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Protocol: DNA Extraction from Mouse Biopsies

Application:

Extraction of DNA for the purposes of genotyping mice. Biopsies generally consist of tail or ear tissue.

Procedure:

1. Obtain biopsy in 1.5-mL screw cap microfuge tube.
2. Add 400 μ L of 50 mM NaOH.
Note: 50 mM NaOH is diluted from 10 N NaOH. In regards to NaOH, 10 N = 10 M.
3. Boil (100 °C) until tissue is fully dissolved (~30-60 minutes).
Note: Intermittent vortexing can speed up dissolution of tissue.
4. Add 50 μ L of 1 M Tris-hydrochloric acid (Tris-HCl), pH 8.0.
5. Vortex and centrifuge at 25 °C, 12,000 x gravity (g) for 2 min.
Note: Unless otherwise noted, all centrifugation steps performed using a benchtop micro centrifuge.
6. Transfer 200 μ L of supernatant to new 1.5-mL microfuge tube.
7. Add 20 μ L of 3 M sodium acetate (NaOAc), pH 5.2.
Note: Volume of NaOAc added is equivalent to 1/10th the sample volume.
8. Add 660 μ L of 95% ethanol (EtOH).
Note: Volume of EtOH added is equivalent to 3x the volume of sample + NaOAc.
9. Vortex and precipitate (PPT) DNA at -20 °C overnight.
Note: DNA can also be PPT at -80 °C for 1 hour in a pre-chilled tube rack.
10. Centrifuge at 4 °C, 12,000 x gravity (g) for 5 min.
11. Aspirate supernatant and resuspend (RSS) DNA pellet in 100 μ L of 0.1x Tris-EDTA (TE) buffer.
Note: 0.1x TE is diluted down from a 10x TE stock solution.