SmartCells™ High-Efficiency Chemically Competent *E. coli*

Instruction Manual

Catalog Number
C101020
C101120

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The SmartCells™ chemically competent *E. coli* and all other components are developed, designed, intended, and sold for research use only. They are not to be used for human diagnostic or included/used in any drug intended for human use. All care and attention should be exercised in the handling of the kit components by following appropriate research lab practices.

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Kit Contents and Ordering Information

The SmartCells™ chemically competent *E. coli* contains sufficient competent cells for 20 transformations.

Catalog Number	Contents	Amount
C101020	SmartCells™ chemically competent *E. coli*. *Contains competent cells at >1 x 10^9 cfu/µg.* SmartCells™ Genotype F$^-$ *recA1 endA1 hsdR17 supE44 thi-1 gyrA96 relA1 ϕ80lacZΔM15 Δ(lacZYA-argF)U169*	20 x 50 µl
	SOC Medium	6 ml
	pUC19 Positive Control Plasmid. *Provided as a positive control to verify the transformation competency of the competent Cells.*	20 µl (10 pg/µl)
C101120	SmartCells™ F' chemically competent *E. coli*. *Contains competent cells at >1 x 10^9 cfu/µg.* SmartCells™ F' Genotype F′ *recA1 endA1 hsdR17 supE44 thi-1 gyrA96 relA1 ϕ80lacZΔM15 Δ(lacZYA-argF)U169*	20 x 50 µl
	SOC Medium	6 ml
	pUC19 Positive Control Plasmid. *Provided as a positive control to verify the transformation competency of the competent Cells.*	20 µl (10 pg/µl)

Stability and Storage

The SmartCells™ chemically competent *E. coli* is shipped frozen. For maximum stability and long-term use, store cells at –70°C upon receipt. The SOC medium should be stored at room temperature.

Product support

Telephone: 858-457-1919 or 888-428-0558 (US toll free)	**Fax:** 858-623-9494 or 858-558-3617
E-mail: tech1@genlantis.com	Web Site: http://www.genlantis.com

Introduction

Most commercially available chemically competent cells today, regardless of the claimed tranformation efficiency, frequently under perform in real transformation experiments when ligation mixtures instead of supercoiled DNA molecules are used. SmartCells™ chemically competent *E. coli* has been prepared by a unique procedure to warrant the highest and most robust transformation performance under diverse conditions. There is no need to dilute or purify your ligation mix before transformation. If needed, over 10 µl of full strength ligation mix can be added to 50 µl competent cells without significantly compromising transformation results. SmartCells™ carry genotypes suitable for most cloning needs such as blue/white selection, generation of plasmid vector based libraries or gene banks, and ability to be transformed efficiently with large plasmids. SmartCells F'™ is also available for use with M13 cloning vectors.

Transformation Protocol

1. Thaw one tube of the SmartCells™ competent cells on ice (10-15 minutes).
2. Add 1-10 µl of ligation mix to the cells; mix gently and incubate on ice for 15 to 30 minutes.
3. Heat the mix at 42ºC for 45 seconds.
4. Add 0.25 ml room temperature SOC medium and incubate at 37ºC for 1 hour in an air incubator. Shaking tubes horizontally at 225 rpm is recommended for the best efficiency.
5. Dilute transformation reaction if necessary and spread 100 µl of transformed cells on LB/Agar plates containing appropriate selection (*e.g.* ampicillin or kanamycin). Alternatively, if maximum numbers of colonies are desired, collect cells by spinning in a microfuge for 10 seconds. Resuspend cell pellet in 50 µl SOC and spread on the agar plates containing antibiotics.
6. Incubate overnight at 37ºC.

Quality Control

Kit Component	Quality Control Standard
SmartCells™ or SmartCells™ F' competent cells	Consistently yield >1.0 x 10^9 cfu/µg transformation efficiency when transformed with pUC19 plasmid.

Notes

- It is not necessary to dilute your ligation mix with TE. The SmartCells™ competent cells are prepared through a unique procedure that they will work with most full-strength ligation buffers. In our test, up to 10 µl of undiluted ligation mix could be used without significantly compromising the transformation efficiency.
- Transformation efficiencies for ligation of inserts to vectors will be slightly lower than supercoiled plasmid (between 2 to 5 fold lower).
- If unexpectedly lower number of colonies are observed, we recommend that customer test the efficiency of competent cells using the provided supercoiled pUC19 plasmid DNA as described below.
    1. Transform 5 µl (50 pg) pUC19 into 50 µl of competent cells.
    2. Follow steps 2-6 on page 4.
    3. Dilute the transformation reaction 50 fold with SOC and plate 30 µl on a LB agar plate containing 100 µg/ml ampicillin.
    4. Incubate overnight at 37ºC and count colonies. Calculate transformation efficiency as follows:

$$\frac{\text{Number of colonies}}{\text{50 pg pUC19}} \times \frac{1 \times 10^6 \text{ pg}}{\mu\text{g}} \times \frac{300\ \mu\text{l}}{\text{30 µl plated}} \times 50 \text{ (dilution factor)} = \text{CFU} / \mu\text{g}$$

For a complete list of international distributors, please visit our web site at www.genlantis.com.
For additional troubleshooting assistance, please contact our Technical Support Department at:

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