



# Champion™

# Competent Cell

For Research Use Only

Mix then Spread

High Efficiency M Easy to Use



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#### General information

Champion™ Competent Cells are chemically competent cells, which were prepared by SMOBIO to make *E. coli* perform excellent transformation efficiency. Standard transformation protocol is recommended for large plasmids or non-ampicillin selection. Time-saving transformation protocol is recommended for simple and rapid transformation. Champion™ Competent Cells are one of the fastest and simplest ready-to-use competent cell products in the world.

#### Kit contents

- (1) Champion™ Competent Cells
- (2) pUC19 Control Plasmid (5  $\mu$ l, 10<sup>-4</sup>  $\mu$ g/ $\mu$ l)
- (3) Champion™ Transformation Protocol Card

#### Shipping condition

Throughout the shipping process, the temperature is maintained under -70°C.

#### Storage and expiration

Champion™ Competent Cells must be stored between -70°C to -80°C. Subsequent freeze-thaw cycles will reduce transformation efficiency. If high efficiency is required for the experiment, do not use aliquots that have gone through several freeze-thaw cycles. The efficiency of Champion™ Competent Cells lasts for 1 year with proper storage.



# Items and ordering information

Product Name	Compatible to E. coli strain	Efficiency (cfu/μg)	Quantity	Cat. No.
Champion IM 100 High	E. coli JM109	9 >1 x 10 <sup>8</sup>	100 μl x 80 vials	CC0202
Champion™ 109 High			100 μl x 24 vials	CC0204
Champion™ 21	Champion™ 21 E. coli BL21 (DE3)	>1 x 10 <sup>7</sup>	100 μl x 80 vials	CC2102
Champion 21			100 μl x 24 vials	CC2104
Champion™ DH5α High	E. coli DH5α	>3 x 10 <sup>8</sup>	100 μl x 80 vials	CC5202
			100 μl x 24 vials	CC5204



# Genotypes and applications

Product Name	Genotype	Application	
Champion™ 109 High	e14( $McrA^-$ ) $recA1$ endA1 $gyrA96$ $hsdR17(n_c^-, m_c^+)$ $phoA$ $supE44$ $thi-1$ $relA1$ $\Delta(lac-proAB)$ (F' $traD36$ $proAB$ $laci$ * $Z\DeltaM15$	Appropriate for blue-white color and robotic screening. It is a fast growing strain forming visible colonies within 8~10 hours.	
Champion™ 21	$F' \ omp T \ \textit{hsdS}_{\beta}(r_{\beta} \ m_{\beta} \ ) \ \textit{dcm gal} \ \lambda \ (DE3)$	Appropriate host for recombinant protein expression using T7-based expression vectors.	
recA1 endA1 gyrA96 hsdR17(rc, m Champion™ DH5α High phoA supE44 relA1 thi-1 Δ(lacZYA- argF)U 169 φ80 Δ(lacZ)M15 F'		Suitable for cloning with large plasmid and cDNA library construction, and also for bluewhite colony selection.	

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### Time-saving transformation protocol for Champion™ Competent Cells

- Quickly thaw -70°C stored competent cells with hand (body temperature) for 10~20 seconds, then keep thawed competent cells on ice.
- Add DNA (ex. ligation products) to cells, keep volume of DNA less than 10% of competent cell volume.
- Vortex 1 sec or tap on the tube with finger to mix well.
- 4. Incubate on ice for 0~10 mins. (The ice-incubation will slightly increase efficiency.) \*
- Plate the bacteria/DNA mixture onto a pre-warmed (room temperature to 37°C) selective LB agar plate (LB + antibiotic).
- 6. Incubate the plates at 37°C until the colonies are suitable for analysis.
- \* Notice: For large plasmid (>6 Kb) or non-ampicillin selection, steps below are highly recommended to be added between step 4 and 5.
  - a. Heat shock the competent cells at 42°C for 45~90 seconds.
  - b. Incubate on ice for 1~5 mins.
  - c. Add 900  $\mu$ I LB or SOC medium to the competent cells and incubate at 37°C for 30~60 mins with a 200 rpm shaking.



