Cellular RNA Extraction Kit (Cat. TC-02R)

Description:

- This kit is designed for rapidly isolating the highest yields and quality of RNA from cells/tissue samples, offering a simple, fast, environmental-friendly protocol for RNA extractions without using vacuum filtration and toxic organic solvents such as Trizol, phenol or chloroform. This kit is specially designed for preparing the pure RNA from small amounts of sample material for RT-PCR amplification, qPCR, and microarray. The kit is compatible with cells/tissues from LCM samples.
- **Key features:**
  1. Obtain the highest yield and integrity of RNA within 1 hour.
  2. No filter column or vacuum filtration is required, able to avoid the loss of RNA during extracting.
  3. Suitable for extracting the pure RNA from the small/tiny tissues or LCM samples.

Kit components and protocol: (keep all samples and buffers on ice during proceeding)

Components:	Quantity (50 extracts)	Storage
One-step Fast Lysis Buffer-A contains nuclease inhibitors	10.0 mL	–20°C
One-step P-Lysis Reagent-B	0.5 mL	–20°C
RNA Precipitation Solution (2x)	10.0 mL	–20°C

RNA Extraction Protocol:

1. Harvest cells (∽1x10e7) as usual in a 1.5ml microcentrifuge tube, and centrifuge at 1000 xg (or 3000rpm) for 5 minutes at 4°C. Carefully remove all the supernatant and save pellet on ice for RNA extraction.
2. Add **200ul one-step fast lysis buffer-A** and **10ul one-step P-Lysis Reagent-B** to the pellet and incubate on ice for 20 minutes. Vortex vigorously at highest speed for 20 seconds every 5 minutes.
3. Centrifuge at highest speed (∽13,000 xg) for 5 minutes at 4°C. Discard the pellet and transfer the supernatant into a new 1.5ml tube.

   Note: Discard the lipoproteins/lipids that may form an upper layer after centrifugation.
4. Add **200ul RNA Precipitation Solution (2x)** into the supernatant, mix thoroughly by pipette up and down several times and incubate at -20°C for 20 minutes or overnight.

   Note: Always keeps the bottle of RNA precipitation solution (2x) at -20° C.
5. Centrifuge at highest speed (∽13,000 xg) for 10 minute at 4°C. Carefully remove all the supernatant and save the pellet. The pellet is the RNA. .

   The RNA pellet will not be visible if the concentration is less than 20ng/ul.
6. Simply rinse the RNA pellets with 500ul ice cold 80% ethanol for 3 times without resuspending the RNA pellet. (Centrifuge at highest speed (∽13,000 xg) for 5 minute at 4°C if the pellets are resuspended.) Aspirate the liquids and Air-dry the pellets for 5-10 minutes.
7. Dissolve the RNA pellets in 20ul nuclease-free H_2O or TE. Centrifuge at highest speed (∽13,000 xg) for 2 minute at 4°C prior to measure the RNA concentration with 260/280nm.
8. Store all the RNA extracts at -80°C.

Precautions and Disclaimer:

This product and procedure described are intended for R&D use only. Purchase of this product does not convey a license to perform any patented process.

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