

gDNA Extraction from Rodent Tail

Please reference the current Genfind v2 protocol for product information and a detailed description of use.

1. On dry ice cut 20 mg of rodent tail. Cut it into 3-4 smaller pieces and place it into the well of a 1.2 mL 96 well deepwell plate (Thermo Fisher DW-1127).
2. Add 200 μ L Lysis Buffer and to each sample. Seal the plate. Incubate at 70°C for 1 hour.
3. Add another 200 μ L l of Lysis buffer and 9 μ L of Proteinase K. Gently pipette tip mix samples 5 times. Seal the plate. Incubate at 55°C overnight.
4. Transfer the entire supernatant into clean 1.2 mL 96 well deepwell plate avoiding bones and hair.
5. Vigorously shake the Binding Buffer bottle to ensure complete resuspension of the beads. For each sample mix 200 μ L water and 300 μ L Binding Buffer. Add 250 μ L of binding buffer/ water to each well and pipette mix 5 times.
6. Add another 250 μ L of binding buffer/ water to each well and pipette mix 5 times.
7. Incubate at room temperature for 5 minutes.
8. Place the sample plate on the SuperMagnet plate for 15 minutes to separate.
9. With the plate still on the magnet, gently aspirate off the supernatant. It may be necessary to perform multiple aspirations to remove the supernatant without disturbing the beads.
10. Take the plate off the magnet and add 500 μ L Wash Buffer 1 and tip mix 10 times. The beads should be resuspended from the bottom of the well.
11. Place the plate back on the magnet for 6 minutes and, while the plate is on the magnet, carefully aspirate the supernatant.
12. Repeat steps 10 and 11 for a total of 2 washes with Wash Buffer 1.
13. Take the plate off the magnet and add 500 μ L of Wash Buffer 2 to each sample and tip mix 10 times.
14. Place the plate back on the magnet for 6 minutes and, while the plate is on the magnet, carefully aspirate the supernatant.
15. Repeat step 13 and 14. After the second Wash Buffer 2 wash, remove as much of the liquid as possible.
Optional for gDNA for restriction digests: resuspend the beads in 500 μ L 70% ethanol, let the beads separate for 6 minutes and remove the supernatant.
16. Add 200 μ L of elution buffer (water or TE) to each sample.
17. Remove the plate from the magnet and resuspend the beads by gently tip mixing 10 times. Incubate the plate for 2 minutes at room temperature and again tip mix the samples 10 times.
18. Place the plate on the magnet for 10 minutes or until the supernatant clears. Transfer the eluate to a clean plate or tubes.