



E. coli Competent Cells DH5α

Cat. No.: S2GNM03h40001 (100 µl /vial)

Description:

Genomics' DH5α are chemically competent cell, which were prepared using our proprietary process to make the cells highly efficient for immediate DNA uptake. The transformation process only takes one minute, and the steps are simplified.

Genotype:

endA1 recA1 relA1 gyrA96 hsdR17(rk-, mk+) phoA supE44 thi-1 Δ(lacZYA-argF)U169 Φ80 Δ(lacZ)M15 F

Efficiency:

>5 x 10⁸ cfu/µg

Application:

Suitable for cloning larger plasmids and constructing cDNA libraries, while also supporting blue-white colony screening.

Kit Contents:

Contents	S2GNM03h40001
E. coli Competent Cells DH5α	100 µl x 10 vial

Storage:

Always store Competent Cells at -80°C. Thaw on ice when ready for use. Do not refreeze thawed, unused aliquots.

Protocol:

Before Starting

- Turn on the water bath and set to 42°C.
- LB plates containing appropriate antibiotics, 0.1 mM IPTG and 40 µl/ml X-Gal (or spreading 40 µl of 20 mg/ml X-Gal and 4 µl of 200 mg/ml IPTG onto LB/antibiotic plates). Solution and



media containing antibiotic, IPTG, and X-Gal must be stored protected from light in order to maintain potency.

- c. Chill sterile polypropylene culture tubes and pipette tips on ice or at -20°C.

Starting

<Heat-shock cold plate>

1. Thaw one tube of competent cells (typically 100 µl) on ice until 1/3 – 1/2 volume is thawed.
2. Add pre-chilled DNA (the volume of DNA should be \leq 5% of competent cells) immediately. Mix by vortexing for 1 second or tap the tube with finger to mix well.
3. Keep the tubes on ice for 1-5 minutes to increase the transformation efficiency - ***Optional step.***
4. Heat-shock the tube in a water bath at exactly 42°C for 15 – 45 seconds.
5. Place the cell using plating beads onto a pre-chilled (4°C) and dried selective LB agar plate (LB+ antibiotics).
6. Incubate the plate at 37°C for 12-16 hours for Genomics' DH5α Competent Cells.

<Non Heat-shock cold plate>

1. Prewarm a selective LB agar plate at room temperature up to 37 °C incubator.
2. Thaw one tube of competent cells (Typically 100µl) on ice until 1/3 – 1/2 volume is thawed.
3. Add pre-chilled DNA (the volume of DNA should be \leq 5% of competent cells) immediately. Mix by vortexing for 1 second or tap the tube with finger to mix well.
4. Keep the tubes on ice for 1 – 5 minutes to increase the transformation efficiency - ***Optional step.***
5. Place the cell using plating beads onto a pre-warmed and dried selective LB agar plate (LB+ antibiotics).
6. Incubate the plate at 37°C for 12-16 hours for Genomics' DH5α Competent Cells.

Composition of Buffers and Solutions:

LB medium with antibiotic

To 1 L of distilled water, add:

10 g Bacto® Tryptone/5 g Bacto® Yeast Extract/10 g NaCl. Adjust the pH to 7.0 – 7.5 with NaOH. Autoclave and allow the autoclaved medium to cool to 55°C and add appropriate antibiotic. For LB plates, include 15 g agar prior to autoclaving.



◎ **Revision History** ◎

Description	Version	Date
Initial Release	GN-CPC-053_Protocol_V1	May 2021
Catalog Number Adjustment	S2GNM03h40001_Protocol_V2	Jan 2025
Kit Contents and Text Revisions	S2GNM03h40001_Protocol_V3	Mar 2026