

Reagents:

- 5X KCM buffer (I have a stock and can aliquot you 1ml)
 - 5.1 mL 1M KCl
 - 1.5 mL 1M CaCl₂
 - 2.5 mL 1M MgCl₂
 - 0.9 mL ddH₂O
- DNA
- TOP10 competent cells (1 tube has 100ul, I use 30ul per reaction)
- ice
- 42C water bath
- SOC media (or LB)

Protocol

1. Aliquot 30ul into a new eppendorf/culture tube.
2. Add 6ul of 5X KCM buffer to the cells.
3. Add at least 10ng of plasmid DNA to the cell-buffer mixture.
4. Incubate on ice for 20 minutes.
5. Heat shock for 45 seconds.
6. Incubate on ice for 2 minutes.
7. Add 200ul SOC/LB and shake at 37C for 30-60min.
8. Plate cells on respective antibiotic plates.
9. Incubate overnight at 37C.