Data sheet

Tissue Total RNA Purification Kit

Cat. No: AN0150 (50 reactions)
Cat. No: AN0152 (100 reactions)

Description

Tissue Total RNA Purification Kit offers a rapid and convenient method for purification of total RNA from a variety of tissue and culture cells. The kit is based in nucleic acid ability to bind silica in the presence of high concentrations of chaotropic salts. Tissue samples can be efficiently homogenized in a microcentrifuge tube using the provided micropestle. Eluted purified RNA is ready for use in a variety of downstream applications: real-time RT-PCR, Northern Blotting, cDNA library construction, etc.

Features

- **High yields**: up tp 50µg; depends on type of sample.
- **Ready to use** RNA.
- **Just a few minutes** procedure (about 30 min).
- **Mini format**

Quality Certifications

Total RNA is isolated from a 30 mg thorax muscle tissue sample. Purified RNA is quantified using a spectrophotometer with a typical yield of more than 10µg of total RNA and a A260nm/A280nm ratio of 1.9-2.1. Quality is further checked by agarose gel electrophoresis.

Kit Components	(Reactions)	
Item	50	100
Buffer BLY*	25 ml	50 ml
Wash Buffer 1 (WB1)	30 ml	60 ml
Wash Buffer 2 ** (WB2)	15 ml	30 ml
RNase-free ddH_2O	10 ml	10 ml
RNAprep spin column	50	100
Filter Column	50	100
Collection tube (2mL)	100	200
1.5 ml microtube	50	100
Micropestle	50	100

Note

*Before beginning the lysis and homogenization steps, prepare a fresh amount of Buffer BLY containing 1% 2-mercaptoethanol (β-ME) [Not included] for each purification procedure. Add 10 µL β-ME for each 1 mL Lysis Buffer

**Add the volume ethanol (96%-100%) specified [Not included] to WB2 Buffer prior to initial use (see bottle label for volume). After ethanol has been added, mark the bottle to indicate that this step has been completed.

Kit Storage:

Tissue Total RNA Purification Kit can be stored at room temperature. The kit components are stable for 1 year, if stored properly.

Please wear gloves when using this product. Avoid all skin contact with kit reagents. In case of contact, wash thoroughly with water.
β-ME is toxic; dispense in a fume hood and wear appropriate protective clothing.

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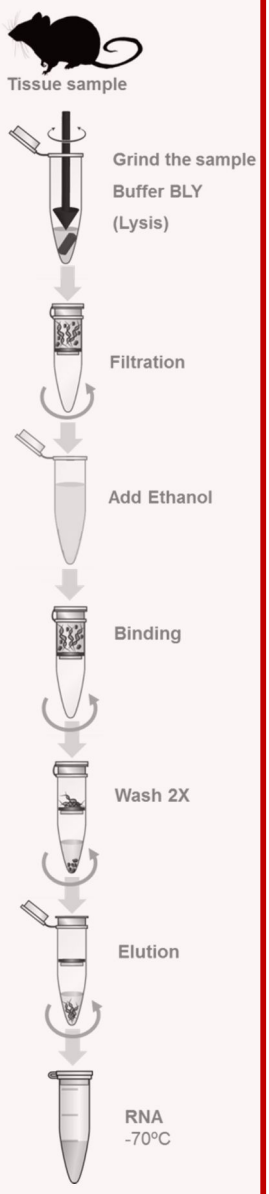
DETAILED PROTOCOL

1. Cut up to 30mg of animal tissue and transfer to a 1.5-ml microcentrifuge tube (not provided). Use the **micropestle** to grind the material to pulp. You can grind the tissue sample in liquid nitrogen.
2. Add 350µl of **Buffer BLY** (β-ME added) and continue to homogenize the sample by grinding. In order to release all RNA in the sample, it is required to disrupt the sample completely.
3. Incubate at room temperature for 5 minutes.
4. Place a **Filter Column** in a 2 ml **Collection tube** and transfer the sample mixture to the filter column. Centrifuge at full speed for 2 minutes.
5. Carefully transfer the clarified filtrate to a new 1.5 ml microcentrifuge tube.
6. Add 1 volume of 70% ethanol to the clarified lysate and mix vigorously by vortexing.
7. Apply the total volume (usually 700 µl) from step 6 to the **RNAprep spin column** by decanting or pipetting.
8. Centrifuge at full speed for 90 seconds. Discard the flow-through.
9. Wash the **RNAprep spin column** by adding 250 µL **WB1** and centrifuging at 10000 g for 90 seconds. Discard the flow-through.
10. [Optional] Place the **RNAprep spin column** in a **collection tube** and add 60 µL of RNase-free DNase I solution (0.5U/µl) (not provided) to the centre of the column matrix. Let stand for 15 minute at room temperature.
11. Add 500 µl of **WB1** and centrifuge at full speed for 30 seconds. Discard the flow-through.
12. Add 750 µl of **WB2** and centrifuge at full speed for 1 minute. Discard the flow-through.
13. Again Centrifuge at full speed for 3 minutes. This step helps to dry the **RNAprep spin column**.
14. Place the **RNAprep spin column** into a new, labelled 1.5 microcentrifuge tube and pipet 50-60µl of **RNase-free Water** directly into the. Close the cap and incubate for 1 minute at room temperature.
15. Centrifuge at full speed for 1 minute to elute RNA.
16. Keep eluted RNA on ice at all times and store at <-70°C.



PRODUCT USE LIMITATION : This product is developed, designed and sold exclusively for research purposes and *in vitro* use only. The product was not tested for use in diagnostics or for drug development, and is not suitable for administration to humans or animals. Please refer to www.canvaxbiotech.com for the Material Safety Data Sheet of the product.