Quick-Start Protocol October 2015

QIAamp® Fast DNA Tissue Kit

The QIAamp Fast DNA Tissue Kit (cat. no. 51404) can be stored at room temperature (15–25°C) for up to one year.

Further information

- *QIAamp Fast DNA Tissue Kit Handbook*: **www.qiagen.com/HB-1931**
- Safety Data Sheets: **www.qiagen.com/safety**
- Technical assistance: **support.qiagen.com**

Notes before starting

- Add ethanol to Buffer AW1 and Buffer AW2 concentrates.
- Add isopropanol to Buffer MVL concentrate.
- Preheat a thermomixer to 56°C for use in steps 4 and 5.
- Unless stated otherwise, all centrifugation steps should be performed at full speed (maximum 20,000 *x g*) for 30 s at room temperature in a conventional tabletop centrifuge.

1. Weigh and cut the tissue sample to a suitable size (5–25 mg), then place in the Tissue Disruption Tube (supplied).
2. Add the following buffers and enzymes: 200 µl AVE, 40 µl VXL, 1 µl DX Reagent, 20 µl Proteinase K, 4 µl RNase A (100 mg/ml). Tightly cap the lid.

   Optional: When processing multiple samples, you may prepare a master digestion buffer mix.
3. Homogenize using one of the following three options:
   a. Vortex-Genie®2 with appropriate 2 ml tube adapter: full speed for 5 min
   b. TissueLyser II with TissueLyser Adapter Set 2 x 24: 24 Hz for 30 s

c. TissueLyser LT: 45 Hz for 2 min

Note: Do not use frequencies higher than 24 Hz if using the TissueLyser II.

Note: Proceed with step 4 regardless of whether there is residual tissue visible or not.

4. Incubate in a thermomixer at 1000 rpm for 10 min at 56°C.
5. If the lysate is homogenous after step 4, proceed directly with step 6. If there is still residual tissue left after step 4, repeat steps 3 and 4 a single time with the same settings as for the first homogenization.

   Note: If there is residual tissue left after repeating steps 3 and 4, increase the incubation (1000 rpm at 56°C) time up to 1 h. Avoid longer incubation periods. Proceed with step 6 even if there is residual tissue remaining after the prolonged incubation.
6. Add 265 µl Buffer MVL and mix by pipetting or vortexing.
7. Apply the mixture from step 6 to the QIAamp Mini spin column and centrifuge for 1 min. Place the QIAamp Mini spin column in a clean 2 ml collection tube (provided) and discard the tube containing the filtrate.
8. Add 500 µl Buffer AW1 to the spin column and centrifuge. Place the spin column into a new 2 ml collection tube (supplied).
9. Add 500 µl Buffer AW2 to the spin column and centrifuge. Place the spin column into a new 2 ml collection tube (supplied).
10. Centrifuge for 2 min. Place the spin column into a clean 1.5 ml microcentrifuge tube (supplied).
11. Add 50–100 µl ATE directly onto the spin column membrane, incubate at room temperature for 1 min and then centrifuge for 1 min.

    Optional: Repeat step 11 for increased yield.

Scan QR code for handbook.