BioDynamics Laboratory Inc.

PRODUCT INFORMATION

Product Name : Giga Competent Cell (DH5α) GX

Code No. : DS229G Size : $100 \mu l \times 10$

Competency: $> 1 \times 10^9 \text{ cfu/µg}$ (pBR322)

Supplied Product: SOC medium, $1 \text{ ml} \times 10$ This product is for research use only

Description:

Giga Competent Cell (DH5 α) 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 α stain (one of the standard strains for molecular biology applications) by advanced chemical procedure. The DH5 α cell has mutation of $\phi 80 lac Z \Delta M15$ and lacks $laq I^q$ gene, which allows blue-white color screening of transformants with X-gal (IPTG is not required).

Genotype of *E coli* Strain DH5α:

supE44, ΔlacU169(φ80lacZΔM15), 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 μ g/ml ampicillin. The efficiency was confirmed to be greater than 1×10^9 cfu/ μ 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 ₄ , 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 μ 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 µl of 20 mg/ml X-Gal onto an LB agar plate and allow the reagent to absorb 30 minutes prior to inoculating cells. As DH5α does not have lacl^q, 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 DH5α
DS225	Jet Competent Cell (DH5α)	DS255	Zip Competent Cell BL21(DE3)