Supplementary Material 1. Protocol of DNA extraction for Nanopore long-reads genome sequencing

Use only materials and reagents DNase free. Never vortex and store DNA extracts on ice from step 9. Avoid DNA freezing-thawing.

1. Using liquid nitrogen and pre-chilled mortar and pestle, grind 0.2/0.3 g of fresh mycelium into powder, in less than 1 minute. Place the powder into a 15 mL Falcon tube and store in liquid nitrogen or at -80°C.
2. Add 5 mL TE 1X pH8 and 300 µL SDS 10 %. Homogenize without vortexing until the complete thawing of the mixture.
3. Add 100 µL of proteinase K at 20 mg / mL. Homogenize by flicking the tube without vortexing and incubate at 37 °C over night in a dry oven.
4. Centrifuge 3 min at 4000 rpm to eliminate tissues debris. Transfer the supernatant in a new 15 mL Falcon tube.
5. Add 200 µL of SDS 10 % and Homogenize by slow inversion.
6. Add 1,3 mL NaCl 5 M and mix vigorously until obtaining white foam.
7. Incubate 30 min in ice and mix again vigorously until obtaining white foam.
8. Centrifuge 25 min at 4000 rpm at 4 °C (with brake and acceleration on 4). Transfer the supernatant in a new 15 mL Falcon tube with caution.
9. Add 7 mL of phenol/chloroform/isoamyl alcohol 25:24:1. Mix by inversion 200 times and centrifuge 10 min at 5000 rpm at 4 °C. Transfer the upper aqueous phase into a new 15 mL Falcon tube taking care to avoid the aqueous/organic interface. Warning: store tubes in ice during the aqueous phase transfer.
10. Add Rnase A for 2% of the final volume, and place the tubes at 37°C for 30min in a heating unit or a water bath.
11. Repeat the step 9.
12. Add 1/10 volume of NaAc 3 M pH = 5.2 and 1 volume of isopropanol and mix gently by inversion. Place the tubes at -20 °C for 3 h.
13. Centrifuge 30 min at 8000 rpm at 4 °C. Eliminate the supernatant by spilling the tube. Briefly spin and eliminate the residual supernatant by pipetting.
14. Rinse the DNA pellet with 6 mL of ethanol 70 % and centrifuge 5 min at 8000 rpm at 4 °C. Eliminate the supernatant as above. Repeat this step twice. Dry the DNA pellet at room temperature by opening the tube until no ethanol remains.
15. Re-suspend the DNA pellet with caution in 500 µL DNase-free water. Possibility to store DNA extracts at -20 °C and avoid freeze-thaw cycles.
16. Make tests of DNA extract quality. If the DNA extract quality if insufficient, perform steps 17 to 22.
17. To remove polysaccharides residues, add 500 µL of NaCl 2.4 M (1.2 M final).

18. Add 1 mL of diethyl ether saturated with DNase-free water (to prepare mix by vortexing 1 volume of diethyl ether and 1 volume of DNase-free water – let the two phases separating and recover the organic upper phase corresponding to diethyl ether saturated in water). Mix by inversion 200 times and centrifuge 20 min at 8000 rpm at 4 °C. Transfer the lower aqueous phase into a new 15 mL Falcon tube.

19. Add 2.5 volumes of pure ethanol (prechilled at -80°C) and place the tubes at -20 °C over night. If the DNA pellet appear while adding the pure ethanol, possibility to recover the pellet with a cut cone and place it in a new 15mL Falcon tube with 3mL of ethanol 70%.

20. Centrifuge 35 min at 8000 rpm at 4 °C. Eliminate the supernatant by spilling the tube. Briefly spin and eliminate the residual supernatant by pipetting.

21. Rinse the DNA pellet with 5 mL of ethanol 70 % and centrifuge again 20 min at 8000 rpm at 4 °C. Eliminate the supernatant as above. Repeat this step twice. Dry the DNA pellet at room temperature by opening the tube until no ethanol remains.

22. Re-suspend the DNA pellet with caution in 100uL of Dnase-free water.

23. Make tests of DNA extract quality. If the DNA extract quality if insufficient, perform steps 24.

24. Purification on Genomic-tip 100/G - Blood & Cell Culture DNA Midi Kit (25) 13343 QIAGEN according to Special Applications protocol 'Purification of genomic DNA prepared by other methods*' , followed by QIAGEN Genomic-tip procedure - Part II: Genomic-tip Protocol Protocol for Isolation of Genomic DNA from Blood, Cultured Cells, Tissue, Yeast, or Bacteria - QIAGEN Genomic DNA Handbook 08/2001, with at step 5 an over night precipitation of the total DNA in isopropanol at – 20 °C, followed day 4 by a centrifugation 20 min at 8000 rpm to finish by step 6B and resuspension of the total DNA in 100 µL DNase-free water or buffer according to the sequencing company. Note: At least 2/3 of DNA is lost throughout the final 100/G purification. At the end 8 µg of DNA must be obtained so the quantity of DNA extracted must be adapted to the prerequisite of the sequencing company.