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TSS Transformation (after Chung et al., 1989, PNAS 86:2172-2175)

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¹ Chung, C.T. et al. , (1989) Proc. Natl. Acad. Sci. USA 86, 2172.

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

ABSTRACT

Transformation & Storage Solution (2X TSS) enables researchers to take advantage of the simple system described by Chung et al. ¹ for the preparation, long-term storage and transformation of competent E. coli. Early log-phase cells are suspended in 1X TSS: a solution containing polyethylene glycol, dimethyl sulfoxide, and divalent cations in a bacterial growth medium.

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Preparing TSS

- 1 2 x TSS (Transformation and Storage Solution):
 - 20 g PEG (3350 or 8000) 20% (w/v)
 - 10 mL DMSO (after autoclave) 10% (v/v)
 - 2,03 g $\text{MgCl}_2 \cdot 6\text{H}_2\text{O}$ 100 mM
 - ad 100mL LB medium ($\rightarrow$ pH 6.5)

Transformation

- 2
 - Inoculate starter culture of 1.5 mL LB medium
 - After 1.5 – 2 hours culture should become turbid ($\rightarrow$ exponential growth)
- 3
 - Add 200 μL of culture to 1.5 mL tube and mix with 200 μL 2xTSS (work on ice)
 - Add 0.5 – 1.0 μL plasmid (high copy: rather 0.3 μL)

Incubation

- 4
 - Incubate on ice for 20-30 min
 - Incubate 45-60 min at 37°C on shaking device (> 200 rpm) – addition of LB is not requested
 - Plate on LB-Agar (incl. appropriate antibiotic)