
# User-Developed Protocol:

# Isolation of DNA from soft tissues using the TissueLyser and QIAamp® DNA Mini Kit

This procedure has been adapted by customers and is for the isolation of DNA from soft tissues using the TissueLyser and the QIAamp DNA Mini Kit.* **The procedure has not been thoroughly tested and optimized by QIAGEN.**

The TissueLyser allows high-throughput, rapid and effective disruption of up to 48 tissue samples.

**Note:** This protocol has only been tested with 'soft' tissues (e.g., liver, spleen, thymus, heart, kidney, and brain) and may not work with 'hard' tissues (e.g., bone, teeth, and skin).

The following guidelines can be used for both fresh and frozen tissues, and for tissues stabilized with RNA*later*™ RNA Stabilization Reagent.

## Reagents and materials to be supplied by the user

- 5 mm stainless steel beads (cat. no. 69989)
- TissueLyser Adapter Set 2 x 24 (cat. no. 69982)
- 2 ml Safe-Lock microtubes (Eppendorf, cat. no. 0030 120.094)

## Important note before starting

- Use 10 mg tissue as starting material, increasing the amount if the protocol works satisfactorily.

# Procedure

1. **Pipet 180 µl Buffer ATL (tissue lysis buffer) into a 2 ml Safe-Lock microtube.**
2. **Add one stainless steel bead to each tube. For best results, use 5 mm (mean diameter) stainless steel beads.**
3. **Add 10 mg tissue to the tube, and assemble the TissueLyser.**
4. **Homogenize on the TissueLyser for 20 s at 30 Hz. Do not exceed this time as it may result in DNA shearing.**
5. **Centrifuge the sample briefly to ensure that all the tissue debris is on the bottom of the tube.**
6. **Add 20 µl proteinase K to the tube.**

   **Note:** Add 40 µl proteinase K if using RNA*later* stabilized tissues.
7. **Incubate for 56°C for 1 h in a shaker incubator.**

* All disruption steps can also be performed using the Mixer Mill MM 300 without modification.


8. **Briefly centrifuge the 2 ml microtube to remove drops from inside the lid.**

   **Optional: RNase treatment of the sample. Add 4 µl RNase A (100 mg/ml solution) to the tube, and mix by pulse-vortexing for 15 s. Incubate at room temperature (15–25°C) for 2 min.**

9. **Briefly centrifuge the 2 ml microtube to remove drops from inside the lid before adding 200 µl Buffer AL to the sample (add 220 µl Buffer AL if using RNA*later* stabilized tissues). Mix again by pulse-vortexing for 15 s, and incubate at 70°C for 10 min. Briefly centrifuge the 2 ml microtube to remove drops from inside the lid.**

10. **Continue with the Tissue Protocol in the *QIAamp DNA Mini Kit* and *QIAamp DNA Blood Mini Kit Handbooks*, from step 4.**

## Ordering Information

| Product | Contents | Cat. No. |
|---|---|---|
| **TissueLyser — for high-throughput disruption of a wide range of biological samples** | | |
| TissueLyser (220–240 V, 50/60 Hz) | Universal laboratory mixer-mill disruptor, 220–240 V, 50/60 Hz | 85220 |
| TissueLyser (120 V, 50/60 Hz) | Universal laboratory mixer-mill disruptor, 120 V, 50/60 Hz | 85210 |
| TissueLyser (100 V, 50/60 Hz) | Universal laboratory mixer-mill disruptor, 100 V, 50/60 Hz | 85200 |
| **Accessories** | | |
| TissueLyser Adapter Set 2 x 24 | 2 sets of Adapter Plates and 2 racks for use with 2.0 ml microcentrifuge tubes on the TissueLyser | 69982 |
| Stainless Steel Beads, 5 mm (200) | Stainless Steel Beads, suitable for use with the TissueLyser system | 69989 |
| TissueLyser Single-Bead Dispenser, 5mm | For dispensing individual beads (5 mm diameter) | 69965 |

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