DNA Extraction Protocol

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Materials Needed:

Zymo Genomic DNA Clean & Concentrator Kit
Homogenization buffer (HB)
- 30 mM Tris HCL (pH 8.0)
- 10 mM EDTA
- 0.1 M NaCl
- 0.5% Triton X-100
SDS 10%
RNase A
Proteinase K

IMPORTANT NOTE: This protocol is adapted from Dr. Jeremy Wang's high molecular weight extraction protocol for nanopore sequencing. Be sure that all solutions from the Zymo kit have the appropriate volume of 100% ethanol added.

1. Add 500 µL of HB to a 1.5 mL tube with 20-40 frozen flies and then homogenize the flies using a clean pestle until you can no longer recognize any fly parts.

2. Add 125 µL of SDS and 10 µL of Proteinase K then vortex for 15 seconds.

3. Incubate at 55C for 1 hour (inverting every 10 minutes) and after 30 minutes add 10 µL of RNase A.

4. Add twice the volume of ChIP DNA Binding Buffer to each volume of DNA sample and mix thoroughly.

5. Pipette the mixture into a Zymo-Spin Column in a collection tube, centrifuge at 16,000 x g for 30 seconds, and discard the flow-through.

6. Add 200 µL of DNA Wash Buffer to the Zymo-Spin Column, centrifuge at 16,000 x g for 1 minute, and discard the flow-through.

7. Repeat step 6.

8. Transfer the Zymo-Spin Column to a 1.5 mL tube and to elute the DNA add 32 µL of DNA Elution Buffer directly onto the column matrix and centrifuge at 16,000 x g for 30 seconds.

9. Store the eluted DNA in the cold room until quantification.