Improved methods for high yield genomic DNA of cat stools using DNeasy PowerSoil Pro Kit.

Hajar Fauzan Ahmad

Faculty of Industrial Sciences and Technology (FIST), Universiti Malaysia Pahang, Malaysia.

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1 Spin the PowerBead Pro Tube briefly to ensure that the beads have settled at the bottom. Add up to 250 mg (ideally 200 to 250 mg) of cat stool and 800 µl of Solution CD1, together with 25 µL of Proteinase K. Vortex briefly to mix.

2 Vortex at maximum speed for 10 min. Upon completion, transfer to Eppendorf ThermoMixer to continue incubating, and mixing almost for 20 minutes at 65°C.

3 Centrifuge the PowerBead Pro Tube at 15,000 x g for 1 min. Transfer the supernatant to a clean 2 ml Microcentrifuge Tube (provided).

4 Add 200 µl of Solution CD2 and vortex for 5 s. Centrifuge at 15,000 x g for 1 min. Avoiding the pellet, transfer up to 700 µl of supernatant to a clean 2 ml Microcentrifuge Tube (provided).

5 Add 600 µl of Solution CD3 and vortex for 5 s.
6 Load 650 µl of lysate to an MB Spin Column. Centrifuge at 15,000 x g for 1 min. Discard the flow-through and repeat step 8 to ensure that all of the lysate has passed through the MB Spin Column.

7 Carefully place the MB Spin Column into a clean 2 ml Collection Tube (provided). Avoid splashing any flow-through onto the MB Spin Column.

8 Add 500 µl of Solution EA to the MB Spin Column. Centrifuge at 15,000 x g for 1 min. Discard the flow-through and place the MB Spin Column back into the same 2 ml Collection Tube.

9 Add 500 µl of Solution C5 to the MB Spin Column. Centrifuge at 15,000 x g for 1 min.

10 Discard the flow-through and place the MB Spin Column into a new 2 ml Collection Tube (provided). Centrifuge at up to 16,000 x g for 2 min. Carefully place the MB Spin Column into a new 1.5 ml Elution Tube (provided).

11 Add 100 µl of Solution C6 to the center of the white filter membrane, and let the C6 remain at filter membrane for 10 minutes.

12 Centrifuge at 15,000 x g for 1 min. Discard the MB Spin Column. The DNA is now ready for downstream applications.