E. coli plasmid Transformation

Before starting: Book heating block @42ºC

Get competent cells from -80ºC freezer

1. Thaw cells on ice
2. Pipette 2 µL of solution, containing plasmid, into the cell stocks. Mix gently by pipetting up and down.
3. Leave the tubes on ice for 30 min.
    - **OBS! Don't mix/shake the tubes after, since that will disperse the DNA again.**
    - **Put rack in fridge to prevent the ice from melting.**
4. Put the tubes in the heating block @42ºC for 90 s.
    - **OBS! If you have many tubes, be sure to remove them from the heating block in the same order that you put them in.**
5. Take the tubes out of the heating block and put them on ice.
6. Pipette 500 µL LB-medium into each tube.
    - **OBS! Use a sterile fume hood. Make sure to wipe the floor of the hood with ethanol and do the same for everything you put in the hood (like pipette, LB-solution and your hands).**
    - **OBS! Pipette carefully, to avoid splash.**
7. Incubate @37ºC for 1h.
    - **TIP: Take the plates, out of the fridge, and put them in 30ºC to remove moisture, making it easier to write on them.**
8. Transfer 10% (50-100 µL) of the solution to the plastes marked 10% (LB/Amp plates)
    - **OBS! Use sterile technique.**
9. Spin down the solution into a pellet.
10. Resuspend the pellet in ~100 µL MQ.
11. Plate the resuspended cells on the plates marked 90% (LB/Amp plates)
12. Incubate the plates @37ºC for 16h (overnight).