Time-saving transformation protocol for Champion™ Competent Cells



Notice: the antibiotics concentration for Champion™ Competent Cells

Ampicillin (Ap)	20 μg/ml
Kanamycin (Km)	25 μg/ml
Tetracycline (Tc)	7.5 µg/ml
Chloramphenicol (Cm)	20 μg/ml



#### **Quality Control**

#### Efficiency assay

The transformation efficiency of each batch of Champion™ Competent Cells is greater than that described in "Items and ordering information" at the time of production. The transformation efficiency is determined following time-saving transformation protocol with incubation on ice for 5 mins.

#### Calculation of transformation efficiency is as follows:

- a. Equation for transformation efficiency= transformed colonies (transformants)/ $\mu g$  of plasmid.
- b. Example:  $100 \,\mu$ I of competent cells have been transformed with  $10^6 \,\mu$ g of pUC19 plasmid. If 550 colonies are observed on the selective plate. The transformation efficiency is:  $550/10^6$ =5.5x108 transformants/ $\mu$ g of pUC19 plasmid.

#### Contamination assay

No colony on the LB agar plate with  $20^{\sim}50~\mu g/ml$  of ampicillin when transformation is performed without plasmid.

#### α-Complementation assay

The ratio of the white colonies/ white & blue colonies of the tested plate is less than 3% (for Champion™ 109 High, and Champion™ DH5α high)



#### **Factors Affecting Transformation Efficiency**

#### Thawing methods

Shorter thawing time is more efficient than a longer thawing time. Slow thawing caused by power shortages and unstable freezers will result in decreased efficiency.

#### Methods of mixing DNA with competent cells

A one-second vortex provides more reliable transformation efficiency (1.1 times compared with mixing by a finger tap).

#### Size of plasmid

Plasmid size is known to affect the efficiency greatly. For instance, the efficiency of a supercoiled 2.7 kb and a 10 Kb plasmid (using time-saving transformation protocol) has a difference of approximately 100 times. For large plasmids (> 6 kb), the heat shock method and the recovery procedure as shown in the standard transformation protocol will improve the efficiency.

#### Heat shock treatment

For large plasmids (> 6kb), transformation efficiency with heat shock treatment will be nearly double to that without heat-shock treatment.

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#### Plating methods

Bent glass rods show the greatest efficiency, while plating loop shows less efficiency than plating beads. When handling a large quantity of samples at the same time, plating beads are recommended.

#### Concentration of antibiotic

Antibiotic concentration is critical to use of the time-saving transformation protocol.

(1) For ampicillin

The recommended concentration is 20  $\mu$ g/ml for fresh ampicillin (Ap) and 50  $\mu$ g/ml for old Ap. If a higher concentration (50~100  $\mu$ g/ml) is used, the efficiency will reduce by 3~10 times.

(2) For other antibiotics

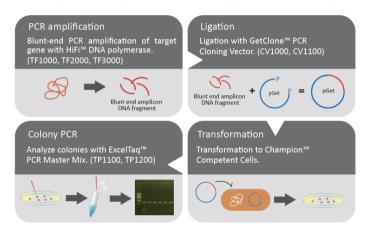
25 μg/ml Kanamycin; 7.5 μg/ml Tetracycline; 20 μg/ml Chloramphenicol.

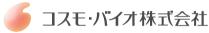
For plasmid size <6 Kb, the efficiency of kanamycin selection is usually  $3^{\sim}10$  times less than the ampicillin selection. For plasmid size > 6 Kb, the efficiency of kanamycin selection is much lower than ampicillin. We suggest using the standard transformation protocol (with heat shock and recovery steps) to enhance the efficiency.

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Fast and easy method for plasmid construction using SMOBIO smart cloning tools







More technical information can be found by visiting our website: http://www.smobio.com/Support.html