

## PRODUCT INFORMATION

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**Product Name :** Giga Competent Cell (DH5 $\alpha$ )

**Code No. :** DS229M

**Size :** 100  $\mu$ l  $\times$  5

**Competency :**  $> 1 \times 10^9$  cfu/ $\mu$ g (pBR322)

**Supplied Product :** SOC medium, 1 ml  $\times$  5

*This product is for research use only*

### Description :

Giga Competent Cell (DH5 $\alpha$ ) is developed to confer extremely high transformation efficiency and an ideal product for DNA cloning, library construction and a variety of other applications. The Giga Competent Cell is prepared from *E. coli* DH5 $\alpha$  stain (one of the standard strains for molecular biology applications) by advanced chemical procedure. The DH5 $\alpha$  cell has mutation of  $\phi$ 80*lacZ* $\Delta$ M15 and lacks *laqI*<sup>q</sup> gene, which allows blue-white color screening of transformants with X-gal (IPTG is not required).

### Genotype of *E coli* Strain DH5 $\alpha$ :

*supE44,  $\Delta$ lacU169( $\phi$ 80*lacZ* $\Delta$ M15), *hsdR17, recA1, endA1, gyrA96, thi-1, relA1**

### Quality Control :

Transformation was carried out according to the method described in this Product Information using supercoiled pBR322 plasmid. Transformants were plated on LB plates containing 50  $\mu$ g/ml ampicillin. The efficiency was confirmed to be greater than  $1 \times 10^9$  cfu/ $\mu$ g.

### Storage Condition :

Stable at -80°C with little or no loss in transformation efficiency for 6 months from the date of receipt. Competent cells are very sensitive to variation in temperature. Must be stored at -80°C. Upon receipt, store the competent cells in a freezer at -80°C directly from a dry ice shipping box and store SOC medium at room temperature or at -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 following thawing cells on ice.
- Use of refrozen competent cells is not recommended.

### 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 <sub>4</sub> , anhydrous
4 g/L	glucose

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### Transformation Procedure :

- Materials to be supplied by user

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

If blue-white screening is required to select transformants,

- 20 mg/ml X-Gal in dimethylformamide (DMF)

- **Transformation**

1. Thaw one tube of Giga Competent Cell on ice. One tube contains 100  $\mu$ l 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  $\mu$ l of competent cells, use  $\leq$  5  $\mu$ l).
3. Incubate the tube on ice for 20 minutes.
4. Heat Shock the cells by placing the tube in 42°C water bath for 45 seconds. Do not mix or shake.
5. Remove the tube from the 42°C bath and place it 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 to 37°C). Culture the cells at 37°C for 1 hr in a shaker.
7. Spread an aliquot of the cells onto an LB agar plate containing appropriate antibiotic.  
If blue-white color screening is required, spread 25  $\mu$ l of 20 mg/ml X-Gal onto an LB agar plate and allow the reagent to absorb 30 minutes prior to inoculating cells. As DH5 $\alpha$  does not have *lacI<sup>q</sup>*, IPTG is not required for blue-white screening.
8. Incubate the plate 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:

DS210	Competent Cell JM109	DS220	Competent Cell DH5 $\alpha$
DS225	Jet Competent Cell (DH5 $\alpha$ )	DS255	Zip Competent Cell BL21(DE3)