

DNA Isolation from Mouse Tissue

1. Homogenize tissue (~ 0.5g of liver) in 2 ml of isotonic buffer.
2. Add 7mM EDTA to 5 ml and mix gently.
3. Add SDS to 1% and mix gently.
4. Add protenase K to 200 μ g/ml and mix gently.
5. Incubate at 55°C for 1-4 hours.
6. Extract once with phenol, once with phenol:chloroform (1:1), and once with chloroform.
7. Add two volumes of ethanol, swirl gently, and pick out DNA fiber with a pipet tip. NOTE: DNA precipitate will not form if too much ethanol is added.
8. Transfer DNA to an eppendorf tube and wash twice with 70% ethanol. Dry pellet under vacuum.
9. Add 0.5 ml of TE and incubate pellet at 55°C for 1-2 hours. Vortex gently to dissolve pellet completely.
10. Check concentration of by measuring its OD at 260 nm. At 260nm, 1 OD equals 50 μ g of DNA/ml.

Solutions

Isotonic buffer: per 100 ml

0.15 mM NaCl 3 ml of 5M NaCl

0.01 M Tris pH 7.5 1 ml of 1M Tris

1.5 mM MgCl₂ 0.15 ml of 1 M MgCl₂

ddH₂O up to 100 ml

Chen 4/25/88

TAA re-entered 1/24/88