CatchGene® Viral RNA Kit

Cat. No.	Rxn
MR12004	4
MR12050	50
MR12250	250

Kit Content

	4rxn	50rxn	250rxn	
Spin Columns	4	50	250	pcs
Collection Tubes (2 ml)	12	150	750	pcs
Buffer AE	1	6	30	ml
Carrier RNA	12x2	140x2	1350	µg
Buffer VRL	2	25	125	ml
Buffer RW1 (concentrated)	3.36	42	105x2	ml
Buffer RW2 (concentrated)	0.68	8.4	42	ml
RNase-Free H_2O	0.96	12	60	ml

Kit Storage

Upon arrival,

1. Please store **Carrier RNA** at **-20 ℃** for long term storage.

Buffer, solvent and consumables, please store at 15-25 ℃.

Kit Preparation

1. **Prepare 1 µg/µl Carrier RNA**
   For 12 µg Carrier RNA, please add 12 µl Buffer AE into the bottom of tube and mix thoroughly for dissolving.
   For 140 µg Carrier RNA, please add 140 µl Buffer AE into the bottom of tube and mix thoroughly for dissolving.
   For 1350 µg Carrier RNA, please add 1350 µl Buffer AE into the bottom of tube and mix thoroughly for dissolving.
   After dissolving, please aliquot into smaller volume and store at -20°C. Do not freeze-thaw more than three times.
2. **Prepare Buffer RW1**
   Add equal volume of 100% EtOH into Buffer RW1 (concentrated) to get Buffer RW1.
   After adding 100% EtOH, please check the sticker on the bottle and close the cap tightly.
3. **Prepare Buffer RW2**
   Add 4 volume of 100% EtOH into Buffer RW2 (concentrated) to get Buffer RW2.
   After adding 100% EtOH, please check the sticker on the bottle and close the cap tightly.

Sample Pretreatment

For whole blood

1. Centrifuge whole blood at 3,000 x g for 10 minute at room temperature.
2. Transfer 100 µl upper serum or plasma layer for following purification.

For nasopharyngeal swab with transport medium

1. Close the cap properly and vortex for 15 sec.
2. Transfer 100 µl clear supernatant for following purification. (Avoid to aspirate any debris or mucus)

For cultured cell

1. Collected cells depend on cells growing type.
2. Harvested the cells grown in suspension by centrifuging for 5 min at 300 x g. Carefully remove supernatant. (cell number recommended not more than 1 x 10^7).
3. Add 100 µl Buffer AE and add 5 µl Carrier RNA (1 µg/µl) into the tube then proceed to **step 3 of general protocol** quickly.

General Protocol

1. Add 5 µl Carrier RNA (1 µg/µl) into the 1.5 ml micro-centrifuge tube.
2. Transfer 100 µl liquid sample into the tube.
3. Add 350 µl Buffer VRL (add 1% β-mercaptoethanol freshly), vortex vigorously for 30 sec, brief spin down then incubate at 25°C (room temperature) for 5 min.
4. Add 250 µl of 100% EtOH, close the cap and mix thoroughly by vortex for 15 sec, brief spin down.
5. Transfer all mixture to Spin Column (with 2ml Tube).
6. Centrifuge at 11,000 x g for 1 min, and change a new collection tube.
7. Add 700 µl Buffer RW1 into spin column, centrifuge at 11,000 x g for 1 min, discard the flow-through.
8. (Optional) On column digest of DNA with DNase I (not provided).
9. Add 700 µl Buffer RW1 into spin column, centrifuge at 11,000 x g for 1 min, discard the flow-through.
10. Add 700 µl Buffer RW2 into spin column, centrifuge at 11,000 x g for 1 min, discard the flow-through.
11. Change a new collection tube, centrifuge at 11,000 x g for 3 min.
12. Place the Spin Column into 1.5 ml micro-centrifuge tube, add 30-100 µl RNase-Free H_2O and incubate at 25°C (room temperature) for 2 min.
13. Centrifuge at 11,000 x g for 1 min for elution.

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