

RNA Extraction Kit (Blood, Tissue, Cell)

Catalog no.: DB9824

(50, 100 prep)

Intended for research use only

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Kit Components

No.	Name	cat #: DB9864 (50,100)
1	<u>Handbook protocol</u>	1
2	Columns and Collection Tubes (pcs)	50, 100
3	BW buffer (in case of blood samples)	50, 100 ml
4	RNLy Buffer	40, 80 ml
5	RN Wash Buffer	40, 80 ml
6	Elution Bufer	5, 10 ml

Required Reagent

- 1. Chloroform
- 2. Ethanol 96%

Procedure

1. In case of blood: add 1m BW buffer in 2 ml vial and add 500 μ l of blood. Invert several times and incubate at RT for 5 min. centrifuge it at 13000 rpm and discard the supernatant. For tissue: using a scalpel cut the tissue into very small pieces on a sterilized dish. Transfer 20-40 mgr (for liver and spleen 20 mgr is enough) or 1-2×10 6 cell (for cell culture) to a 1.5 ml tube and add 800 μ l of RNLy Buffer.



- 2. Pipetting the sample to avoid clump. In case of hard tissues, you can use homogenizer on ice. Incubate at RT for 5 min.
- 3. Add 150 μ l of chloroform to the mixture, and mix thoroughly by shaking. Incubate for 3 min at room temperature.
- 4. Centrifuge at 12000 rpm for 12 min at 4° C.
- 5. Transfer **450** μ l of upper phase in a new 1.5 ml tube. Add **400** μ l of 96% ethanol and mix it.
- 6. Place the column in a collection tube and transfer mixture to the spin column. centrifuge for 1 min at 12000 *rpm*.
- 7. Discard flow-through. Add 700 μ l of RN wash buffer to the spin column and centrifuge at 12000 rpm for 1 min. (optional: to gain more pure RNA, repeat the wash step).
- 8. Place back the column in the collection tube and centrifuge for 2 min at 12000 *rpm* to dry the spin filter.
- 9. Place the spin column into a clean 1.5 ml tube and apply 50 μ l Elution Buffer or DEPC water to the center of the membrane and wait for 1 min.
- 10. Close the lid and centrifuge at **12000** x g for 1 min.
- 11. Store eluted RNA at -70° C.