# High Efficiency Yeast Transformation

1. Start an overnight culture of the yeast cells to be transformed, in YPD.

2. Next morning, reinoculate 5 mls of YPD with 50 ul of O/N culture. Allow cells to grow
   approximately 4 hours, or until in log phase growth.

3. Take 250 ul of culture, pellet the cells at top speed in a microfuge for 5 sec.

4. Resuspend the cells in 1 ml of $dH_2O$, pellet in microfuge. Remove $dH_2O$.

5. Resuspend the cells in 1 ml of 100 mM LiAc and incubate for 10 min at 30˚ C. Pellet the cells in a microfuge.

6. Add the following components into the tube on top of the cell pellet in this order;

   240 µl of PEG (50% w/v)
   36 µl 1.0 M. LiAc
   50 µl SS-DNA (2.0 mg/ml)
   5.0 µl of plasmid or linear DNA
   20 µl of sdd water.

7. Vortex the cell pellet for at least 1 min to resuspend the cell pellet in the transformation mix. Incubate the cells for 30 minutes at 30˚ C. Incubate for 20 minutes at 42˚ C.

8. Pellet the cells at top speed in a microcentrifuge for 10 sec. Remove the supernatant using a micropipet.

9. Gently resuspend the pellet in 100 µl of sdd water by slowly pipetting up and down.

10. Plate the cell suspension onto a plate of omission medium that selects for the presence of the plasmid (or insert). Colonies should be visible in 2 -4 days at 30˚ C.