

Extra Quick High Salt Extraction

1. Turn on heat block to 55°C.
2. Add 500 µl of STE buffer – the same tip can be used for all tubes.
3. Pick out one piece of muscle (~3x2 mm), liver or a fin clip. Use an open flame to clean the cutting equipment after each sample is taken.
4. Add 15 µl of 1% proteinase K to each tube. Proteinase K is heat stable, and therefore it is not necessary to keep on ice. Use a new tip for each tube.
5. Add 75 µl of 10% SDS to each tube.
6. Thoroughly mix contents of the tubes by inversion.
7. Place samples in the heat block (55°C) till the tissue is completely dissolved (up to overnight or add more Prot. K). Shake samples to dislodge any solid pieces.
8. Add 60 µl of 5M NaCl to each tube (10% total volume).
9. Thoroughly mix contents of the tube by inversion.
10. Place on ice or fridge for at least 1 hour to overnight.
11. Centrifuge samples at full speed (14K) for 10 minutes.
12. Label new tubes.
13. Collect supernatant – be conservative by leaving what can not be cleanly collected. Place the pipette tip on the side away from the pellet. Place the supernatant into the newly labeled tubes – keeping them on ice.
14. Centrifuge the collected supernatant for additional 5 minutes at full speed.
15. Collect supernatant, and place into a newly labeled tube.
16. Add 1000 µl of absolute alcohol (or 95%) to the supernatant.
17. Thoroughly mix.
18. Precipitate the samples overnight in the -20°C freezer.
19. Centrifuge for 10 minutes at full speed (14K).
20. Pour off alcohol, leaving the pellet of DNA behind.
21. Wash the pellet by adding ~500 µl of 70% alcohol. The same tip can be used if the alcohol is dribbled in from above the tube.
22. Repeat steps 21 and 22. If the DNA pellet becomes loose, spin for 2 minutes at 14K.
23. Remove all alcohol, leaving the pellet of DNA behind.
24. Let the pellets either air dry or dry in the speedvac.
25. When the tubes are dry resuspend the pellet in 50 µl of water and let dissolve.
26. Store suspended DNA at -20°C freezer.