

NUCQX® Magnetic Tissue Total RNA Kit

Cat. no. LM101101

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Product Information

Product name	NUCQX® Magnetic Tissue Total RNA Kit
Cat.No.	LM101101
Size	20T/50T/100T
Storage and shipping	Part I: Store and transport at room temperature. Shelf life 1 year. Part II: Store at -20°C, and transport with ice bag. Shelf life 1 year

2. Product Description

This product is designed for the extraction of total RNA from animal tissues (e.g., liver, kidney, spleen). Utilizing uniquely formulated magnetic beads and carefully optimized buffers, it efficiently captures released nucleic acids, resulting in high-purity RNA of consistent quality. The extracted RNA is suitable for applications such as RT-PCR, Northern blotting, and in vitro translation. When used in conjunction with automated nucleic acid extraction instruments, this product enables high-throughput nucleic acid extraction.

3. Components

Form	Cat NO.	Component	Size		
			20T	50T	100T
Part I	LM101101-A	Lysis Buffer	10mL	25 mL	50mL
	LM101101-B	Binding Buffer	12mL	30mL	60mL
	LM101101-C	Wash Buffer A	14mL	35mL	70mL
	LM101101-D	Wash Buffer B	10mL	24 mL	48 mL
	LM101101-E	Elution Buffer	1mL×2	5mL	10mL
	LM101101-F	Magnetic Bead Suspension	0.4mL	1mL	1mL×2
Part II	LM101101-G	DNase I	60µL	150µL	300µL
	LM101101-H	DNase I Reaction Buffer (10×)	0.2mL	0.5mL	1mL
	LM101101-I	PT Solution	90µL	225µL	450µL

4. Instructions for Use

Reagent Preparation

- For Wash Solution B, please add the corresponding anhydrous ethanol according to the label instructions before use.
- Before operation, add PT solution to the lysis buffer to a final concentration of 1%. For example, add 10 µL of PT solution to 1 mL of lysis buffer. Once prepared, the solution can be stored at 4 °C for one week.
- The PT solution is frozen at -20 °C . After dissolving at room temperature, there may be trace amounts of crystallization. Gently vortex to mix, and use the solution after it has completely dissolved.

Sample Preparation

- **Tissue Samples:** Add 450 µL of lysis buffer (ensure PT solution has been added) to a centrifuge tube. Use liquid nitrogen to grind the sample into a powder. Weigh 10-15 mg of the sample and add it to the lysis buffer. Vortex to mix. If there is any precipitate, use a pipette to gently resuspend. Let the mixture sit at room temperature for 5 minutes to obtain the pretreated sample.
- **Suspended Cell Samples:** Take no more than 5 million cells. Centrifuge at low speed to remove the supernatant. Add 450 µL of lysis buffer to the centrifuge tube and vortex to mix. Let it sit at room temperature for 5 minutes to obtain the pretreated sample.
- **Adherent Cell Samples:** Use trypsin to digest the adherent cells and centrifuge at low speed to remove the supernatant. Add 450 µL of lysis buffer to the centrifuge tube and vortex to mix. Let it sit at room temperature for 5 minutes to obtain the pretreated sample. If the cell count is less than 1×10^6 , you can directly remove the medium from the culture plate and add 450 µL of lysis buffer. Vortex to mix and let it sit at room temperature for 5 minutes to obtain the pretreated sample.

1. Manual Extraction Steps

- 1.1. Add 550 µL binding buffer to the preprocessing sample and vortex for 5 seconds to mix.
- 1.2. Add 20 µL magnetic bead suspension and vortex for 5 seconds to mix. Place the tube on a rotary shaker or vortex mixer and mix for 5 minutes at room temperature.
- 1.3. After incubation, briefly centrifuge the tube, place it on a magnetic rack until the beads are fully captured, and carefully discard the liquid.
- 1.4. Add 700 µL wash buffer A to the tube, vortex for 2 minutes to mix thoroughly, then place the tube on the magnetic rack until the beads are fully captured, and carefully discard the liquid.
- 1.5. Add 700 µL wash buffer B (ensure that wash buffer B contains no ethanol), vortex for 2 minutes to mix thoroughly, then place the tube on the magnetic rack until the beads are fully captured, and carefully discard the liquid.

