## Plasmid transfer from S. cerevisiae to E. coli

- 1. Prepare a 5 ml over night culture of yeast in SD-aa medium
- 2. Centrifuge 1.4 ml of the liquid culture in microtube for 30 seconds
- 3. Decant supernatant and resuspend pellet in remaining drop of liquid
- 4. Add 0.2 ml of 2 % Triton X-100, 1 % SDS, 100 mM NaCl, 10 mM Tris pH 8.0, 1 mM EDTA
- 5. Add 0.2 ml PCIA (Phenol : Chloroform : Isoamyl alcohol = 24:23:1)
- 6. Add 0.3 g of acid washed glass beads
- 7. Disrupt cells on vortex for 2 minutes or in Fast-Prep for 45 seconds
- 8. Centrifuge for 5 minutes in microcentrifuge
- [Optional: Extract again with 0.2 ml PCIA Extract with 0.2 ml CIA Precipitate with 1/10 volume 3 M NaOAc and 2.5 volumes Ethanol Dissolve in 50 µl of TE]
- 9. Transform 5 µl of DNA into E. coli DH5