

Quick-Start Protocol

DNeasy[®] 96 PowerSoil[®] Pro QIAcube[®] HT Kit

This protocol is for the purification of microbial genomic DNA from all soil types and stool samples using the DNeasy 96 PowerSoil Pro QIAcube HT Kit with the QIAcube HT Prep Manager software.

Solution CD2 should be stored at 2–8°C upon arrival. All other reagents and kit components should be stored at room temperature (15–25°C), until the expiry date printed on the box label.

Further information

- *DNeasy 96 PowerSoil Pro QIAcube HT Kit Handbook*: www.qiagen.com/HB-2592
- QIAcube HT protocols and installation files: www.qiagen.com/QIAcubeHT-Resources
- Safety Data Sheets: www.qiagen.com/safety
- Technical assistance: support.qiagen.com

Notes before starting

- If Solution CD3 has precipitated, heat at 60°C until precipitate dissolves.
- Prepare Buffers AW1 and AW2 according to the instructions on the bottles.
- For soil samples, use a maximum of 250 mg. If processing stool samples, perform a vacuum check on the QIAcube HT, and use no more than 200 mg as starting material. For dehydrated stool samples, start with no more than 100 mg.

Sample preparation

1. Spin the PowerBead Pro Plate or PowerBead Pro Tube briefly to ensure that the beads have settled at the bottom of the wells or the tube.
2. Add up to 250 mg of soil or 200 mg of stool, and 800 µl of Solution CD1. Seal the plate or recap the tube. Vortex briefly.
3. Homogenize samples thoroughly using one of these methods:

3a. **TissueLyser II:**

If using a PowerBead Pro Plate, place the sealed plate between two Plate Adapters (cat. no. 11990). Shake for 5 min at 25 Hz.

Reorient the plates so that the side that was closest to the machine body becomes furthest from it. Shake again for 5 min at 25 Hz.

If using PowerBead Pro Tubes, place the tubes into a TissueLyser Adapter Set 2 x 24 (cat. no. 69982) or a 2 ml Tube Holder (cat. no. 11993) and Plate Adapter Set (cat. no. 11990). Fasten the adapter into the TissueLyser II. Shake for 5 min at 25 Hz. Reorient the adapter so that the side that was closest to the machine body becomes furthest from it. Shake again for 5 min at 25 Hz.

3b. **PowerLyzer 24:**

Place PowerBead Pro Tubes symmetrically into the tube holder and secure the finger plate in place over the carousel. Operate the instrument at 2000–4000 rpm for 2 x 30 s, with a 30 s break in between.

Note: Higher forces, such as 4000 rpm, increase the risk of DNA fragmentation but provide higher yields.

3c. **Vortex-Genie® 2:**

Secure PowerBead Pro Tubes horizontally using a Vortex Adapter for 24 (1.5–2.0 ml) tubes (cat. no. 13000-V1-24). Vortex at maximum speed for 10 min.

Note: If using a 24-tube Vortex Adapter for more than 12 preps, increase the vortex time by 5–10 min. Avoid using tape, which can become loose and result in reduced homogenization efficiency, inconsistent results, and reduced yields.

4. Centrifuge the PowerBead Pro Plate at $4500 \times g$ for 6 min, or the PowerBead Pro Tubes at $15,000 \times g$ for 1 min.
5. Transfer the supernatant to a new S-Block (provided).
Note: Expect 480–600 μl . The supernatant may still contain some soil particles.
6. Add 250 μl of Solution CD2 and mix by pipetting. Seal the plate with Sealing Tape.
Note: If processing samples with very high inhibitor content, use 300 μl Solution CD2.
7. Centrifuge the S-Block at $4500 \times g$ for 6 min at room temperature.
8. Avoiding the pellet, transfer 550 μl of the supernatant into a new S-Block.
Note: For more convenience, transferring the supernatant can be done using the QIAcube HT instrument. Choose **Transfer Protocol** under setup/selected protocol. For detailed information, refer to the *DNeasy 96 PowerSoil Pro QIAcube HT Kit Handbook*.
9. Transfer the S-Block (from step 8) containing 550 μl of supernatant to the QIAcube HT instrument. Proceed to “DNeasy 96 PowerSoil Pro protocol on the QIAcube HT” below.

DNeasy 96 PowerSoil Pro protocol on the QIAcube HT

1. Start the QIAcube HT Prep Manager software. Click **Home** in the main toolbar to access the Home screen.
2. Select **DNeasy 96 PowerSoil Pro** from the “Create experiment” list. Follow the instructions in the wizard and fill in all required fields.
3. In the “Setup” step, select **soil**, **stool**, or **any** as Sample type, and **manual** under Pre-treatment.
4. Select **DNeasy 96 PowerSoil Pro Protocol**. For information about optional steps and advanced options, refer to the *DNeasy 96 PowerSoil Pro QIAcube HT Kit Handbook*.
5. Define samples in the “Labware selection” step.
6. Arrange samples to the output plate in the “Assignment” step.
Note: The instrument must be switched on and connected to the software before entering the “Worktable” step.

7. Follow the instructions on the virtual worktable to prepare the instrument worktable. Sample volume is 550 µl (in the S-Block from step 8 in the “Sample preparation” section of this protocol).
8. Save the experiment by clicking **Save** in the button bar.
9. Click **Start run** to start the run.

Important: If the optional “Vacuum performance” step has been selected, the software will show a dialog that needs to be confirmed after defined vacuum steps.
10. When the protocol is complete, cover the elution plate (EMTR) with the lid and remove it from the elution chamber.

Note: If using Top Elute fluid, there may be two liquid phases in the elution microtubes. Top Elute fluid will be the top layer, above the elution buffer.
11. Create a report if required.
12. Follow the cleaning procedure.

Revision History

Document	Changes	Date
HB-2554-002	Updated storage conditions. Corrected the related handbook link and QR code.	May 2019



Scan QR code for handbook.

For up-to-date licensing information and product-specific disclaimers, see the respective QIAGEN kit handbook or user manual.

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