Quick Yeast Transformation

When just a few transformants are sufficient, such as the transformation of a test plasmid or a GAL4BD fusion plasmid, the following protocol can be used. This procedure is very flexible and can be applied to yeast cells from a number of different sources; however, for best results, use cells from a freshly grown plate.

1. Scrape a 25 µl yeast inoculum for EACH transformation planned from a plate with a sterile toothpick or loop and resuspend in 1 ml of sdd water. (Each Transformation reaction requires about a 25 µl volume of yeast cells, therefore if you scrape about 100 µl volume of yeast cells into 1 ml this represents 4 transformation reactions)**

   TIP:The plate can be up to a week old, yet, fresh inocula are preferred. Yeast cells taken from most types of media plates can be transformed by this method, although cells grown on YPAD perform best. Remove cells from the edge of a cell mass or colony to ensure recovering healthy cells

2. Pellet the cells at top speed in a microfuge for 5 sec.
3. Resuspend the cell pellet in 1 ml of 100 mM LiAc and incubate for 5 min at 30° C.
4. Place the volume representing a single TRANSFORMATION REACTION (or 25 µl volume of yeast cells, If you scraped a 50 µl volume of yeast cells into 1 ml of sterile water then this volume would be 500 µl) into a separate microcentrifuge tube and spin the suspension at top speed in a microcentrifuge for 5 sec. Remove the supernatant with a micropipet. .
5. Add the following components into the tube on top of the cell pellet in this order or as a premix;.

Premix (µl)	1X	3X	5X
Peg (50% w/v)	240	720	1200
LiAc (1.0 M)	36	108	180
SS-DNA (2mg/ml)	50	150	250
H_2O	14	42	70

6. Use 350 µl per tranformation
   5.0 µl of plasmid DNA (100 ng to 5 µg)
7. Vortex the cell pellet for at least 1 min to resuspend the cell pellet in the transformation mix and incubate at 42° C for 20 min.
8. Pellet the cells at top speed in a microcentrifuge for 10 sec. Remove the supernatant using a micropipet. Gently resuspend the pellet in 200 - 400 µl of sdd water by slowly pipetting up and down.

Plate the cell suspension onto 1 or 2 plates of SC ommision medium that selects for the presence of the plasmid. Colonies should be visible in 2 - 4 days at 30^{o} C.

Yield will range from a few hundred to a few thousand transformants.