YEAST TRANSFORMATION PROTOCOL

Adapted by Vinh Pham from previously published protocols.
Last modified: July 29, 2002.

MATERIALS:

Salmon Sperm DNA (10ug/ul): Boil 5', quick cool on ice for 5'.
40% PLT -560ul 50% PEG
-70ul 10X LiOAc (1M LiOAc)
-70ul 10X TE, pH 7.6 (0.1M Tris, 10mM EDTA)
DMSO
TE, pH 7.6

PROCEDURE:

1. Dry selective plates 2-3 days ahead of time.
2. Add 5ug pDNA (less is more) and 5ul salmon sperm DNA to a thawed 150ul aliquot of competent yeast cells. Do *not* use herring sperm DNA.
3. Vortex gently and flick bottom of tube to resuspend settled cells.
4. 30 deg. C for 30 min. on shaker.
5. Add 700ul 40% PLT (make fresh).
6. Vortex gently and flick bottom of tube to resuspend settled cells.
7. 30 deg. C for 1 hr. on shaker.
8. Optional: Add 85ul DMSO and mix by inversion (do not vortex).
9. Heat shock at 42 deg. C for 5 min.
10. Cold shock on ice for 2 min.
11. Pellet at 3Krpm for 1'. Discard supe.
12. Wash 2X with 1ml TE.
13. Pellet at 3Krpm for 1'. Discard supe.
14. Resuspend in 1ml TE.
15. Plate 200ul cells per plate and incubate at 30 deg. C.