**Genomic DNA from Flies**

1. Collect 50-200 flies on ice. (With fewer than 100 flies, it is difficult to get enough DNA to spool at the end.)

   50 mL

2. Homogenize in 5 ml 0.35 M Sucrose 6 g
   (use 7 ml dounce) 0.1 M EDTA 10 mL of 0.5 M
   50mM Tris pH 8.0 2.5 mL of 1M

   Pipette through wide Nitex into 15 mL polypro tubes (Sarstedt).

3. Spin @ 4000 rpm for 10 minutes at 4º C to pellet nuclei.

   25 mL

4. Resuspend in 2 mL 0.1 M NaCl 0.5 mL of 5M
   20 mM Tris pH 8.0 0.5 mL of 1M
   10 mM EDTA 1.0 mL of 250 mM

   Add RNase to 10 ug/mL (2 uL of 10 mg/mL).
   Incubate @ RT 15 minutes.

5. Add Proteinase K to 100 ug/mL (10 uL of 20 mg/mL) and SDS to 0.5% (100 uL of 10 %).

   Incubate at 65ºC for 1 hour.

6. Phenol extract 1X
   Phenol/Chloroform extract 1X
   Chloroform 1X
   ETOH precipitate: add 200uL of 3M NaOAC, 4.5 ml ETOH, -20ºC O/N.

   Mix gently and fish out DNA into Eppendorf tube with 1 mL 70% EtOH.
   Spin down, remove supernatant and dry.

7. Resuspend in TE 1 uL per fly.