

# Certificate of Analysis

## Competent Cell Starter Kit

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**One Shot® TOP10 Chemically Competent *E. coli*, 20 Reactions:**

### Transformation Efficiency

50 µl of competent cells are transformed with 10 pg of supercoiled pUC19 plasmid DNA (non-saturating conditions). Test transformations are performed on a minimum of 3 vials per lot. Transformed cultures are plated on LB plates containing 100 µg/ml ampicillin and incubated overnight at 37°C.

Transformation efficiency must be greater than  $1.0 \times 10^9$  cfu/µg pUC19.

### Antibiotic Sensitivity

Cells must exhibit growth on LB medium plates.

Untransformed cells must show no growth on LB plates containing 100 µg/ml ampicillin, indicating the absence of any ampicillin resistance markers.

Untransformed cells must show no growth on LB plates containing 50 µg/ml kanamycin, indicating the absence of any kanamycin resistance markers.

Untransformed cells must show no growth on LB plates containing 15 µg/ml tetracycline, indicating the absence of any tetracycline resistance markers.

Untransformed cells must show no growth on LB plates containing 15 µg/ml chloramphenicol, indicating the absence of any chloramphenicol resistance markers.

Untransformed cells must show no growth on LB plates containing 50 µg/ml Zeocin™, indicating the absence of any Zeocin™ resistance markers.

Untransformed cells must show growth of no more than 5 colonies on LB plates containing 100 µg/ml spectinomycin, indicating the absence of any spectinomycin resistance markers and a low rate of

spontaneous mutation.

Untransformed cells must exhibit growth on LB plates containing 25 µg/ml streptomycin, indicating the presence of streptomycin resistance markers.

### **Absence of Bacteriophage**

To verify the absence of phage contamination, 0.5-1.0 ml of TOP10 competent cells are added to LB top agar and poured over LB plates. After overnight incubation at 37°C, no plaques should be detected.

### **One Shot® OmniMAX™2-T1:**

#### **Transformation Efficiency**

50 µl of competent cells are transformed with 10 pg of supercoiled pUC19 plasmid DNA (non-saturating conditions). Test transformations are performed on a minimum of 6 vials per lot. Transformed cultures are plated on LB plates containing 100 µg/ml ampicillin and incubated overnight at 37°C.

The average transformation efficiency must be greater than  $5.0 \times 10^9$  cfu/µg pUC19.

#### **Antibiotic Sensitivity**

Cells must exhibit growth on LB medium plates.

Untransformed cells must show no growth on LB plates containing 100 µg/ml ampicillin, indicating the absence of any ampicillin resistance markers.

Untransformed cells must show no growth on LB plates containing 50 µg/ml kanamycin, indicating the absence of any kanamycin resistance markers.

Untransformed cells must show no growth on LB plates containing 15 µg/ml chloramphenicol, indicating the absence of any chloramphenicol resistance markers.

Untransformed cells must show no growth on LB plates containing 50 µg/ml Zeocin™, indicating the absence of any Zeocin™ resistance markers.

Untransformed cells must show no growth on LB plates containing 100 µg/ml spectinomycin, indicating the absence of any spectinomycin resistance markers.

Untransformed cells must show growth of no more than 5 colonies on LB plates containing 100 µg/ml streptomycin, indicating the absence of streptomycin resistance markers and a low rate of spontaneous mutation.

Untransformed cells must exhibit growth on LB plates containing 15 µg/ml tetracycline, indicating the presence of tetracycline resistance markers.

## **Growth On Minimal Media**

Cells must exhibit growth on 2B minimal medium plates, indicating the absence of any auxotrophic markers.

## **Lac Phenotype**

Untransformed cells must exhibit growth of white colonies on LB plates containing 400 µg/ml X-Gal and 1 mM IPTG, indicating a Lac<sup>-</sup> phenotype.

## **Gal Phenotype**

Cells must exhibit growth of bright red colonies on MacConkey galactose plates, indicating a Gal<sup>+</sup> phenotype.

## **RecA Phenotype**

Cells must exhibit inhibited growth on LB medium plates containing 8 µg/ml nitrofurantoin, indicating a RecA<sup>-</sup> phenotype.

## **T1 Phage Resistance**

Ten individual colonies of the OmniMAX™2-T1R strain are exposed to T5 phage, along with 2 colonies of a T1/T5-sensitive strain. After overnight incubation at 37°C, the OmniMAX™2-T1R cells should exhibit normal growth, and the sensitive strain should show no growth.

## **Absence of Bacteriophage**

To verify the absence of phage contamination, 0.5-1.0 ml of OmniMAX™2-T1R competent cells are added to LB top agar and poured over LB plates. After overnight incubation at 37°C, no plaques should be detected.

## **One Shot® Stbl3™ Chemically Competent *E. coli*:**

### **Transformation Efficiency**

50 µl of competent cells are transformed with 10 pg of supercoiled pUC19 plasmid DNA (non-saturating conditions). Test transformations are performed on a minimum of 3 vials per lot. Transformed cultures are plated on LB plates containing 100 µg/ml ampicillin and incubated overnight at 37°C.

