TurboCells®

Chemically Competent *E. coli*

Cat. #	Contents	Quantity
AMS.C300020	TurboCells® Chemically Competent *E. coli*	20 x 50 µl
	TurboCells® Transformation Buffer	5 ml
	pUC19 Positive Control Plasmid	20 µl (10pg/µl)
AMS.C301020	TurboCells® F' Chemically Competent *E. coli*	20 x 50 µl
	TurboCells® Transformation Buffer	5 ml
	pUC19 Positive Control Plasmid	20 µl (10pg/µl)

Shipping	Shipped on Dry Ice
Storage	Store cells at -70°C; store TurboCells Transformation Buffer at room temperature. Stable for 6 months.

Related Products	Catalog #
EZ-Spread ™ Beads, Single-Use Tubes, 50 tubes	AMS.C400050
EZ-Spread ™ Beads, Dispenser Bottle	AMS.C400100

Introduction: TurboCells® Chemically Competent *E. coli* are specially optimized to achieve excellent transformation efficiency with a novel 3-minute transformation protocol. Together with the TurboCells® Transformation Buffer, TurboCells Chemically Competent *E. coli* allow efficient transformation without the need for a lengthy transformation reaction or hour-long recovery after heat shock. In addition, with TurboCells, you can perform the heat shock step at 37ºC, eliminating the inconvenient 42ºC water bath. TurboCells Competent *E. coli* are great for routine cloning experiments. The fast and easy protocol also makes them ideal for high-throughput transformations.

When using the 3-minute transformation protocol, you can achieve transformation efficiencies of up to 5 x 10^7 – 1 x 10^8 cfu/µg of supercoiled DNA, which is more than sufficient for most cloning experiments. When using the traditional 1.5 – 2 hour transformation protocol with the TurboCells™ Competent *E. coli*, efficiencies of 1 x 10^9 cfu/µg or greater can be achieved.

TurboCells Chemically Competent *E. coli* have been prepared using a unique procedure to allow robust performance under diverse conditions. For example, it is not required 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. The genotype of TurboCells is suitable for most cloning needs such as blue/white color selection, generation of plasmid vector based libraries, and the ability to efficiently transform large plasmids (Table 1). TurboCells F´ is also available for use with M13 cloning vectors.

TurboCells® F$^-$: *recA1 endA1 hsdR17 supE44 thi-1 gyrA96 relA1 ϕ80lacZΔM15 Δ(lacZYA-argF)U169*	
TurboCells® F': *recA1 endA1 hsdR17 supE44 thi-1 gyrA96 relA1 ϕ80lacZΔM15 Δ(lacZYA-argF)U169*	
recA1	Reduced homologous DNA recombination
endA1	Low non-specific endonuclease activity and improved plasmid yield
hsd	Improved cloning efficiencies and representations of methylated DNA
lacZΔM15	Allow blue/white screening through β-galactosidase complementation

METHODS AND PROCEDURES

A. Transformation Protocol

1. Thaw one tube of the TurboCells competent cells on ice.
2. Add 5 to 50 ng of DNA, in a volume of 1 to10 µl, to the cells; mix by tapping gently and incubate on ice for 3 to 5 minutes.
3. Heat shock the mix at 37ºC for 60 seconds.
4. Dilute the transformation reaction with 200 µl TurboCells Transformation Buffer and spread 50–100 µl of transformed cells on room temperature LB/Agar plates containing the appropriate antibiotic selection (e.g. ampicillin or kanamycin). We recommend using EZ-Spread™ Plating Beads (Cat. No. C400050 & C400100) to obtain optimal plating results.
5. Incubate overnight at 37ºC.

B. Notes

a. If maximum numbers of colonies are desired, replace Step 4 with the following: collect cells by spinning in a microfuge for 10 seconds. Resuspend cell pellet in 50 µl TurboCells Transformation Buffer and spread on an agar plate containing antibiotic.

b. If satellite colonies appear on the plates after overnight incubation, we recommend the following: 1) reduce the volume of transformation reaction used for plating, 2) if ampicillin is used, replace it with more stable homologue such as Carbenicillin (disodium salt), or 3) increase the antibiotic concentration in the plate.

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c. It is not necessary to dilute your ligation mix with TE. The TurboCells competent cells are specially prepared to work with most ligation buffers. In our tests, up to 10 µl of undiluted ligation mix could be used without significantly compromising the transformation efficiency.

d. To increase transformation efficiencies and obtain higher number of colonies, we recommend the following traditional protocol:

1. Thaw one tube of the TurboCells® 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 the transformation reaction if necessary and spread 100 µl of transformed cells on LB/Agar plates containing the appropriate antibiotic selection (e.g. ampicillin or kanamycin).
6. Incubate overnight at 37ºC.

e. Transformation efficiencies for ligation of inserts to vectors will be slightly lower than supercoiled plasmid (between 5 to 10 fold lower).

f. To test the efficiency of TurboCells® Chemically Competent E. coli using the provided supercoiled pUC19 plasmid, please use the following protocol:

1. Transfer 50 µl thawed cells to chilled 2059 tubes.
2. Add 5 µl (50 pg) pUC19 control DNA to each tube. Gently tap the cells to mix with DNA.
3. Incubate cells on ice for 30 min.
4. Heat shock the cells at 42°C for 45 seconds.
5. Place on ice for 2 minutes.
6. Add 900 µl of room temperature SOC medium.
7. Shake in a 37°C incubator for 1 hour.
8. Dilute the reaction 1:50 with SOC medium (add 10 µl reaction to 490 µl SOC).
9. Plate 100 µl of this dilution on LB agar plate containing 100 µg/ml carbenicillin.
10. Incubate overnight at 37ºC and count colonies. Calculate transformation efficiency as follows:

$$\frac{\text{Number of Colonies}}{\text{20 pg pUC19}} \times \frac{1 \times 10^6 \text{ pg}}{\mu\text{g}} \times \frac{50\ \mu\text{l}}{50\ \mu\text{l plated}} \times 20^{\dagger} = \text{CFU}/\mu\text{g}$$

†20 is the dilution factor

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