Bacterial transformation

Stephen Floor

Abstract

Standard protocol to transform bacteria with a plasmid using chemically competent E. coli and antibiotic resistance.

Citation: Stephen Floor Bacterial transformation. **protocols.io**
dx.doi.org/10.17504/protocols.io.mnvc5e6
Published: 16 Jan 2018

Materials

✓ MACH1 or DH5a or TOP10 or NEB stable or etc for DNA purification/miniprep by Contributed by users

✓ BL21(DE3) or BL21-Star(DE3) or Rosetta2(DE3) or etc for protein purification by Contributed by users

Protocol

Step 1.
All steps in a microcentrifuge tube.

Step 2.
Thaw cells on ice.

Step 3.
Incubate 10-500 ng DNA with cells (10 to 50 µL) on ice for 25 minutes in a microcentrifuge tube.

Step 4.
Heat shock at 42 degrees for one minute in water bath.

Step 5.
Recover for two minutes on ice.

Step 6.
Add 180 µL LB or SOC media, mix.

Step 7.
Grow at 37 degrees for one hour with shaking. Warm the plates during this step.

Step 8.
Plate 50 to 200 µL of the transformation on plate with appropriate antibiotic.

Step 9.
Leave at 37 degrees overnight or room temperature for two days.