Product Data Sheet

Rapid Transformation Kit

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Product Description

The solutions provided in this kit allow the researcher to take advantage of the quick and easy transformation system described by Chung *et al*.[1] This protocol allows for the transformation of plasmid DNA into any bacterial strain of choice without needing to purchase competent cells or prepare competent cells according to the lengthy hexaminecobalt chloride/DHMO or calcium chloride procedures as described in Sambrook *et al*.[2] and other molecular biology handbooks.

Bacterial colonies propagated overnight on TSA or other plate medium are resuspended in LB medium and then in 2x TSS to a final 1x concentration. TSS contains polyethelyene glycol, dimethyl sulfoxide and divalent cations, which "condition" the cells, rendering them competent. Once treated with TSS, cells are incubated on ice, heat-shocked and incubated in growth medium, as performed in most standard transformation protocols. Transformation efficiences range from 10^6 - 10^8 colonies/µg DNA. (Precise transformation efficiencies will depend on both the host strain as well as the nature and quality of the transforming DNA.) However, the intent of the kit is to transfer plasmid DNA into other bacterial strains easily without the need for lengthy host cell preparation protocols (e.g., when performing a strain screen with a recombinant protein construct of interest).

Instructions for Use

1. **Materials:**
   - 1.1. 2x TSS (DO NOT store or use near flame - DMSO is flammable).
   - 1.2. *E. coli* strains to be transformed.
   - 1.3. Tryptic Soy Agar (TSA) plates or comparable non-antibiotic containing plate medium.
   - 1.4. Sterile micro(centri)fuge tubes, 1.5mL (or other convenient tube).
   - 1.5. LB medium.
   - 1.6. SOC medium.
   - 1.7. Antibiotic-containing plate medium.

2. **Methods:**
   - 2.1 Streak strain(s) on non-antibiotic plate medium and incubate at 37ºC overnight. (This protocol MUST be performed with fresh, overnight bacterial colonies.)
   - 2.2. Dispense 0.1 mL LB medium into microfuge tubes using sterile technique.
   - 2.3. Pick 4 colonies using a 1µL loop or sterile toothpick and resuspend in the 0.1 mL LB using sterile technique.
   - 2.4. Add 0.1 mL 2x TSS (DO NOT FLAME) and mix well.
   - 2.5. Incubate on ice for 15 min. Once chilled, do not allow the cells to warm above 14ºC.

Kit Components

Provided in Kit
2x TSS
SOC medium
Bacterial Strains
Materials to be Supplied by User
TSA (Tryptic Soy Agar) or LB agar plates (for initial strain propagation)
TYE (Tryptone Yeast Extract), LB or other plate medium containing antibiotics (for selection of transformants)

Storage

2x TSS & SOC medium	**Store at 4°C** Can be stored at Room Temperature during periods of frequent use. DO NOT STORE OR USE 2X TSS NEAR A FLAME
Bacterial Stabs	**Initial Storage at 4°C** *Strains should eventually be propagated and archived as frozen glycerol stocks at -80°C.

Material Safety Data

FOR RESEARCH USE ONLY. NOT INTENDED OR APPROVED FOR HUMAN, DIAGNOSTICS OR VETERINARY USE. Do not ingest, swallow or inhale. Do not get in eyes, on skin, or on clothing. Wash thoroughly after handling. For complete safety information see full Material Safety Data Sheet.

2.6. Add 100 ng plasmid DNA and incubate on ice for 20 min.

2.7. Heat shock in a 42°C water bath for 1 min.

2.8. Add 1 mL SOC medium using sterile technique.

2.9. Incubate at 37°C for 30 min.

2.10 Plate 0.1 mL of transformation mix on antibiotic-containing plate medium and incubate overnight at 37°C for 30 min.

2.11. Streak purify 2 or 3 colonies on appropriate antibiotic plates.

2.12. Prepare master cell bank(s) or cryostock(s) of new strain(s).

References

1. Chung, C.T. *et al*. (1989) *Proc. Natl. Acad. Sci.* USA 86, 2172.
2. Sambrook, Fritsch, Maniatis (1989) *Molecular Cloning: A Laboratory Manual, Second ed*., Cold Spring Harbor Press: Plainview, NY, pp. 1.76-1.84.

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