Tribio Cell RNA Lysis Buffer for Direct RT-qPCR (TBS6044)

Catalog	Unit Size
TBS6044-10	10 ml
TBS6044-50	50 ml
TBS6044-100	100 ml

Description

Tribioscience Cell RNA Lysis Buffer enables rapid preparation of RNA free of genomic DNA (gDNA) directly from cell, suitable for reverse transcription quantitative PCR (RT-qPCR) applications, including adherent or primary cells.

Application

- Rapid isolation of genomic DNA-free RNA directly from cell cultures.
- Suitable for downstream reverse transcription quantitative PCR (RT-qPCR or RT).
- Compatible with adherent cells and primary cells.

Main Features

- Fast preparation in approximately 20 minutes
- Eliminates genomic DNA contamination
- Works directly from cultured cells without additional purification steps
- Ideal for sensitive RT-qPCR applications

Storage

Store all components at -20°C for up to 1 year.

Procedure

Part A. Processing of Adherent Cells in a 96-Well Culture Plate

1. Pre-seed cells in a 96-well culture plate such that the cell density at harvest is 10–1 × 10^5 cells per well.
   Note: Excessive cell numbers may result in incomplete cell lysis and can inhibit RT-qPCR.
2. Completely remove the cell culture medium by aspiration.
3. Wash the cells with 120 µL of PBS at room temperature, then completely aspirate and remove the PBS.
4. Add 50 µL of the cell RNA lysis buffer into each well.
5. Incubate at room temperature for 10 minutes without agitation.
   Note: Do not mix by pipetting. Do not exceed 20 minutes at room temperature.
6. Transfer the lysate to a PCR plate or centrifuge at 75 °C for 5 min, then cool down on ice, quick spin to bring all liquid to the bottom of the tube or plate.
   Note: Use a thermal cycler to ensure optimal temperature uniformity.
7. Keep the cell lysate on ice for immediate use. For longer-term storage, keep at −20 °C or −80 °C.
8. Take 1-2 ul for RT-qPCR Reactions. We recommend to use our One-Step Sybr qRT-PCR (TBS4007).

Part B. Processing of Nonadherent Cells in a 96-Well PCR Plate

1. Prepare the appropriate volume of cell lysis master mix fresh on ice, mix thoroughly, and briefly centrifuge before use. Use the master mix for 2 hours.
2. Count and transfer 10–10^5 cells per well into a 96-well PCR plate or tube.
3. Centrifuge at 500–1,000 × g for 3 min and carefully remove the medium without disturbing the pellet.
4. Add 125 µL of room-temperature PBS to wash the cells. Centrifuge at 500–1,000 × g for 3 minutes and carefully aspirate 20 µL of the supernatant.
5. The following steps are the same as Part A, steps 5–8.

Relative Products

Tribo™ 2x Sybr qPCR Super Mix (TBS4001)
Tribo™ 2x TaqMan qPCR Super Mix (TBS4002)
Tribo™ Reverse Transcription Reaction Kit (TBS4006)
One-Step Sybr qRT-PCR (TBS4007)
One-Step TaqProbe qRT-PCR Kit (TBS4008)
Pfu PCR MasterMix (2x) (TBS4012)

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