

DNA from mouse tails

<u>Materials:</u>	Lysis buffer	10 mM Tris/HCl pH8.0 400 mM NaCl 2 mM EDTA 0.1% SDS
	Proteinase K [20 mg/ml] 5 M NaCl Isopropanol 70% Ethanol 10 mM Tris/HCl pH8.0	

Procedure:

- Add 500 µl lysis buffer and 3 µl Proteinase K to tail biopsy (ca. 2 mm)
- Incubate at 50-60°C (we use 58°C) for 3 hours – overnight (or until tail is dissolved) with shaking
- Add 200 µl 5M NaCl, mix well!!
- Spin for 5 minutes (') at full speed (ca. 12-16 000 g) in a microcentrifuge
- Transfer supernatant (ca. 650 µl) to new tube, discard pellet (tissue debris)
- Add 1 volume (ca. 650 µl) isopropanol, mix well (!!) by rigorously inverting the tube several times
- Spin 15-30' at full speed (ca. 12-16 000 g) at room temperature
- Discard supernatant, wash pellet (DNA) by adding 500 µl 70% Ethanol
- Spin 5' at full speed (ca. 12-16 000 g) at 4°C
- Discard supernatant, air-dry pellet at room temperature (or 37°C if it needs to be fast)
- Dissolve DNA in 100 µl 10 mM Tris pH8.0 by pipetting up and down several times
- Use 1 µl of this solution as a template for (genotyping) PCR reaction