

This protocol was adapted from an online protocol by "MFT," as well as advice from Marc Ammerlaan.
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⊘⊘⊘⊘ **Heat Shock Protocol**

Transformation of competent cells can be achieved using a fairly straightforward process. This process is between QT and Electroporation in terms of complexity and efficiency.

Procedure *I* will transform the bacteria; after a 20 minute recover stage, procedure *II* will plate them.

⇒ Competent cells should be made in accordance with the **Production of QT competent cells** protocol.

Compounds

Competent cell solution, thawed
Plasmid solution

Materials

LB plates (labeled, with antibiotic)
1.5mL centrifuge tubes
5µL pipette, tips (chilled to 4°C)
200µL pipette, tips (chilled to 4°C)
1000µL pipette, tips (chilled to 4°C)
Sterile glass spreader
ICE

You will also need access to an:

Water bath at 42°C

External protocols:

Production of QT competent cells

Procedure I

1. Add **50ng plasmid solution** to a **1.5mL centrifuge tube** with a **5µL pipette**. Keep the tube on **ICE**.

⇒ Calculate the volume of DNA solution necessary to generate 50ng ahead of time. Make sure to label the plates.

2. Add **200µL competent cell solution** to the tube with a fresh, chilled, sterile **1000µL pipette**. Mix the solutions by repeatedly uptaking and expelling it from your pipette about 2-3 times.
3. Chill the centrifuge tubes for **5 minutes** on ICE.
4. Place the tubes in the **water bath** at **42°C** for **45 seconds**.
5. Place the tubes back on ice for **20 minutes**. This will limit damage to the cells.

Procedure II

1. Using **careful sterile technique**, pipette **100µL** of the contents of the centrifuge tube onto the center of an **antibiotic LB plate** with a sterile **200µL pipette**.
2. Carefully lift the lid just high enough to fit in a **sterile glass spreader**. Spread the solution evenly across the plate using gentle linear motions, as if drawing an asterisk (*).
3. **Incubate** the plates for **16 hours** at **37°C** or until colonies are observed.