Quick-Start Protocol August 2016

QIAamp® PowerFecal® DNA Kit

The QIAamp PowerFecal DNA Kit can be stored at room temperature (15–25°C) until the expiry date printed on the box label.

Further information

- Safety Data Sheets: **www.qiagen.com/safety**
- Technical assistance: **support.qiagen.com**

Notes before starting

- Perform all centrifugation steps at room temperature (15–25°C)
- If Solution C1 has precipitated, heat at 60°C until precipitate dissolves.
- Shake to mix Solution C4 before use

1. Add 0.25 g of stool or biosolid to the Dry Bead Tube provided.
   Note: For fecal samples that are especially high in lipids, polysaccharides and protein (e.g. meconium or some bird feces), smaller amounts of starting material (~0.10 g) may improve DNA yield and purity.
2. Add 750 µl of PowerBead Solution to the Dry Bead Tube.
3. Add 60 µl of Solution C1 and invert several times or vortex briefly.
4. Heat the tubes at 65°C for 10 min.
5. Secure tubes horizontally using a Vortex Adapter tube holder (cat. no. 13000–V1–24). Vortex at maximum speed for 10 min.
6. Centrifuge the tubes at 13,000 x *g* for 1 min.
7. Transfer the supernatant to a clean 2 ml collection tube (provided). Expect between 400 to 500 µl of supernatant.
8. Add 250 µl of Solution C2 and vortex briefly to mix. Incubate at 2–8°C for 5 min.

Note: You can skip the 5 min incubation. However, if you have already validated the PowerFecal extractions with the incubation we recommend you retain the step.

9. Centrifuge the tubes at 13,000 x *g* for 1 min.
10. Avoiding the pellet, transfer up to 600 µl of supernatant to a clean 2 ml collection tube.
11. Add 200 µl of Solution C3 and vortex briefly. Incubate at 2–8°C for 5 min.

    Note: You can skip the 5 min incubation. However, if you have already validated the PowerFecal extractions with the incubation we recommend you retain the step.
12. Centrifuge the tubes at 13,000 x *g* for 1 min.
13. Avoiding the pellet, transfer the supernatant to a clean 2 ml collection tube (provided). Do not transfer more than 750 µl at this step.
14. Add 1200 µl of Solution C4 to the supernatant and vortex for 5 s.
15. Load 650 µl of supernatant onto a MB Spin Column and centrifuge at 13,000 x *g* for 1 min. Discard the flow through and repeat until all the supernatant has been processed.
    Note: Each sample processed will require a total of three loads.
16. Add 500 µl of Solution C5 and centrifuge for 1 min at 13,000 x *g*.
17. Discard the flow through and centrifuge again for 1 min at 13,000 x *g*.
18. Carefully place the MB Spin Column in a clean 2 ml Collection Tube (provided).

    Note: Avoid splashing any of Solution C5 onto the MB Spin Column.
19. Add 100 µl of Solution C6 to the center of the white filter membrane. Alternatively, you may use sterile, DNA-free, PCR-grade water or TE buffer (cat. no. 17000-10).
    Note: Eluting with 100 µl of Solution C6 will maximize DNA yield. For more concentrated DNA, a **minimum** of 50 µl of Solution C6 can be used.
20. Centrifuge at 13,000 x g for 1 min and discard the Spin Filter basket. The DNA in the tube is now ready for any downstream application.
    Note: We recommend storing DNA frozen (–20° to –80°C) as Solution C6 does not contain EDTA. To concentrate DNA see the Hints & Troubleshooting Guide.