Quick Genomic DNA Prep

Materials

- Microcentrifuge tubes (1.5 mL)
- Disposable tissue grinders
- Microcentrifuge

Solutions and Reagents

- Isopropanol
- Ethanol (70%)
- TE
- Buffer A (Store at room temperature)
  100 mM Tris-Cl (ph 7.5)
  100 mM EDTA
  100 mM NaCl
  0.5% SDS
- Buffer B (Mix together and store at 4^{o}C
  2 mL of 5 M potassium acetate (use 2430 uL for 10)
  5 mL of 6 M lithium chloride (use 6070 uL for 10

Methods

1. Collect 30 anesthetized flies in a 1.5 mL microcentrifuge tube placed on ice.
   Note: Flies can be stored at -80 ^{o}C indefinitely or DNA can be prepared immediately w/o freezing the flies.

2. Grind flies in 200 uL of Buffer A with a disposable tissue grinder. Add an additional 200 uL of Buffer A (total volume of 400 uL) and continue grinding until only cuticles remain (about 1-2 min., grinding by hand).

3. Incubate samples at 65 ^{o}C for 30 min.

4. Add 800 uL of Buffer B to each sample, mix well by inverting the tube multiple times, and incubate on ice for at least 10 min. and up to a few hours.

5. Centrifuge in a microcentrifuge at 12,000 rpm at room temperature for 15 min.

6. Transfer 1 mL of the supernatant into a new microcentrifuge tube. Some of the precipitate will not pellet, but instead float on top of the supernatant; be extremely careful to avoid transferring any floating precipitate. Discard the pellet.

7. Repeat step 5 and 6 to get rid of any contaminating precipitate.

8. Add 600 uL of isopropanol to each sample, and mix well by inverting the tube several times. If 800 uL of supernatant, then add 480 uL of isopropanol.

9. Centrifuge in a microcentrifuge at 12,000 rpm at room temperature for 15 min.

10. Discard supernatant (use big 1000 uL pipettes, then 200 uL pipettes) and wash pellet with 70% ethanol (0.5-1 mL).

11. Vortex (30 sec.) and centrifuge at 14,000 rpm at room temperature for 10 min.

12. Remove ethanol.

13. Repeat steps 10-12.

14. Air dry the microcentrifuge tubes for about 30 min. with kim wipe covering tubes.

15. Resuspend pellet in 150 uL of TE and then vortex for 30 sec.

16. Mix the solution with a pipette by sucking.

17. Incubate at 60^{o}C for 10 min.

18. Vortex for 30 sec.

19. Store the DNA at -20 ^{o}C.