WizPrep™ Total RNA Mini Kit (Tissue)

- W72070-50 **50 prep**
- W72070-150 **150 prep**

Description

The WizPrep™ Total RNA Mini Kit (Tissue) provides a fast and simple method to isolate total RNA from various animal tissue.
The WizPrep™ Total RNA Mini Kit (Tissue) uses silica-membrane technology to eliminate the cumbersome steps associated with loose resins or slurries. The kit is ready for use and can purify the total RNA from a wide variety of animal tissue samples, and the whole process is completed in less than 20 minutes.
Purified RNA is suitable for RT-PCR, northern blotting, primer extension and cDNA library construction.

Kit Contents

Contents	50 prep	150 prep	Storage
RL Buffer	30 ml	90 ml	R/T
W1 Buffer	30 ml	90 ml	R/T
W2 Buffer(1) (concentrate)	14 ml	44 ml	R/T
RNase-Free Water	5 ml	15 ml	R/T
Spin Columns*	50	150	R/T
Filter Columns*	50	150	R/T
Collection Tubes (2.0ml)	100	300	R/T
Instruction Manual	1	1	

(1) : Add absolute ethanol to the W2 Buffer prior to initial use (see the bottle label for volume).
* All Spin Columns are sterilized by electron beam.
• R/T : Room temperature

Reagents and equipment to be supplied by user

- 96~100% ethanol (to prepare W2 Buffer)
- 1.5 ml microcentrifuge tubes
- Sterile RNase-free pipette tips and Manual pipettors
- Centrifuge for microcentrifuge tubes
- Equipment for sample disruption and homogenization
- Personal protection equipment (lab coat, gloves, goggles)

Kit specifications

Parameter	Characteristics
Format	Silca-membrane spin column
Sample materials	< 25 mg tissue
Typical yield	5 ~ 35 µg (depending on sample)
Elution volume	50 µl
Preparation time	< 20 minutes

Quality Control Analysis

The kit was qualified by isolating total RNA from 20 mg of animal tissue following the protocols outlined in the manual.

Quality Authorized by : Jamie Ahn

Protocol

Before starting ;
1) Add absolute ethanol to the W2 Buffer prior to initial use (see the bottle label for volume).
2) If a precipitate has formed in RL Buffer, dissolve by incubating at 56°C before use.

Step 1. Preparation animal tissue (< 25 mg), for spleen (< 10 mg)

- Cut up to 25 mgs of animal tissue (or 0.5 cm of mouse tail) then transfer it to a 1.5 ml tube.

NOTE : If tissue has a higher number of cells (e.g. spleen or liver), reduce the starting material to 10 mgs.

- Add **500 µl of RL Buffer, 5 µl of β-mercaptoethanol** and homogenize the sample tissue by grinding.
- Incubate at room temperature for 5 min.
- Transfer the homogenized sample to Filter Column (violet color).
- Centrifuge at 13,000 rpm for 1 min.
- Discard the Filter column.

Step 2 : Binding step

- Add **400 µl of 70% Ethanol** to filterate (collection tube) and mix by pipetting 5 times.
- Connect Spin Column to Collection tube.
- Transfer 800 µl of the mixture to the Spin Column and centrifuge at 13,000 rpm for 1 min.
- Discard the flow-through and re-connect with the Spin Column.

(Optinal) DNA residue degradation

Add 100µl of DNase I solution (2U/µl) in center of Spin Column matrix and incubate at room temperature for 10 min.

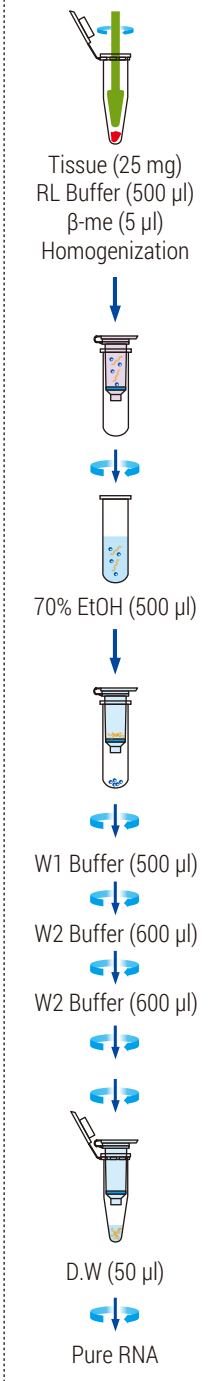
Step 3 : Wash Step

- Add **500 µl of W1 Buffer** to the Spin Column and centrifuge at 13,000 rpm for 1min then discard the flow-throw.
- Add **600 µl of W2 Buffer** (ethanol added) in the Spin Column and centrifuge at 13,000 rpm for 1 min then discard the flow-throw.
- Add **600 µl of W2 Buffer** (ethanol added) in the Spin Column and centrifuge at 13,000 rpm for 1 min then discard the flow-throw.
- Centrifuge at 13,000 rpm for 3 min.

Step 4 : Elution Step

- Connect the Spin Column and new 1.5 ml tube.
- Add **50 µl of RNase-Free Water** into the center of Spin Column and incubate at room temperature for 1 min.
- Centrifuge at 13,000 rpm for 3 min.
- Discard the Spin Column.
- Eluted RNA are stored at –20 °C for a few days, –70 °C for long term storage.

Quick Protocol



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Troubleshooting

Problem	Solution and Explanation
The Spin column is clogged	• Inefficient disruption and/or homogenization • Too much starting material • Centrifugation temperature was too low (should be 20-25ºC)
Low yield of RNA	• Insufficient disruption and homogenization • Too much starting material • RNA still bound to RNA spin column membrane • Ethanol carryover
RNA Degradation	• Harvested animal tissue not immediately stabilized • Inappropriate handling of starting material • RNase contamination