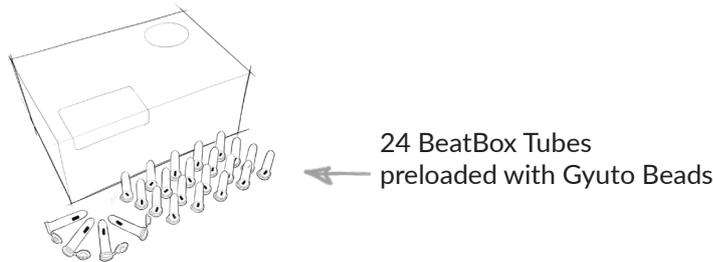




KIT CONTENT



BEATBOX ACCESSORIES



Pre-Requisites

Common lab equipment is required for the sample preparation.

Equipment	Quantity and Description
SAMPLE	Mammalian tissue (fresh/frozen), 5-50 mg wet. For other sample types contact PreOmics for adapted protocols.
LYSIS BUFFER	8–25 mL PreOmics® LYSE or RIPA buffer. See protocol for details. Contact info@preomics.com to order the required volume of PreOmics LYSE buffer. For SP3-iST kits, contact PreOmics for adapted protocols
2-FOLD iST LYSE BUFFER	Optional: Required to continue with iST sample preparation if RIPA was used as lysis buffer. See protocol for details. 2-fold iST LYSE needs to be ordered in addition to the iST kits from PreOmics.
CENTRIFUGE	Benchtop centrifuge for 2 mL tubes is required for spin-down of the homogenate.
BeatBox instrument	Tissue homogenizer with accessory kit.

Method

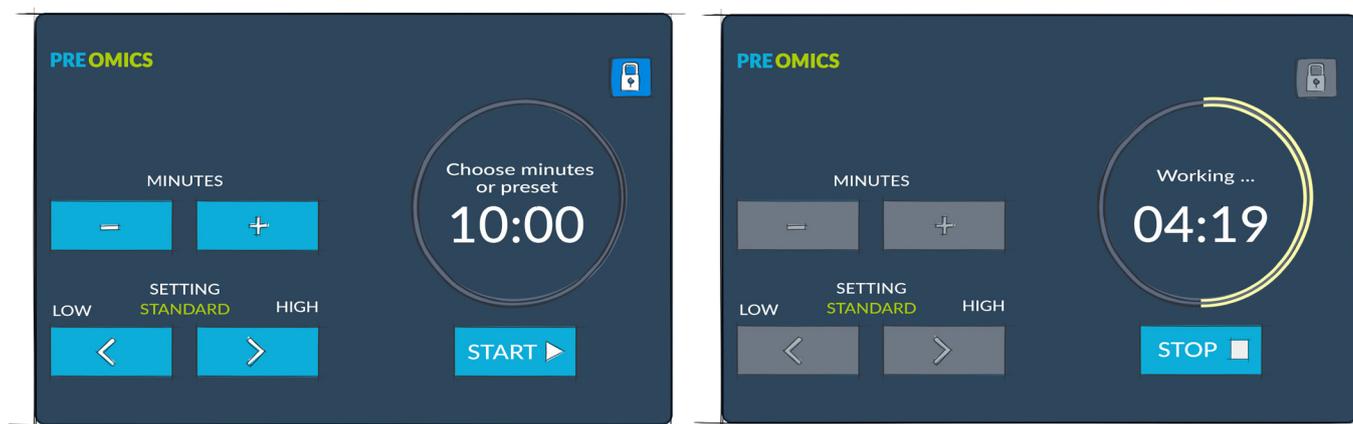
For a detailed description and graphical representation on how to use the BeatBox, please refer to the BeatBox Quick Start Manual 24x.

1. TUBE PREPARATION *NOTE1*

- 1.1. Carefully open the **BEATBOX TUBE**. Add wet **SAMPLE** to **BEATBOX TUBE**. *NOTE2*
- 1.2. Add 300–1000 µL of **LYSIS BUFFER** to each **BEATBOX TUBE**. **LYSIS BUFFER** can be PreOmics' LYSE or RIPA buffer. If you intend to freeze surplus homogenate, use RIPA buffer for homogenization and subsequent storage. *NOTE3*
- 1.3. Close the **BEATBOX TUBE**.

2. BEATBOX HOMOGENIZATION

- 2.1. Turn ON the BeatBox, place the **BEATBOX TUBES** into the **BEATBOX TUBE ADAPTER**, and insert the **BEATBOX TUBE ADAPTER** into the GARAGE
- 2.2. Use default configurations (SETTING: STANDARD; MINUTES: 10 min) or select optimal lysis conditions for your tissue sample by adjusting SETTING and MINUTES in the BEATBOX menu:



BeatBox screen for settings

BeatBox screen during processing

SETTING: User can choose between LOW, STANDARD, or HIGH. The power level increases from LOW to HIGH.

MINUTES: User can choose between 1–40 min (up to 10 min in 30 sec increments; above 10 min in 5 min increments).

- 2.3. Insert the GARAGE and press START.
- 2.4. After the BeatBox run is completed, remove the GARAGE from the instrument and the **BEATBOX TUBES** from the **BEATBOX TUBE ADAPTER**. If intact tissue is still visible, repeat BeatBox run (steps 2.1–2.5).
- 2.5. Spin down the **BEATBOX TUBES** (1500 rcf; 30–60 sec).

3. CONTINUE WITH PREOMICS' KITS

- 3.1. Determine the protein concentration of the homogenate.
- 3.2. When iST LYSE BUFFER is used, continue with the iST sample preparation using up to 100 µg of extracted protein (if the volume is <50 µL, fill up to 50 µL with LYSIS BUFFER).
Continue with step "2. DIGEST" and follow the protocol.
- 3.3. When RIPA buffer is used, continue with the iST sample preparation using up to 100 µg of extracted protein.
Add 2-FOLD iST LYSE BUFFER in ratio 1:1 (v/v) with tissue homogenate (if the volume is <50 µL, fill up to 50 µL with iST LYSE BUFFER).
Continue with step "2. DIGEST" and follow protocol.

NOTE1

SINGLE USE ONLY: Each BEATBOX TUBE and GYUTO BEAD can be used only once. It is recommended not to run the BEATBOX TUBES more than 40 minutes in total (independent of the setting).

NOTE2

For sample handling, plastic tweezers are preferable to metal tweezers to avoid sticking of the **GYUTO BEADS** to the tweezer. Allow deep-frozen and frozen tissue to thaw on ice for 5–10 min.

NOTE3

Buffer to sample ratio should be adjusted individually to create optimal conditions. Lower buffer volumes down to 100 µL are possible but recovering the full sample volume may be difficult. If lower lysis buffer volume is used, make sure that the **SAMPLE** is covered with buffer.

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