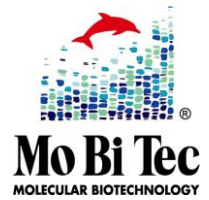


# MC1061 Chemically Competent *Escherichia coli*

Product Information Sheet  
# VS-ELS10710-01



## SUMMARY

shipped on dry ice; store at -80 °C

For research use only

## Product

Chemically competent *Escherichia coli* cells of strain MC1061.

## Description

The chemically competent *Escherichia coli* cells of strain MC1061 are prepared for heat shock transformation. MC1061 is a recombinant positive strain (*recA*<sup>+</sup>) provided for cloning and amplification of plasmid DNA of diverse Gram-positive bacteria, e.g., plasmids for expression in *Lactococcus lactis* or *Bacillus subtilis*.

## Genotype

*araD139, Δ(ara, leu)7697, ΔlacX74, galU-, galK-, hsr-, hsm+, strA* (Casadaban MJ and Cohen SN, 1980; Analysis of gene control signals by DNA fusion and cloning in *Escherichia coli*.)

## Storage and Handling

Competent cells should be stored at -80 °C. Storage at -20 °C will result in a significant decrease in transformation efficiency. Care must be taken not to interrupt the cooling chain since any storage above -80 °C will lead to a loss in transformation efficiency, even if cells do not thaw.

For handling please also consider the handbook or data sheet of the plasmid or expression vector that will be transformed.

## Transformation Efficiency

1-3 x 10<sup>6</sup> cfu/μg plasmid DNA (tested with vector pNZ2103 of *Lactococcus lactis* Constitutive Expression System, #VS-ELV01200)

## Product Contents

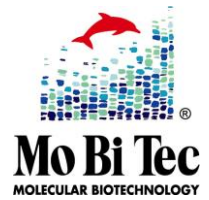
5 x 100 μl chemically competent MC1061 *E. coli* cells in transformation buffer containing 10.5% glycerol.

## Transformation Protocol

1. Thaw a tube of competent MC1061 cells on ice for 10 min.
2. Aliquot 50 μl of the competent cells to a new tube (tube 2: negative control)
3. Add 1-5 μl containing 50-100 ng plasmid DNA to the cells in the original tube (tube 1: plasmid transformation) and 1-5 μl sterile H<sub>2</sub>O<sub>dd</sub> to the cells in the control tube (tube 2).
4. Carefully mix by flicking the tubes 4-5 times. Do not vortex.
5. Place the DNA/cell mixture (tube 1) and the negative control (tube 2) on ice for 30 min.
6. Heat shock the DNA/cell mixture and the control tube at exactly 42 °C for exactly 45 sec. Do not mix!
7. Place on ice for 5 min.

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- Pipette 950  $\mu$ l of SOC medium into each tube and shake them vigorously (250 rpm) at 37 °C for 60 min or rotate (Note: SOC gives 2-fold higher transformation efficiency than LB medium).
- Prepare several dilutions (e.g., 1:10, 1:20, and 1:100) of the transformed cells (tube 1) and plate 50-100  $\mu$ l of each dilution on an LB agar plate containing an antibiotic for selection. Plate 100  $\mu$ l of the control preparation (tube 2) on an LB agar plate containing the same antibiotic. The kind of antibiotic is depending on the resistance gene of the transformed plasmid.
- Incubate 1-2 days at 37 °C.

## SOB medium (1 liter)

20 g Tryptone  
5 g Yeast extract  
0.5 g NaCl  
950 ml H<sub>2</sub>O<sub>dd</sub>

shake until all is dissolved  
add 10 ml of 250 mM KCl  
adjust to pH 7.0 with 5 M NaOH  
add H<sub>2</sub>O<sub>dd</sub> to 1 liter and autoclave

Just before use add 5 ml of a sterile 2 M MgCl<sub>2</sub> solution

## SOC medium (1 liter)

980 ml SOB medium (sterile)  
20 ml 1 M glucose (sterilized by filtration)

## Quality Warranty

Transformation efficiency ( $1-3 \times 10^6$  cfu/ $\mu$ g plasmid DNA) of the chemically competent MC1061 *E. coli* cells has been tested and verified. Untransformed cells have been controlled to be sensitive to diverse antibiotics: ampicillin (100  $\mu$ g/ml), chloramphenicol (10  $\mu$ g/ml), and kanamycin (50  $\mu$ g/ml).

## Order Information, Shipping and Storage

Order#	Product	Amount
VS-ELS10710-01	MC1061 Chemically Competent <i>Escherichia coli</i>	5 x 100 $\mu$ l
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