

Chemical competent Transformation

Aim of the experiment

This protocol can be used to transform chemical competent cells with DNA from various sources such as Ligation, Gibson Assembly or pure plasmid.

Materials

- Chemical competent cells (various strains, prepared according to protocol)
- SOC Media, see corresponding protocol
- LB-Plates with corresponding antibiotics
- Heat block set to 42 °C
- DNA(various source)
- 1.5 ml Eppendorf tube

Procedure

All steps must be done on ice!

1. Thaw cells on ice for 10 min.
2. Add 1 μ l of DNA.
3. Wait 1-30 min, note that the transformation efficiency will increase by 2 for every 10 min of incubation.
4. Heat-shock 30 s at 42 °C.
5. Regenerate on ice for 2 min.
6. Add 950 μ l SOC Media.
7. Incubate at 37 °C and 250 rpm for 1 h.
8. Spin down for 30 s, discard 900 μ l and transfer the cells onto the plate by resuspension in the remaining media.

9. Spread the cells with plating beads by shaking for at least 10 s.
10. Incubate plates overnight at 37 °C.