

Worm DNA prep.

1. Grow several large plates of worms on NGM plates until starved
2. Wash worms several times in TEN solution. (Resuspend in about 500 ul.)
3. Add SDS to 0.5% and Proteinase K to 200 mg/mL

For 500 ul worms, add:

- 12.5 ul of 20% SDS
- 5 ul 20 mg/mL proteinase K

4. Incubate at 65° for 1 or 2 hours.
5. Phenol extract several times

- a. add phenol
- b. mix well (don't vortex)
- c. spin 2 min.

d. take the top phase

e. repeat if necessary

6. Add 3M Sodium Acetate solution to 0.3M and 0.4 mL ethanol (100%)

For 500 ul worms, add:

- 50 ml of 3M sodium acetate solution
- 0.4 mL of 100% ethanol

7. Collect DNA by spooling on a glass rod.

8. Wash briefly in 70% ethanol

9. Dry

10. Resuspend DNA in ~ 500 ml of TE solution

11. Add RNase A to 40 ng/ul

For 500 ul worms, add:

- 1 ul 20mg/mL RNaseA

12. Phenol extract once

13. Add 3M Sodium Acetate solution to 0.3M and 0.4 mL ethanol (100%)

For 500 ul worms, add:

- 50 ml of 3M sodium acetate solution

- 0.4 mL of 100% ethanol

14. Collect DNA by spooling on a glass rod.

15. Wash briefly in 70% ethanol

16. Dry

17. Resuspend DNA in ~ 100 ml of TE solution

Solutions

TE - off the shelf TEN - TE + 100mM NaCl (or 2% 5M NaCl)