

MultiShot™ StripWell TOP10 Chemically Competent E. coli

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Description

The MultiShot™ StripWell TOP10 Chemically Competent *E. coli* Kit is chemically competent TOP10 *E. coli* packaged in a 96-stripwell plate to simplify medium- and high- throughput bacterial transformation. The stripwell format allows you to use only the number of wells you need for your particular application.

Contents

Fifty microliters of chemically competent TOP10 E. coli are supplied per well with a transformation efficiency of > 1×108 cfu/µg pUC19 DNA. Each MultiShot[™] StripWell Kit contains the following reagents.

Catalog no.	Number of plates	S.O.C.	pUC19
C4096-01	1	2 × 15 mL	50 μL (500 pg)
C4096-05	5	10 × 15 mL	50 μL (500 pg)
C4096-10	10	20 × 15 mL	2 × 50 μL (1 ng)

Genotype

F- mcrA Δ(mrr-hsdRMS-mcrBC) φ80lacZΔM15 ΔlacX74 recA1 araD139 galU galK Δ(ara-leu)7697 rpsL (Str^R) endA1 nupG

Shipping and Storage

Each MultiShot $^{\text{\tiny TM}}$ StripWell Kit is shipped on dry ice. Upon receipt, store the kit at -85° C to -68° C.

Note: You may store the S.O.C. medium at 15°C to 30°C or at 2°C to 8°C.

Composition of SOC Medium and pUC19

S.O.C. Medium:

2% Tryptone; 0.5% Yeast Extract; 10 mM NaCl; 2.5 mM KCl; 10 mM MgCl2; 10 mM MgSO4; 20 mM glucose.

pUC19:

Each tube contains 10 pg/ μ L pUC19 in 5 mM Tris-HCl, 0.5 mM EDTA, pH 8.

IMPORTANT!

Do not transform chemically competent cells by electroporation. The salt content of the buffer will cause arcing and kill the cells.

Procedure introduction

The following procedure describes how we qualify the MultiShot^{TM} StripWell Kit. In addition, this information will help you determine how to best use the MultiShot^{TM} StripWell Kit for your own applications.

Total well volume

Each well holds 1.2 mL.

Before you begin

- Prepare a container of ice large enough to chill the number of wells you will be using.
- Bring the vial of S.O.C. to room temperature.
- Pre-heat a water bath to 42°C.

Procedure

- Remove a MultiShot™ StripWell plate from the freezer and remove the number of wells you need. Return any unused wells to the freezer. Place the wells in the container of ice. The cells should thaw within 1 minute.
- 2. Carefully remove the strip of caps from each set of 8 wells and keep them for further use.
- 3. Use a multi-channel pipette to add 2–5 μL DNA (2 pg–20 ng) to the wells. Keep the volume around 2 μL for uniform results.
- 4. After adding the DNA, cover the wells with the caps and incubate the cells and DNA on ice for 30 minutes.
- 5. Transfer the wells to the water bath and heat-shock them for 30 seconds at 42°C.
 - **Note:** Be careful not to contaminate the cells.
- **6.** Transfer the wells back to the ice and allow the wells to cool for 1 minute.
- 7. Remove the caps and add 250 μL S.O.C. to each well. Re-cap the wells tightly.
- 8. Incubate the wells at 37°C for 1 hour with shaking (225 rpm). We turn the wells on their side to increase aeration and secure them to the shaker.
- **9.** Plate the appropriate volume from each well. See the next page for examples and expected number of colonies.

Plating volumes and expected results

The following table describes the type of DNA, amount transformed into MultiShot™ StripWell chemically competent cells, the volume plated, and the number of colonies.

Note: We use pUC19 to qualify the kit. Transformation efficiency should be > 1×10^8 cfu/ μg and yield 100–300 colonies per plate. Variability should be no more than 5-fold between wells.

DNA	Туре	Amount Transformed	Volume Plated	Number of Colonies
pUC19	Supercoiled	5 pg plasmid	10 μL	100-300
pCR™2.1- TOPO® plus amplified 750 bp insert	TOPO® Cloning Vector	3.4 ng vector + insert	25 μL	100-300

Safety Data Sheets (SDSs)

Safety Data Sheets (SDSs) are available at www.lifetechnologies.com/support.

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