

## Chemical Transformations

1. Thaw frozen cells on ice for ~10-15 min.  
(**Note:** Leaving these on ice longer does not appear to affect transformation efficiency)
2. Label 1.5 mL Eppendorf tubes
3. Aliquot 1-2  $\mu\text{L}$  of DNA plasmid to corresponding Eppendorf tube
  - o 2  $\mu\text{L}$  for uncoiled DNA (i.e. re-suspended parts and ligation products)
  - o 1  $\mu\text{L}$  for supercoiled DNA (i.e. miniprep products)
4. Add 40  $\mu\text{L}$  competent cells to DNA aliquots  
(**Technique:** Aspirate (draw & release) w/ pipet several times to mix)
5. Incubate on ice for ~30 min.  
(**Note:** Leaving these on ice longer does not appear to affect transformation efficiency)
6. Heat shock cells in water bath for ~60 s @ 42° C  
(**Note:** ~10 min. @ 37° C also works well)
7. Incubate on ice for ~5 min.
8. Add 200  $\mu\text{L}$  SOC and put in shaker for ~2 hr. @ 37° C  
(**Note:** If short on time, 1 hr. recovery is sufficient)
9. Distribute transformed cells uniformly over appropriate LB plates
  - o ~100  $\mu\text{L}$  for uncoiled DNA
  - o ~25  $\mu\text{L}$  for supercoiled DNA
10. Once dry, turn upside down and incubate overnight @ 37° C  
(**Note:** Over weekend @ R.T. provides similar results)