



Transformation of E. coli with plasmid

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This protocol can be used to transform electrocompetent *E. coli* cells with a plasmid.

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Preparation 30m

Wash cuvette with [M]100 % (v/v) ethanol and UV in the hood for ⊙ 00:20:00

20m

2 Pre-thaw electrocompetent cells and plasmid on wet ice for © 00:10:00

10m

Electroporation 20m 40s

3 Add $\Box 1 \mu L$ of plasmid DNA to $\Box 50 \mu L$ electrocompetent cells

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- 4 Transfer cell-DNA mixture into the cuvette and add equal amount (50 μL) of MilliQ

 5 Let the cuvette sit in wet ice for © 00:20:00

 6 Place cuvette in BioRad pulser and pulse at 2.5 keV

 7 Within © 00:00:40 add 100 μL of 2X LB to the cuvette

 40s

 Incubation 30m

 8 Place cuvette in an incubator at 8 37 °C for © 00:30:00

 9 Then use a bent pipette tip to spread the cuvette's contents onto a plate made with LB agar and the appropriate antibiotics. Spread until plate is dry
 - 11 Make glycerol stocks of transformed cells and store for future use

Place the plate in an incubator at § 37 °C © Overnight

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