



Cat #: 42-654



NSN #: 655000L068515

# GC5™ Competent Cells – High Efficiency Cat #: 42-654

**Unit Size:** 1ml

**Amount:** 5 x 200µl

**pUC19 control (10 pg/µL):** 1 x 10µl

**Storage:** Store at -70°C. FOR RESEARCH USE ONLY

## General Description

GC5 Competent Cells are chemically competent *E. coli* cells. The cells are transformed by heat-shock methods. These competent cells are resistant to the lytic bacteriophages T1 and T5.

GC5 Competent Cells are suitable for many molecular biology applications, like generating cDNA libraries from plasmid-based vectors or constructing gene banks. Blue/white screening for recombinants can be performed by including X-gal and IPTG in the agar plates.

The GC5 strain is sensitive to these common antibiotics: ampicillin, kanamycin, chloramphenicol and tetracycline. It is resistant to naladixic acid.

## General Handling

- Competent cells are very sensitive to any change in temperature. Cells must be thawed on ice. The transformation should be started immediately after the cells are thawed.
- Competent cells must be treated gently. Mix cells by swirling or gently tapping the reaction tube. Do not mix by pipetting or vortexing.
- Once thawed, the cells should be used. Re-freezing thawed competent cells will result in a significant drop in transformation efficiency.

## Genotype

F-  $\phi 80lacZ\Delta M15 \Delta(lacZYA-argF)U169 recA1 endA1 hsdR17(r_k^-, m_k^+)$   
*phoA supE44 thi-1 gyrA96 relA1  $\lambda^- tonA$*

## Efficiency

$\geq 10^9$  transformants/µg pUC19 DNA

## pUC19 DNA Storage Buffer

Control DNA is supplied in TE Buffer [10mM Tris-HCl pH 8.0, 0.1mM EDTA].

## Notes on Ligation Reactions

**Ligation reactions inhibit transformation.** Less transformants are observed from ligation reactions than from transformations with plasmid DNA.

Use 0.5µL of a ligation reaction per 50µL of competent cells. For best results, either purify the ligation mixture by ethanol precipitation prior to transformation or dilute the ligation reaction 3-fold in TE buffer and use 1µL per 50µL competent cells.

## Advance Preparations

- Equilibrate a non-shaking water bath to 42°C.
- Place SOC Medium at room temperature.
- Prepare LB agar plates with the appropriate antibiotic. If blue/white screening for recombinants is desired, the plates should include 40µg/mL X gal and 1mM IPTG.
- Agar plates should be placed in a 37°C incubator for about 30min. prior to plating.

## Transformation Protocol for Chemically Competent GC5 Cells

- Remove competent cells from -70°C and place directly in ice. Thaw cells for 5 to 10min.
- Gently mix cells by tapping tube.
- Add 1-50pg of DNA [or 1µL control DNA] into the 50µL competent cells. Swirl the pipettor tip through the cells while dispensing DNA. Gently tap tube to mix.
- Place the tubes on ice for 30min.
- Heat-shock the cells for 45sec. in a 42°C water bath. Do not shake.
- Add 450µL of room temperature SOC medium to each transformation reaction.
- Incubate at 37°C for one hour, with shaking (225 to 250rpm).
- Spread on LB agar plates containing appropriate antibiotic (e.g., 100µg/mL ampicillin for control pUC19).
- Incubate the plates at 37°C overnight (12 to 16 hours).

## SOC Medium Formulation

2% Tryptone, 0.5% Yeast Extract, 0.4% glucose, 10mM NaCl, 2.5mM KCl, 5mM MgCl<sub>2</sub>, 5mM MgSO<sub>4</sub>.

## Quality Control

Cells must have a transformation efficiency of  $\geq 1.0 \times 10^9$  transformants/µg pUC19 DNA (non-saturating conditions). Cells must show resistance to T5 phage.

## Related Products

Description	Cat #	Size*
BL21 Competent Cells	42-600 42-601	20 x 50µl 5 x 200µl
BL21 (DE3) Competent Cells	42-602 42-603 42-604	20 x 50µl 5 x 200µl 96 x 20µl plate
BL21 (DE3)pLysS Competent Cells	42-606 42-607 42-608	20 x 50µl 5 x 200µl 96 x 20µl plate
Value Efficiency GC5™ Competent Cells	42-650	10 x 200µl
High Efficiency GC10™ Competent Cells	42-658 42-660	20 x 50µl 5 x 200µl
Thunderbolt™ GC10 ElectroCompetent Cells	42-664 42-666	5 x 80µl 5 x 100µl
High Efficiency JM109 Competent Cells	42-670	5 x 200µl
SOC Medium	42-750	10 x 10mL
X-gal	20-108	1 gram
IPTG	20-109	5 grams

\*Additional sizes available. See [www.geneseesci.com](http://www.geneseesci.com)