

High-efficiency transformation of yeast with a plasmid library***

1. Inoculate 100 mL YPD with an overnight culture of cells so that the final concentration is 0.3 OD₆₀₀/mL
*it is OK to inoculate YPD with cells grown in selective medium
2. Spin out cells; resuspend pellet in 10 mL sterile water
3. Spin out cells; resuspend pellet in 2 mL 0.1M LiOAc-TE
4. Incubate at room temperature for 10 min
5. Add 0.2 mL salmon sperm DNA, 50 µg plasmid library DNA; vortex
6. Add 14 mL 40% PEG/0.1M LiOAc-TE; vortex
7. Incubate at room temperature for 30 min
8. Transfer cells to 1-L flask
9. Add 1.8 mL DMSO and immediately heat shock at 42°C in a water bath shaking at ~150 rpm
10. Cool cells to room temperature in an ice bucket filled with water
11. Spin out cells; resuspend pellet in sterile TE
*use TE to help harvest the cells from the 1-L flask
12. Spin out cells; resuspend pellet in 100 mL YPD and shake for 1 hr
13. Spin out cells; resuspend pellet in 10 mL TE
14. Spin out cells; resuspend pellet in 2 mL TE
15. Spread 0.2 mL aliquots of resuspension on 10 YNB agar plates

***It is usually best to do a small-scale pilot transformation to gauge the number of colonies you can expect, then resuspend the cells in step 14 according to the desired density of colonies when the transformation is scaled up