

## DH5α Fast Chemically Competent Cell

### Product description

DH5α Fast Chemically Competent Cell from YEASEN is a competency cell obtained by a special process of DH5α strain. It can be used for the chemical transformation of DNA with the conversion efficiency up to  $10^8$  cfu/μg plasmid DNA. It is widely used for efficient DNA cloning and plasmid amplification, and can ensure stable inheritance of high-copy plasmids. At the same time, it can be used to construct clones, blue-white spot screening. Deletion of *recA1* and *endA1* genes in DH5α cells ensured the stability of cloned DNA. F-DH5α competent cells were prepared by special technology, without heat shock at 42 °C or incubated at 37 °C, only 10 minutes to complete a transformation.

DH5α competent cell genotypes: F-  $\phi 80$  *lacZ*ΔM15 Δ(*lacZYA-argF*) U169 *endA1 recA1 hsdR17(r<sub>K</sub><sup>-</sup>,m<sub>K</sub><sup>+</sup>) supE44λ- thi-1 gyrA96 relA1phoA*

### Components

Components No.	Name	11803ES80
11803-A	DH5α Fast Chemically Competent Cell	10×100 μL
11803-B	pUC19 (control vector,10 pg/μL)	10 μL

### Specifications

Species	<i>Escherichia coli</i>
Cell type	Chemically competent cell
Whether it contains an F' episome	No
Efficiency	$\geq 1 \times 10^8$
Blue/white colony screening	Yes
Bacterial or yeast strains	DH5α
Amplification of the ccdB-containing vector	It is not suitable for amplification of ccdB-containing vectors

### Storage

The product is shipped with dry ice and can be stored at -85°C~-65°C for six months. Do not store the product in -20 °C or liquid nitrogen.

### Instructions

#### 1. Rapid transformation steps (10 min)

- 1) Preheat the screening plate to 37°C 15 minutes in advance.
- 2) Take out a F-DH5α from -80°C and quickly insert into the ice. After the bacteria melted, add the target DNA (plasmids or linking products), stir gently and leave in the ice bath for 5 min.

\*The volume of DNA added should not exceed one tenth of the suspended fluid volume of the competent cell.

- 3) The mixture from the previous step was transferred to LB medium already prewarmed at 37°C using a 200μL

pipetting gun and evenly coated.

4) Plates were placed in an incubator at 37°C and incubated upside down overnight. If blue and white spots were selected, the plates were incubated upside down at 37°C for at least 15 hours.

## **2. Rapid heat shock transformation steps (25 min)**

1) Take out a F-DH5a from -80°C and quickly insert into the ice. After the bacteria melted, add the target DNA (plasmids or linking products), stir gently and leave in the ice bath for 5 min.

\*The volume of DNA added should not exceed one tenth of the suspended fluid volume of the competent cell.

2) Place the EP tube in a 42°C water bath for 45 sec, then quickly transfer to ice and let stand for 2 min.

\*Do not shake during this process, otherwise it will reduce the conversion efficiency.

3) Add about 700 µL of antibiotic-free LB or 2YT medium to the centrifuge tube, mix well and recover at 37°C at 200 rpm for 10 min.

4) Evenly spread the plate and incubate the plate at 37°C.

## **3. Conventional transformation steps**

1) Take 100 µL of competent cells, ice bath, thaw (approximately 5 min).

2) Add the DNA of interest to the thawed competent cell suspension immediately, gently flick well, and let stand in an ice bath for 25 min.

\*Do not add more than one-tenth of the volume of competent cell suspension.

3) Place the EP tube in a 42°C water bath for 45 sec, then quickly transfer to ice and let stand for 2 min.

\*Do not shake during this process, otherwise it will reduce the conversion efficiency.

4) Add about 700 µL of antibiotic-free LB or 2YT medium to the centrifuge tube, mix well and recover at 37°C at 200 rpm for 60 min.

5) Collect the bacteria by centrifugation at 5,000 rpm for 1 min, leave about 100 µL of supernatant coating onto a plate containing the corresponding antibiotic, and incubate at 37°C overnight. For blue-white spot screening, incubate the plate at 37°C for at least 13 h.

## **Notes**

1. This product should not be freeze-thawed repeatedly, so as not to reduce the conversion efficiency of the competent cells.

2. The thawing time of this product on ice should not be too long.

3. For your safety and health, please wear lab coats and disposable gloves for operation.

4. This product is for research use ONLY!