

CLEANING UP DIRTY DNA PREPS

(Modified from "current protocols" 2.3.3 by H. Judelson)

1. To DNA add 1 volume of: 2% CTAB, 100 mM Tris 8.0, 20 mM EDTA, 1.4 M NaCl (preferably at 55-65⁰C). Mix by inversion. Incubate at 65⁰C for 10 mm.
2. Add equal volume of 24:1 chloroform: isoamyl alcohol [or octanol]. Invert; vortex briefly (up to 30 sec).
3. Spin 5 min in microfuge to separate phases.
4. Save supernatant, being careful not to remove the interface (if desired to increase the yield, back extract the chloroform-interface with 150 µl of the 2% CTAB solution).
5. Add 1.1 volume of (preferably at 55-65⁰C; this increases the speed of precipitation): 1% CTAB, 50 mM Tris 8.0, 10 mM EDTA. Mix well by inversion. A precipitate should be visible.
6. Spin 5 min, 3k x g. If pellets are seen, discard the supernatant.
7. Add to the pellet 0.5 ml of: 10 mM Tris 8.0, 0.1 mM EDTA, 1 M NaCl. Heat 30 min, 65⁰C to dissolve the pellet. Flick the tube to make sure the pellet is dissolved.
8. Add 0.6 vol. isopropanol (0.3 ml). Invert several times. Spin 8 min in microfuge. Discard supernatant.
9. Add 0.5 ml 70% ethanol, spin 1 minute in microfuge, and discard pellet.
10. Add TE and heat 65⁰C for 30 min.