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Quick-Start Protocol

QIAwave DNA Blood & Tissue Kit

The QIAwave DNA Blood & Tissue Kit (cat.no. 69556) can be stored at room temperature (15–25°C) for up to 1 year after delivery.

Further information

- *QIAwave DNA Blood & Tissue Handbook*: **www.qiagen.com/HB-2987**
- 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).
- Redissolve any precipitates in Buffer AL and Buffer ATL.
- Preparation of Buffer AW1: Transfer the entire volume of Buffer AW1 from the 125 ml bottle into a glass bottle larger than 230 ml, either by using a pipet or by pouring. Add 130 ml ethanol (96–100%) to Buffer AW1/C to obtain a final volume of 228 ml. Cap the glass bottle tightly and mix thoroughly by inverting the bottle several times. To label the glass bottle, peel off the upper label from the piggyback label on the 125 ml plastic bottle and transfer it onto the glass bottle.
- Preparation of Buffer AW2/C: Transfer the entire volume of Buffer AW2/C from the 15 ml bottle into a glass bottle larger than 230 ml, either by using a pipet or by pouring. Add 60 ml ultrapure water such as nuclease-free water (1000 ml, cat. no. 129115; 5 liters, cat. no. 129117) and 160 ml ethanol (96–100%) to obtain a final volume of 226 ml. Cap the glass bottle tightly and mix by inverting the bottle several times. To label the glass bottle, peel off the upper label from the piggyback label on the 15 ml plastic bottle and transfer it onto the glass bottle.

- Preparation of Buffer AE/C: Transfer the entire volume of Buffer AE/C from the 15 ml bottle into a glass bottle larger than 120 ml, either by using a pipet or by pouring. Add 110 ml ultrapure water such as nuclease-free water (1000 ml, cat. no. 129115; 5 liters, cat. no. 129117) to obtain a final volume of 120 ml. Cap the glass bottle tightly and mix by inverting the bottle several times. To label the glass bottle, peel off the upper label from the piggyback label on the 15 ml plastic bottle and transfer it onto the glass bottle.
- Equilibrate frozen tissue or cell pellets to room temperature.
- Preheat an incubator to 56°C.
- Refer to the handbook for pretreatment of fixed tissue, insect, bacterial, or other materials.
- Preassemble DNeasy® Mini Spin Columns with Waste Tubes.

Procedure

1. **Tissue**: Cut tissue (≤10 mg spleen or ≤25 mg other tissue) into small pieces, and place in a 1.5 ml microcentrifuge tube (not provided). For rodent tails, use 1 (rat) or 2 (mouse) 0.4–0.6 cm lengths of tail. Add 180 µl Buffer ATL. Add 20 µl proteinase K, mix by vortexing and incubate at 56°C until completely lysed. Vortex occasionally during incubation. Vortex 15 s directly before proceeding to step 2.

   Nonnucleated blood: Pipet 20 µl proteinase K into a 1.5 ml or 2 ml microcentrifuge tube (not provided). Add 50–100 µl anticoagulant-treated blood. Adjust volume to 220 µl with PBS. Proceed to step 2.

   Nucleated blood: Pipet 20 µl proteinase K into a 1.5 ml or 2 ml microcentrifuge tube (not provided). Add 5–10 µl anticoagulant-treated blood. Adjust volume to 220 µl with PBS. Proceed to step 2.

   Cultured cells: Centrifuge a maximum of 5 x 10^6 cells for 5 min at 300 x *g* (190 rpm). Resuspend in 200 µl PBS. Add 20 µl proteinase K. Proceed to step 2.
2. Add 200 µl Buffer AL. Mix thoroughly by vortexing. Incubate blood samples at 56°C for 10 min.
3. Add 200 µl ethanol (96–100%). Mix thoroughly by vortexing.

4. Pipet the mixture into a DNeasy Mini spin column placed in a 2 ml Waste Tube (supplied). Centrifuge at ≥6000 x *g* (8000 rpm) for 1 min. Discard the flow-through and reuse the Waste Tube.
5. Add 500 µl Buffer AW1/C, and centrifuge for 1 min at ≥6000 x *g*. Discard the flow-through and reuse the Waste Tube.
6. Add 500 µl Buffer AW2/C and centrifuge for 3 min at 20,000 x *g* (14,000 rpm). Discard the flow-through and Waste Tube.
7. Transfer the spin column to a new 1.5 ml or 2 ml microcentrifuge tube (not provided).
8. Elute the DNA by adding 200 µl Buffer AE/C to the center of the spin column membrane. Incubate for 1 min at room temperature (15–25°C). Centrifuge for 1 min at ≥6000 x *g*.
9. **Optional**: Repeat step 8 for increased DNA yield.

Document Revision History

Date	Changes
01/2022	Initial release

Scan QR code for handbook.

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