High Efficiency Transformation

New England Biolabs[1]

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Sep 08, 2022

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ABSTRACT

Protocol for high efficiency heat shock transformation of competent *E. coli* cells. Source: New England Biolabs

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MATERIALS TEXT

SOC Outgrowth Medium - 100 ml (New England Biolabs Catalog #B9020S)

1 Thaw a tube of competent E. coli cells on ice for **00:10:00** . Mix gently and carefully pipette **50 µL** of cells into a transformation tube on ice. 10m

2 Add **1 µL** containing **100 ng** of plasmid DNA to the cell mixture. Carefully flick the tube 4-5 times to mix cells and DNA. Do not vortex.

3 Place the mixture on ice for **00:30:00** . Do not mix. 30m

4 Heat shock at exactly **42 °C** for exactly **00:00:30** . Do not mix. 30s

5 Place on ice for **00:05:00** . Do not mix. 5m

6 Pipette **950 µL** of room temperature SOC into the mixture.

7 Place at **37 °C** for **01:00:00** . Shake vigorously (**250 rpm**) or rotate. 1h

8 Warm selection plates to **37 °C** .

9 Mix the cells thoroughly by flicking the tube and inverting, then perform several 10-fold serial dilutions in SOC.

10 Spread 50-100 µl of each dilution onto a selection plate and incubate overnight at **37 °C** . 1d
Alternatively, incubate at **30 °C** for **24:00:00** or **25 °C** for 48 hours.

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