TurboLyse Protease Tissue Mix Protocol

for efficient lysis of tissue samples

This protocol has been developed for fresh, frozen and stabilized human and animal tissue samples including DNase-rich (e. g., spleen, liver, kidney) and lipid tissues (e.g. brain, fat).

Materials and equipment needed

- **1 to 20 mg tissue** per sample depending on tissue type.
  Sample input recommendation:

Tissue type	Generic	High DNA content (e g., spleen, liver, kidney)	Low DNA content (e. g., muscle, cartilage)
Amount	10 mg	5 mg	20 mg

- **Microcentrifuge** with rotor for 1.5 and 2 ml reaction tubes.
- **Thermal shaker** with **agitation** (for fastest performance), capable of heating to 60°C and 80°C. Alternatively: Heating Block or heat chamber
- **Vortexer**
- **Pipets** for 10 µl and 200 µl scales, corresponding pipet tips
- One reaction tube (1.5 ml) per sample for the lysis step (preferably safe-lock).
- Ceramic Blade Scalpels. E.g. BioEcho product no. 050-002-001

Preparation before starting

- Heat the thermal shaker or thermo block to 60°C

PROTOCOL:

Lysis

1. Transfer tissue sample to the **bottom** of a 1.5 ml reaction tube (preferably safe-lock) while the tube is cooled on ice (or cooling block) to avoid DNA degradation during sample loading.

   Note:
   - If possible, cut tissue into small pieces to speed up lysis.
   - For stabilized tissue samples briefly rinse with water to remove traces of stabilization solution before adding samples to the reaction tube.
2. Add **90 µl Tissue Lysis Buffer** (LB) and **5 µl TurboLyse T Protease** (P) to each tissue sample. Mix by flicking or vortexing.

   If working with more than two samples, prepare a **Lysis Master Mix** with 10% excess volume for the number of tissue samples (see table).

Lysis Master Mix:

No of samples	**1**	**6 (+10%)**	**12 (+10 %)**	**Yours**
(LB) Tissue Lysis Buffer (µl)	90	600	1,190	
(P) TurboLyse T Protease (µl)	5	35	70	
Final volume (µl)	**95**	**635**	**1,260**	

Add **95 µl of the Lysis Master Mix** to each tissue sample.

3. Place the reaction tube(s) in the thermal shaker and incubate at **60°C** for **30 min** with max. agitation (for **60 min** if agitation is not feasible, in this case pulse-vortex 3 times during lysis).

   Note: If samples are **not** completely lysed after the time period described above, continue with the next step. Residual cellular debris will not interfere with the purification performance.

   Note: For some tissue types, lysis is already complete after 15 min. Step 3 may be shortened accordingly.
4. After incubation at 60°C, increase the temperature to **80°C** and incubate for additional **10 min** with max. agitation.
5. After having performed step 4, add **10 µl Clearing Solution T** (CS) to each sample. Vortex **3 sec**. The sample becomes cloudy.
6. Centrifuge for **2 min** at **full speed**.
7. Optional: Transfer **lysis supernatant (max. 100 µl)** containing the DNA into a new reaction tube.

The TurboLyse Protease Tissue Mix can be used to efficiently lyse tissues. For downstream applications, where high quality DNA is needed, we recommend our EchoLUTION Tissue DNA Extraction Kits to remove contaminants and inhibitors.

EchoLUTION Tissue DNA Kits	Reactions	Product No.
EchoLUTION Tissue DNA Kit (10)	10	010-002-010
EchoLUTION Tissue DNA Kit (50)	50	010-002-050
EchoLUTION Tissue DNA Kit (250)	250	010-002-250
EchoLUTION Tissue DNA 96 Kit (2 x 96)	2 x 96	010-102-002
EchoLUTION Tissue DNA 96 Kit (8 x 96)	8 x 96	010-102-008
EchoLUTION Tissue DNA 96 Core Kit (8 x 96)	8 x 96	010-102-108

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Product no.	010-122-001 (1 mL)	010-122-020 (20 mL)
Kit contents	Tissue Lysis Buffer, TurboLyse T Protease, Clearing Solution T	

Quick PROTOCOL (please read protocol first)

Lysis

- Transfer **10 mg tissue** to reaction tube
- Add **90 µl** (LB)
- Add **5 µl** (P), vortex briefly
- Incubate **30 min/60°C**, then **10 min/80°C**, max. agitation
- Add **10 µl** (CS) and vortex shortly
- Centrifuge 2 **min** at **max. speed**
- Transfer **lysate supernatant (max. 100 µl)** into new reaction tube

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BioEcho Life Sciences GmbH
Nattermannallee 1
50829 Köln (Cologne)/Germany

Phone: +49 (0) 221-99 88 97-0
E-Mail: contact@bioecho.de
www.bioecho.de