**Genomic DNA Isolation**

**DNA Extraction (macro scale)**

1. Homogenize the whole spleen using a mortar and a pestle in 4ml of **1M NH4OH** in **0.2% triton**.
2. Transfer the homogenized liquid into a 15 ml tube. Add 4 l **RNAse A** and incubate the homogenized at 37C for 30 min. Then, add 4 l of 10 g/ l **Proteinase K** and continue incubating for another 30 min.
3. Do 2 times phenol extraction: Add 4 ml **Phenol, pH 6.6**, invert the tube several times, and spin down for 10 min at max speed. Move supernatant to the new tube, avoiding taking the protein interface.
4. Do 2 times phenol/chloroform extraction: Add 2 ml **Phenol, pH 6.6** + 2 ml **CIA**. Invert the tube several times and spin down for 10 min at max speed. Move supernatant to the new tube, avoiding taking the protein interface. At the end, we should barely see the protein interface.
5. Do one time chloroform extraction: Add 4 ml **CIA**, invert the tube several times, and spin down for 10 min at max speed.
6. Move supernatant to the new tube, leaving some behind. Precipitate DNA pellet by adding 10ml **Isopropanol**, and inverting the tube several times.
7. Spin down the pellet at max speed for 1 min. Remove all the liquid with a pipette, avoiding attaching or taking the precipitated DNA strands.
8. Wash the DNA pellet with 10ml **70% EtOH**. Again, remove all the liquid, avoiding attaching the precipitated DNA strand.
9. Dry the pellet in the vacuum dryer. The clean dry DNA pellet should turn clear.
10. Resuspend the pellet in **0.1 X TE**. The pellet can be left to be dissolved in the fridge overnight or incubated at 37C in a water bath for at least an hour. DNA yield extracted from one spleen should be about 100-800 g.