

Jun 08, 2018

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

DOI

<https://dx.doi.org/10.17504/protocols.io.qtndwme>

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**External link:** <http://www.epibio.com/docs/default-source/protocols/transformation-storage-solution-%282x-tss%29.pdf?sfvrsn=6>

**Protocol Citation:** Lutz Berwanger 2018. TSS Transformation (after Chung et al., 1989, PNAS 86:2172-2175). **protocols.io**  
<https://dx.doi.org/10.17504/protocols.io.qtndwme>



## Manuscript citation:

<sup>1</sup> 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**

**Created:** June 08, 2018

**Last Modified:** June 08, 2018

**Protocol Integer ID:** 12878

**Keywords:** tss transformation, 1x tss, 2x tss, bacterial growth medium, phase cell, transformation, storage solution, divalent cation, cell

## Abstract

Transformation & Storage Solution (2X TSS) enables researchers to take advantage of the simple system described by Chung et al. <sup>1</sup> 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.



## 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  $\mu\text{L}$  of culture to 1.5 mL tube and mix with 200  $\mu\text{L}$  2xTSS (work on ice)
  - Add 0.5 – 1.0  $\mu\text{L}$  plasmid (high copy: rather 0.3  $\mu\text{L}$ )

## 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)