# Yeast Transformation

## Low Efficiency (suitable for episomal plasmid)

- grow yeast on YPD plate overnight
- resuspend in buffer
  - 100ul of buffer100ul **2M Lithium Acetate** (LiAc)
  - 100ul **1M DDT**
  - 800ul **50%PEG** (3350 or 4000)
  - 50ul **ssDNA** (10mg/ml)
- add DNA (50ng+)
- vortex
- heat shock 42C 14min
- plate on selective media and wait

## High Efficiency (more than one plasmid, integrative plasmids, endogenous tagging)

**Competent cells preparation**:

- grow yeast in 5ml YPD overnight
- dilute in 30ml to 0.1-0.2 OD
- grow to 0.5-0.8 OD ~3h
- spin cells (3000rpm 5min), wash **ddW** (~20ml)
- spin cells, wash with **0.1M LiAc**
- spin cells, discard (there will be some LiAc left for ~5 transformations
- keep in +4C up to 1week

**Transformation:**

- prepare mix:
  - 480ul **PEG50%**
  - 100ul competent cells
  - 72ul **1M LiAc**
  - 40ul **ssDNA** (10mg/ml)
  - DNA (plasmid/PCR product...)
- vortex
- 30min-1h 30C without rotation
- add 72ul **DMSO**, vortex
- heatshock 42C 14min
- spin cells, discard, resuspend in 100ul of water and plate on selective media, if you're working with antibiotic resistance markers plate on YPD, next day replate on selective media