

Electroporation

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Abstract

Protocol for E. coli transformation with electrocompetent cells.

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Protocol

Step 1.

Add 100 ng of plasmid into 40 uL competent cells in a 1.5 uL eppendorf tube. Mix gently by flicking the tube a couple of times.

Step 2.

Transfer mixture to electroporation cuvettes. Tap the cuvette e.g. on a table to make sure that all the mixture is on the bottom.

Step 3.

Carry out electroporation; set kV to 2.2, low range to 200 and the capacitance to 25. A read of at least 5 milliseconds is required.

Step 4.

Add 450 uL of SOC or SOB medium immediately after the electroporation, mix by pipetting up and down.

Step 5.

Transfer mixture to 1.5 mL eppendorf tubes and outgrow for 1 hour at 37 C with shaking.

Step 6.

Plate appropriate amounts (20-200 uL) of the mixture on LB plates containing the correct antibiotic. (Remaining unplated transformation mixute can be stored on the bench and plated the next day if necessary.)