

Chemical transformation (heat shock)

1. Pre-heat a bath to 42°C

2. Defrost 100 µl of competent cells for 10-15 minutes on ice. If the Eppendorf contain more than 100 µl competent cells divide them into a 100 µl each. (Each competent eppendorf contains 220 µl.)

- for circular plasmids : Add 1-2 ng plasmid to the competent cells
- If the plasmid is after ligation: Add 5 µl from ligation reaction to the competent cells.
- If you use Biobrick add 2 µl.

3. Put for 30min on ice. During this time make SOC and pre-heat it to 37°C (in shaker)
(2-stage, 14 min) → מכלי המכונה הם אלה המיועדים.

4. 1 minute heat shock at 42 °C.

5. 2 minutes on ice.

6. Add 900 µl SOC and shake for 1 hour recovery at 37 degrees.

7. Warm plates in 37 °C incubator

7. Plate the cells on appropriate antibiotic plates (100 µl and "rest").

Rest = transfer the rest of the volume to eppendorf tube and centrifuge for 1 min, max speed. Discard most of the supernatant, pipette the rest and plate.

8. Incubate overnight in 37°C incubator.
הנותרת מהתערובת = Rest

