

Making electrocompetent cells and electroporation

Materials

- LB medium

Making electrocompetent cells

1. Scrape cells off a plate into 1ml cold water, spin down and resuspend the cells in 50 ul water.

Transformation of electrocompetent cells

2. Add 40ul of electrocompetent cells to 50ng miniprep plasmid. Place on ice for 15min. Don't forget the control without DNA.
3. Add cells in an electroporation cuvette (2mm), don't insert bubbles and keep on ice as much as possible. Ensure the liquid level by tapping.
4. Electroporate at 200 Ω and 2,5 kV. Be sure the metal touch each other. Hold both buttons until you hear the bleep.
5. Immediately add 500ul LB medium (as quick as possible after the 'bleep') to the cells and transfer to a test tube.
6. Incubate horizontally 1hr, 37°C in a shaker
7. Plate on selective media