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 proteinase K to 200 µg/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 DNA by measuring its OD at 260 nm. At 260nm, 1 OD equals 50µg 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