Transformation

Chemical transformation

1. Thaw cells (50 µL aliquots) on ice for 10 minutes.
2. Add 1-5 µL of plasmid DNA (1 pg-100 ng) to cells and mix without vortexing.
3. Place on ice for 30 minutes.
4. Heat shock at 42°C for 30 seconds.
5. Place cells on ice for 5 minutes.
6. Add 950 ul of room temperature SOC medium.
7. Incubate the cells at 37 °C for 60 minutes, shake vigorously or rotate.
8. Harvest the cells by centrifugation at 3000x g for 1 min.
9. Decant the supernatant and resuspend the pellet in the remaining medium.
10. Spread 50-100 µl onto pre-warmed selection plates and incubate at 37°C.

Electrocompetent transformation

1. Thaw cells (50 µL aliquots) on ice for 10 minutes.
2. Place electroporation cuvettes on ice.
3. Add 1-5 µL of plasmid DNA (1 pg-100 ng) to cells and mix without vortexing.
4. Transfer the cell/DNA mix into a cold cuvette without and make sure that the cells deposit across the bottom of the cuvette.
5. Electroporate using the following conditions
    a. 2500 V for *Escherichia coli* DH5α
    b. 400 V for *Vibrio natriegens*
6. Immediately add 950 µl of 37°C SOC (*E. coli*)/BHIN (*V. natriegens*) to the cuvette, gently mix up and down, then transfer to microcentrifuge tubes.
7. Incubate the cells at 37 °C for 60 minutes, shake vigorously or rotate.
8. Harvest the cells by centrifugation at 3000x g for 1 min.
9. Decant the supernatant and resuspend the pellet in the remaining medium.
10. Spread 50-100 µl onto pre-warmed selection plates and incubate at 37°C.