

Genomic DNA Mini-prep

(Adapted for Fastprep machine)

Solutions

Lysis buffer

- 100 mM Tris pH 8.0
- 50 mM EDTA
- 1% SDS

For 50 ml: 5 ml 1 M Tris, 5 ml 0.5 M EDTA, 5 ml 10% SDS

Procedure

1. Grow 5 ml cells overnight at 30° C.
2. Spin, wash once with 1 ml H₂O.
3. Resuspend in 500 µl lysis buffer.
4. Transfer to a screw-tube with acid washed glass beads.
5. Fastprep at 6.0 speed for 20 min.
6. Recover liquid phase with blue tip into another tube.
7. Add 275 µl 7 M ammonium acetate pH 7.0.
8. Incubate 5 min at 65° C, then 5 min on ice.
9. Add 500 µl chloroform, vortex, spin 2 min in microfuge.
10. Take supernatant and precipitate with 1 ml isopropanol.
11. Incubate 5 min at room temperature, then spin 5 min.
12. Wash pellet with 70% EtOH, dry and dissolve in 50 µl H₂O.

For Southern, digest 5 µl DNA

For PCR, use 0.5-1 µl DNA.