- 1.6. Briefly centrifuge to remove any residual liquid, then leave the tube open on the magnetic rack at room temperature to air dry for 5 minutes, ensuring no ethanol remains on the beads.
- 1.7. Prepare DNase digestion solution (should be prepared in advance to avoid over-drying of beads in step 6 :
 - For standard samples such as liver, kidney, spleen, and heart: 10 µL 10x buffer + 3 µL DNase I + 87 µL DEPC water, gently mix by pipetting. The digestion solution can be stored briefly at 4 °C (no more than 30 minutes).
 - For small samples such as brain tissue: 10 µL 10x buffer + 0.3 µL DNase I + 89.7 µL DEPC water, gently mix by pipetting. The digestion solution can be stored briefly at 4 °C (no more than 30 minutes).
- 1.8. Add 100 µL DNase digestion solution to the tube, gently invert or pipette to disperse the beads. Incubate at room temperature for 15 minutes, gently mixing every 5 minutes.
- 1.9. Add 700 µL wash buffer B to the tube, vortex for 2 minutes to mix thoroughly, then place the tube on the magnetic rack until the beads are fully captured, and carefully discard the liquid.
- 1.10. Repeat step 1.9.
- 1.11. Briefly centrifuge to remove any residual liquid, then leave the tube open on the magnetic rack at room temperature for 5 minutes to allow ethanol to evaporate completely. The beads should not be overly dry; slight cracking is acceptable. (Note: Do not overdry the beads as this will reduce the elution efficiency. If the room temperature is too low, you may extend the drying time appropriately.)
- 1.12. Add 50-100 µL elution buffer to the tube, place it in a thermomixer at 60 °C, 1600 rpm, and shake for 5 minutes. Then place the tube on the magnetic rack until the beads are fully captured, transfer the eluted liquid to an RNase-free tube, and avoid transferring any magnetic beads. Store the nucleic acid solution at -80 °C.

2. Automated Extraction Steps with AP-96N Nucleic Acid Extractor

- 2.1. Add reagents as follows. Before adding, ensure that wash buffer B contains no absolute ethanol. The DNase digestion solution should be prepared fresh as needed, following the procedure in step 7 of the manual extraction steps.

Plate	Reagents
1	Binding Plate, add 550 µL binding buffer to each well
2	Wash A Plate, add 700 µL wash buffer A to each well
3	Magnetic Bead Plate, add 700 µL wash buffer B and 20 µL magnetic bead suspension to each well
4	DNase Digestion Plate, add 100 µL DNase digestion solution to each well
5	Wash B Plate, add 700 µL wash buffer B to each well Plate
6	Elution Plate, add 50-100 µL elution buffer to each well

- 2.2. Perform the sample preprocessing steps to obtain the processed sample, then add it to Plate 1 and pipette 2-3 times to mix thoroughly.

2.3. Correctly place the 96-magnetic rod rack into the nucleic acid extractor and load the reagent-added 96-well plates into the extractor.

2.4. Set the program according to the table below and start the operation. After the program finishes, the solution in the elution plate will be the nucleic acid solution. If storage is required, transfer the solution to a clean RNase-free centrifuge tube and store it at -80 °C .

AP-96N Nucleic Acid Extraction Instrument Program

Steps	1	2	3	4	5	6	7	8	9
Position	3	1	2	3	4	6	5	5	3
Waiting Time	00:00:00	00:00:00	00:00:00	00:00:00	00:05:00	00:00:00	00:03:00	00:00:00	00:00:00
Mixing Mode	2	1	2	2	3	2	2	2	2
Mixing Time	00:00:20	00:05:00	00:02:00	00:02:00	00:15:00	00:02:00	00:05:00	00:00:00	00:00:10
Pause	NO	NO	NO	NO	NO	NO	NO	NO	NO
Magnetic Separation Time	00:00:45	00:01:30	00:00:45	00:00:45	00:01:30	00:00:45	00:00:45	00:00:45	00:00:00
Volume (μL)	700	1000	700	700	100	700	100	100	700
Temperature	---	25	---	---	---	---	65	65	---
Mixing Mode	M1	Mixing time 10 s, mixing speed 300,000 (instrument-defined units)							
	M2	Mixing time 10 s, mixing speed 200,000 (instrument-defined units)							
	M3	Mixing time 10 s, mixing speed 50 (instrument-defined units)							

3. Automated Extraction Steps with AP-48N Nucleic Acid Extractor

3.1. Add reagents as follows. Before adding, ensure that wash buffer B contains no absolute ethanol. The DNase digestion solution should be prepared fresh as needed, following the procedure in step 7 of the manual extraction steps.

