

Transformations

Long (best for BL21(DE3) & Rosetta)

1. Thaw cells from -80° freezer on ice (each tube has enough for 4 50ul transformations).
2. Add 1ul of DNA + 50ul of cells to a 1.5ml tube.
3. Mix gently by tapping tube a few times and leave on ice for 30 min.
4. Put in 37°C water bath for 2 min.
5. Put back on ice for 5 min.
6. Add 900ul of LB and incubate for 30min – 1hr at 37°C (either in a water bath or put in a flask and shake in the shaker).
7. Spin down tube (3 min @4k rpm) and pour off all but ~100ul of super, or add 100ul of fresh LB back to the tube. Gently resuspend pellet by pipetting up and down only as many times as needed.
8. Plate cells on LB+appropriate antibiotic plate and incubate at 37°C overnight.

Fast (fine for HMS-174(DE3) cells)

1. Thaw cells from -80° freezer on ice (each tube has enough for 4 50ul transformations).
2. Add 1ul of DNA + 50ul of cells to a 1.5ml tube.
3. Mix gently by tapping tube a few times and leave on ice for 2min
4. Put in 37°C water bath for 3 min.
5. Put back on ice for 5 min.
6. Add 150ul LB and incubate for 20min at 37°C
7. Plate cells on LB+appropriate antibiotic plate and incubate at 37°C overnight.