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Product information

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Rapid Animal Total RNA Extraction Kit

Catalog #: AT4181 / AT4182
Size: 50 preps / 250 preps
Storage: 4°C *

*: Check storage conditions for more details.

Product Description:

This kit uses a reagent similar to TRIzol for rapid RNA isolation from animal tissue, cells, and some of the plant tissue.

Features:

- Rapid and handy: the whole procedure takes approximately 10 minutes.
- Very low genomic DNA contamination (no detectable by RTPCR).
- Flexible, ideal for most animal tissue and cell culture.

Components:

Components	AT4181	AT4182
Solution A	50 ml	250 ml
Protocol	1	1

NOTE: Care must be taken when working with RNA. It is important to maintain an RNAse-free environment starting with RNA sample preparation and continue through purification and analysis. Use RNAse free tubes, tips, gels. Wear gloves at all times.

Procedures:

1. Preparation
   A. Estimate the amount of Solution A to be used. For 1 ml of Solution A, the maximum amount of tissue to be used is 100 mg; the maximum amount of cells to be used is 1×10^7.
   B. Prepare enough chloroform, isopropanol, 75% ethanol and RNase-free water.

2. Homogenization
   A. *For adherent cells*
   Discard the culture medium, add 1 ml of Solution A to ~10 cm^2 cultured cell, and mix gently by pipetting.
   B. *For suspension cells*
   Collect cells by centrifugation, discard the supernatant and add 1 ml of Solution A to 1-5 $\times 10^6$ cells, and mix gently by pipetting. The amount of cells should not exceed 1×10^6 for fibroblasts or carcinoma cell.
   C. *For fresh animal tissue*
   Cut the tissue into pieces; add 1 ml of Solution A to 50-100 mg tissue, homogenate for 30 seconds using a homogenizer.
   D. *For samples in RNAlocker*
   Remove the liquid on the surface prior to cut, and then treat as fresh tissue.

3. Fraction Separation
   A. Add 0.2 volume of chloroform to the lysate, (eg. 0.2 ml chloroform for 1 ml lysate), vortex for 30 seconds.
   B. Centrifuge at no more than 12,000 x g (13,000-15,000 rpm) for 3 minutes at room temperature, deposit may form at the bottom of the centrifuge tube.
   C. Transfer the supernatant into a new 1.5 ml centrifuge tube.
   D. Repeat step 3 for 1-2 times for tissues rich in lipid.
4. Precipitation
   A. Add equal volume of isopropanol to the supernatant, vortex for 30 seconds.
   B. Centrifuge at no more than 12,000 x g (13,000-15,000 rpm) for 3 minutes at room temperature, deposit may form at the bottom of the centrifuge tube.
   C. Discard the supernatant carefully.
5. Primary Wash
   A. Add 1 ml of 75% ethanol to the RNA deposit, vortex for 30 seconds.
   B. Centrifuge at no more than 7,500 x g (13,000-15,000 rpm) for 3 minutes at room temperature.
   C. Discard the supernatant carefully.
6. Secondary Wash
   A. Repeat step 5 once.
   B. Short spin at no more than 7,500 x g (13,000 rpm), remove the residual ethanol carefully.
7. Dissolving the RNA
   A. Incubate the tube with lid open at room temperature for 1-2 minutes.
   B. Add 50-100 µl of RNase-free water to dissolve the pellet, store RNA solution at -80°C
8. RNA integrity, yield and purity can be analyzed via electrophoresis and spectral analysis.

Storage:

Transport at room temperature, store all components at 4°C upon received.