is not recommended.

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Quality Control :

Transformation was carried out according to the method described in this Product Information using supercoiled pUC19 plasmid. Transformants were plated on LB plates containing 50 µg/ml ampicillin. The efficiency was confirmed to be greater than 5×10^7 cfu/µg.

Transformation Procedure :

- Materials to be supplied by user

- LB plates containing antibiotic
- 15 ml sterilized-polypropylene culture tubes
- 37°C shaker
- Ice bucket with ice
- 42°C water bath
- Sterile spreaders

- Transformation

1. Thaw one tube of competent cells on ice. One tube contains 100 µl of cells for each transformation.
2. Add DNA sample* directly into the competent cells and mix by flicking the tube.
 - * The volume of DNA sample should not exceed 5 % of that of competent cells (i.e. for 100 µl of competent cells, use ≤ 5 µl).
3. Leave the tube on ice for 20 minutes.
4. Heat Shock the cells by placing the tube in a 42°C water bath for 45 seconds. Do not mix or shake during this time.
5. Remove the tube from the water bath and place them on ice for 2 min.
6. Transfer the cells to a 15 ml sterilized culture tube containing 0.9 ml of SOC medium (pre-warmed at room temperature). Culture the cells at 37°C for 1 hr in a shaker.
7. Spread an aliquot of the cells* to a LB agar plate containing the appropriate antibiotic.
 - * If plating <100 µl of the cells, mix SOC medium to the cells up to 100 µl then spread them.
8. Incubate the plates at 37°C overnight.

Reference:

Sambrook, J. and Russell, D.W. (2001) Molecular Cloning: A Laboratory Manual, 3rd ed., Cold Spring Harbor Laboratory Press, Cold Spring Harbor, NY.

Related Products:

DS250	Competent Cell BL21(DE3)	DS255	Zip Competent Cell BL21(DE3)
DS258	Electrocompetent Cell BL21(DE3)	DS260	Competent Cell BL21(DE3)pLysS