Preparation of genomic DNA for genotyping

Tail Lysis Buffer (100 mM Tris.Cl pH8.0, 5 mM EDTA, 0.2% SDS, 200 mM NaCl)

50 ml	1 M Tris.Cl pH8.0
5 ml	500 mM EDTA pH8.0
10 ml	10% SDS
20 ml	5 M NaCl

Add dH2O to 500 ml
Store at room temperature

1. Obtain a piece of tail (about 5 mm long is enough), put into an Eppendorf tube
   For adult mice, anesthetize the mice before cutting the tail. For embryos, decapitate the embryos before cutting the tail.
2. Add 0.5 ml Tail Lysis Buffer and 5-10 µl of 20 mg/ml Proteinase K
3. Shake at 50-55°C overnight
   Efficient digestion is critical. If the digestion is not complete, it will be difficult to get a compact DNA pellet in later steps. Robust shaking greatly improves the digestion efficiency. To maximize the shaking, place the Eppendorf tubes flat instead of upright on a shaker. It may be helpful to use Eppendorf clamps to prevent the tubes from poping open during the shaking.
4. Microcentrifuge at top speed for 10 min to pellet undigested hair, transfer the supernatant to a new Eppendorf tube. (Omit this step for embryonic tails)
5. Add 0.5 ml isopropanol. Invert tubes a few times.
6. Microcentrifuge at top speed for 10 min at 4°C.
7. Pour off supernatant, wash pellet with 70% ethanol
8. Dry the DNA pellet by Speed Vac (5 min, setting at Medium).
9. Resuspend the DNA pellet in 100 µl of dH2O by pipetting up and down with a P200 pipetman.
10. Use 2 µl of DNA for PCR (total PCR volume: 20 µl).