Well Position	Reagent
Wells 1/7	Add 550 μL of Binding Buffer to each well
Wells 2/8	Add 700 μL of Wash Solution A to each well
Wells 3/9	Add 700 μL of Wash Solution B and 20 μL of Magnetic Bead Suspension to each well
Wells 4/10	Add 100 μL of DNA Enzyme Digestion Solution to each well
Wells 5/11	Add 50-100 μL of Elution Buffer to each well
Wells 6/12	Add 700 μL of Wash B

3.2. Follow the sample preprocessing steps to obtain the processed sample. Add it to wells in column 1/7, and pipette 2-3 times to mix thoroughly.

3.3. Correctly place the 8-magnet rod set into the magnetic rod rack of the nucleic acid extractor, and then properly place the 96-well plate with added reagents into the nucleic acid extractor.

3.4. Set the program according to the table below and start the operation. After the program finishes, the solution in the elution wells (columns 5 and 11) will be the nucleic acid solution. If storage is required, transfer the solution to a clean RNase-free centrifuge tube and store it at -80 °C.

AP-48N Nucleic Acid Extraction Instrument Program

Steps	1	2	3	4	5	6	7	8	9
Position	3	1	2	3	4	6	5	5	3
Waiting Time	00:00:00	00:00:00	00:00:00	00:00:00	00:05:00	00:00:00	00:03:00	00:00:00	00:00:00
Mixing Mode	2	1	2	2	3	2	2	2	2
Mixing Time	00:00:20	00:05:00	00:02:00	00:02:00	00:15:00	00:02:00	00:05:00	00:00:00	00:00:10
Pause	NO	NO	NO	NO	NO	NO	NO	NO	NO
Magnetic Separation Time	00:00:45	00:01:30	00:00:45	00:00:45	00:01:30	00:00:45	00:00:45	00:00:45	00:00:00
Volume (μL)	700	1000	700	700	100	700	100	100	700
Temperature	---	25	---	---	---	---	65	65	---
Mixing Mode	M1	Mixing time 10 s, mixing speed 300,000 (instrument-defined units)							
	M2	Mixing time 10 s, mixing speed 200,000 (instrument-defined units)							
	M3	Mixing time 10 s, mixing speed 50 (instrument-defined units)							

4. Automated Extraction Steps with AP-16S Nucleic Acid Extractor

4.1. Add reagents to the 96-well deep plate according to the 48-channel liquid addition method.

4.2. Follow the sample preprocessing steps to obtain the processed sample, and add it to wells in columns 1/7. Pipette 2-3 times to mix thoroughly.

4.3. Correctly place the 8-magnet rod set into the magnetic rod rack of the nucleic acid extractor, and properly place the 96-well plate with added reagents into the nucleic acid extractor.

4.4. Set the program according to the table below and start the operation. After the program finishes, the solution in the elution wells (columns 5 and 11) will be the nucleic acid solution. If storage is required, transfer the solution to a clean RNase-free centrifuge tube and store it at -80 °C .

AP-16S Nucleic Acid Extraction Instrument Program

Steps	1	2	3	4	5	6	7	8	9
Position	3	1	2	3	4	6	5	5	3
Waiting Time	00:00:00	00:00:00	00:00:00	00:00:00	00:05:00	00:00:00	00:03:00	00:00:00	00:00:00

Mixing Mode	3	2	3	3	5	3	3	3	3
Mixing Time	00:00:20	00:05:00	00:02:00	00:02:00	00:15:00	00:02:00	00:05:00	00:00:00	00:00:10
Pause	NO	NO	NO	NO	NO	NO	NO	NO	NO
Magnetic Separation Time	00:00:45	00:01:30	00:00:45	00:00:45	00:01:30	00:00:45	00:00:45	00:00:45	00:00:00
Volume (µL)	700	1000	700	700	100	700	100	100	700
Temperature	---	25	---	---	---	---	65	65	---
Mixing Mode	M1	Mixing time 10 s, mixing speed 300,000 (instrument-defined units)							
	M2	Mixing time 10 s, mixing speed 200,000 (instrument-defined units)							
	M3	Mixing time 10 s, mixing speed 50 (instrument-defined units)							

5. NOTES

1. If any components precipitate or appear cloudy (especially in low-temperature environments, such as winter), heat the solution to 45 °C until it becomes clear.
2. Magnetic beads may remain during elution; when aspirating the elution solution, avoid aspirating the magnetic beads as much as possible.
3. For frozen samples, avoid repeated freeze-thaw cycles, as this may degrade the quality of the nucleic acids in the sample.
4. For your safety and health, please wear a lab coat and disposable gloves while operating.