

Yeast transformation

1. grow cells over night in 3-5 ml YPD.
2. Transfer to a 50 ml YPD medium (need about 2 to 3ml). Grow cells to 1×10^7 cells/ml. (O.D 600=0.5 to 1.0).
3. Spin cells down in a 50 ml tube at 3000rpm for 5 min.
4. Discard the supernatant, resuspend in 1 ml steriled water and transfer to 1.5 ml microtube.
5. Spin cells down, and wash with 1 ml TE/LiAC.
6. Resuspend cells to about 10^9 cells/ml. (about 250 ul to 500ul).
7. Heat carrier DNA (ssDNA) at 95-100°C for about 5 min to melt te DNA strands and store on ice.
8. Mix 1 ug plasmid DNA(1 to 2 ul) with 5 ul ssDNA, and add 50 ul of the yeast cells.
9. Add 300 ul PEG4000 (PEG3350/LiAC/TE) and vortex thoroughly.
10. Incubate at 30°C with agitation for 30 min.
11. Add 35ul DMSO.
12. Heat shock cells at 42°C for 15 min.
13. Spin down and wash with 1 ml water.
14. Resuspend in 100 ul water and plate on the selective CM medium.