

## Quick-Start Protocol

# QIAwaye DNA Blood & Tissue Kit

The QIAwave DNA Blood & Tissue Kit (cat.no. 69554 and 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
- The QIAwave DNA Blood & Tissue Kit can be automated on the QIAcube Connect using the DNeasy Blood & Tissue Kit protocols that be downloaded at www.qiagen.com/qiacubeprotocols.

# Notes before starting

- Perform all centrifugation steps at room temperature (15–25°C).
- Redissolve any precipitates in Buffer AL and Buffer ATL.
- 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.
- Preparation of final buffers from concentrates: Transfer the entire volume of buffer
  concentrates from the 2 mL tube or 15 mL bottle into a glass bottle appropriate for the
  final volume (Table 1), either by using a pipette or by pouring. Add ultrapure water

and/or ethanol (96–100%) according to Table 1. To label the glass bottle, use the enclosed label and transfer it onto the glass bottle.

Table 1. Preparation of Buffer concentrates

Kit (cat. no.)	Final buffer	Buffer*	Ultrapure water	Ethanol (96-100%)	Final volume
69554	AW1 AW2 AE	AW1/C AW2/C AE/C	– 15 mL 22 mL	20 mL 40 mL -	35 mL 56.5 mL 24 mL
69556	AW1 AW2 AE	AW1/C AW2/C AE/C	– 60 mL 110 mL	130 mL 160 mL	228 mL 226 mL 120 mL

### 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  $\mu$ l proteinase K into a 1.5 ml or 2 ml microcentrifuge tube (not provided). Add 50–100  $\mu$ l anticoagulant-treated blood. Adjust volume to 220  $\mu$ l 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  $5 \times 10^6$  cells for 5 min at  $300 \times g$  (190 rpm). Resuspend in 200  $\mu$ l PBS. Add 20  $\mu$ l proteinase K. Proceed to step 2.

2. Add 200  $\mu$ l Buffer AL. Mix thoroughly by vortexing. Incubate blood samples at 56°C for 10 min.

- 3. Add 200  $\mu$ l 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, and centrifuge for 1 min at ≥6000 x g. Discard the flow-through and reuse the Waste Tube.
- 6. Add 500  $\mu$ l Buffer AW2 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  $\mu$ l Buffer AE to the center of the spin column membrane. Incubate for 1 min at room temperature. Centrifuge for 1 min at  $\geq$ 6000 x g.

Optional: Repeat step 8 for increased DNA yield.

### **Document Revision History**

Date	Changes
01/2022	Initial release
06/2023	Addition of cat. no. 69554 and necessary procedures
	Updated QSP according to new brand template



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