Transformation efficiency must be greater than  $1.0 \times 10^8$  cfu/µg pUC19.

## **Antibiotic Sensitivity**

Cells must exhibit growth on LB medium plates.

Untransformed cells must show no growth on LB plates containing 100 µg/ml ampicillin, indicating the absence of any ampicillin resistance markers.

Untransformed cells must show no growth on LB plates containing 50 µg/ml kanamycin, indicating the absence of any kanamycin resistance markers.

Untransformed cells must show no growth on LB plates containing 15 µg/ml chloramphenicol, indicating the absence of any chloramphenicol resistance markers.

Untransformed cells must show no growth on LB plates containing 50 µg/ml Zeocin™, indicating the absence of any Zeocin™ resistance markers.

Untransformed cells must show no growth on LB plates containing 15 µg/ml tetracycline, indicating the absence of any tetracycline resistance markers.

Untransformed cells must exhibit growth on LB plates containing 100 µg/ml streptomycin, indicating the presence of streptomycin resistance markers.

## **Leu and Pro Phenotypes**

Cells must exhibit growth on 2B minimal medium plates supplemented with 30 µg/ml leucine and 30 µg/ml proline after overnight growth at 37°C. Cells should show inhibited growth on 2B minimal medium plates, 2B minimal plates supplemented only with leucine, or 2B plates supplemented only with proline under the same incubation conditions. This indicates both Leu<sup>-</sup> and Pro<sup>-</sup> phenotypes.

## **Gal Phenotype**

Cells must exhibit growth of white or light pink colonies on MacConkey galactose plates, indicating a Gal<sup>-</sup> phenotype.

## **RecA Phenotype**

Cells must exhibit inhibited growth on LB plates containing 8 µg/ml nitrofurantoin, indicating a RecA<sup>-</sup> phenotype.

## **Absence of Bacteriophage**

To verify the absence of phage contamination, 0.5-1.0 ml of Stbl3™ competent cells are added to LB top agar and poured over LB plates. After overnight incubation at 37°C, no plaques should be detected.

## **One Shot® Mach1™-T1R Chemically Competent *E. coli*:**

### **Transformation Efficiency**

50 µl of competent cells are transformed with 10 pg of supercoiled pUC19 plasmid DNA (non-saturating conditions). Test transformations are performed on a minimum of 3 vials per lot. Transformed cultures are plated on LB plates containing 100 µg/ml ampicillin and incubated overnight at 37°C.

Transformation efficiency must be greater than  $1.0 \times 10^9$  cfu/µg pUC19.

### **Antibiotic Sensitivity**

Cells must exhibit growth on LB medium plates.

Untransformed cells must show no growth on LB plates containing 100 µg/ml ampicillin, indicating the absence of any ampicillin resistance markers.

Untransformed cells must show no growth on LB plates containing 50 µg/ml kanamycin, indicating the absence of any kanamycin resistance markers.

Untransformed cells must show no growth on LB plates containing 15 µg/ml chloramphenicol, indicating the absence of any chloramphenicol resistance markers.

Untransformed cells must show no growth on LB plates containing 50 µg/ml Zeocin™, indicating the absence of any Zeocin™ resistance markers.

Untransformed cells must show no growth on LB plates containing 100 µg/ml streptomycin, indicating the absence of streptomycin resistance markers.

Untransformed cells must show no growth on LB plates containing 100 µg/ml spectinomycin, indicating the absence of spectinomycin resistance markers.

Untransformed cells must show no growth on LB plates containing 15 µg/ml tetracycline, indicating the absence of tetracycline resistance markers.

### **Growth On Minimal Media**

Cells must exhibit growth on 2B minimal medium plates, indicating the absence of any auxotrophic markers.

### **Lac Phenotype**

Untransformed cells must exhibit growth of white colonies on LB plates containing 400 µg/ml X-Gal and 1 mM IPTG, indicating a Lac<sup>-</sup> phenotype.

## Gal Phenotype

Cells must exhibit growth of bright red colonies on MacConkey galactose plates, indicating a Gal<sup>+</sup> phenotype.

## RecA Phenotype

Cells must exhibit inhibited growth on LB medium plates containing 8 µg/ml nitrofurantoin, indicating a RecA<sup>-</sup> phenotype.

## T1 Phage Resistance

Ten individual colonies of the Mach1™-T1R strain are exposed to T5 phage, along with 2 colonies of a T1/T5-sensitive strain. After overnight incubation at 37°C, the Mach1™-T1R cells should exhibit normal growth, and the sensitive strain should show no growth.

## Absence of Bacteriophage

To verify the absence of phage contamination, 0.5-1.0 ml of Mach1™-T1R competent cells are added to LB top agar and poured over LB plates. After overnight incubation at 37°C, no plaques should be detected.

## Results

All products meet all specifications.

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For Research Use Only. Not for use in diagnostic procedures.

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