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TSS transformation of non-competent *E. coli* cells

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Protocol status: Working
We use this protocol and it's working

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Abstract

Transformation & Storage Solution (2X TSS) enables researchers to take advantage of the simple system described by Chung et al., 1989 (DOI:10.1073/pnas.86.7.2172) for the transformation of *E. coli* cells. Early log-phase cells are suspended in 1X TSS, a solution containing polyethylene glycol, dimethyl sulfoxide, and divalent cations in a bacterial growth medium.
This protocol is an alternative to the transformation method with chemically $CaCl_2$-competent cells.

Advantage: You can use any fresh *E. coli* cells
Disadvantage: Low efficiency. Use only for transformation of plasmids, not for ligation mixtures in molecular cloning. It is possible to transform at least two plasmids at the same time.
$CaCl_2$-Transformation does not work for *E. coli* strains W3110 and W3110 Z1, thus TSS transformation is a method of choice.

Materials

2 x TSS (Transformation and Storage Solution):

- 20 g PEG (3350 or 8000) 20% (w/v)
- 10 mL DMSO, 10% (v/v)
- 2,03 g $MgCl_2$ * $6H_2O$ 100 mM
- ad 100mL LB medium (→ pH 6.5)

- prepare solution without DMSO and autoclave
- add DMSO
- make 10 ml aliquots and store at 4 °C

Transformation

1 Inoculate 3 ml LB-medium with colonies from a fresh agar plate.

2 Incubate at 37 °C and 230 rpm for 1.5 to 2.0 h (exponential growth). Culture should become turbid.

02:00:00

3 Meanwhile prepare 1.5 ml tubes with 200 µl of 2x TSS-buffer and keep on ice.
Add 200 µl cells and 0.5 - 1.0 µl Plasmid. Vortex and incubate on ice for 20 - 30 min.
Longer incubation time will reduce transformation efficiency!

00:20:00

4 Incubate 45-60 min at 37 °C on the Thermomixer with shaking (900 rpm). Addition of LB is not required.

01:00:00

5 Plate on agar plates containing the appropriate antibiotic