

Kit Content

	2rxn	30rxn	
LV Module (with 50ml tube)	2	30	set
MC21 Column	2	30	pcs
Collection Tube (2ml)	4	60	pcs
Buffer RCL1	0.66	9.9	ml
Buffer RCL2	0.22	3.3	ml
Buffer CRW1 (concentrated)	1.32	20	ml
Buffer CRW2 (concentrated)	0.62	9.3	ml
RNase-Free H ₂ O	0.5	5	ml

Kit Storage

Upon arrival,

1. Please store **MC21 Column** at 4°C for long term storage.
2. Buffer, solvent and consumables, please store at 15-25 °C.

Kit Preparation

1. Prepare Buffer CRW1

Add 4 volume of 100% EtOH into concentrated Buffer CRW1 to get Buffer CRW1.
After adding 100% EtOH, please tick the sticker on the bottle and close the cap tightly.

2. Prepare Buffer CRW2

Add 4 volume of 100% EtOH into concentrated Buffer CRW2 to get Buffer CRW2.
After adding 100% EtOH, please tick the sticker on the bottle and close the cap tightly.

Sample Pretreatment

The half life of cfRNA in whole blood is very short. So, after sampling by blood tube, please must perform pretreatment as soon as possible.

For Whole Blood Sample

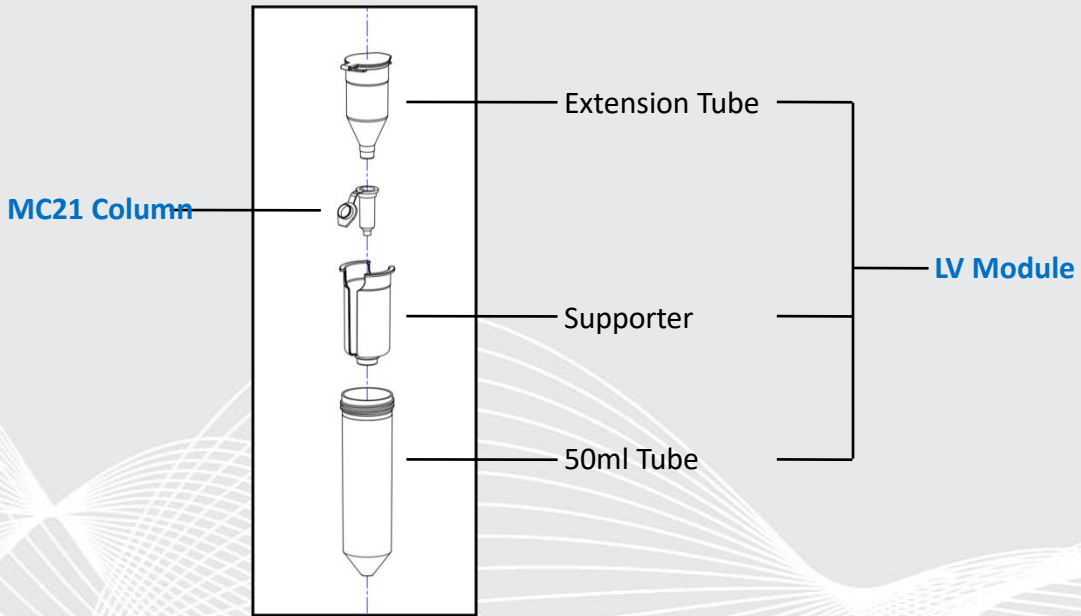
1. Centrifuge whole blood at 3,000 x g for 10 minute at room temperature.
2. Transfer upper layer to the 1.5 ml micro-centrifuge tube (not provided). Please avoid aspirating any cell debris or WBC, and intermediate layer, otherwise will co-extract cell total RNA form intact cell
3. Centrifuge at 11,000 x g for 10 min and transfer the supernatant for following extraction.

Please keep samples into -20°C or -70°C if extraction won't be performed immediately after pretreatment.

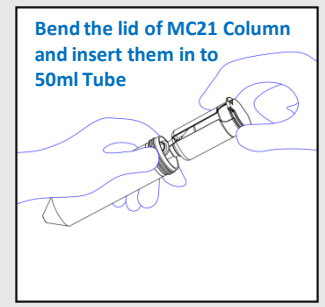
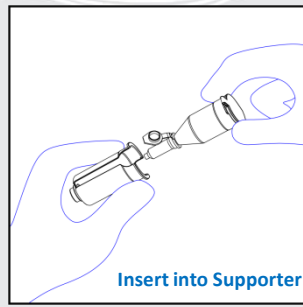
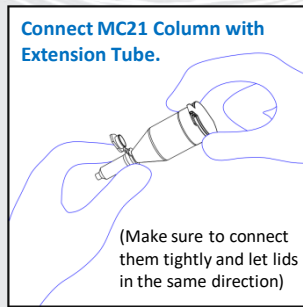
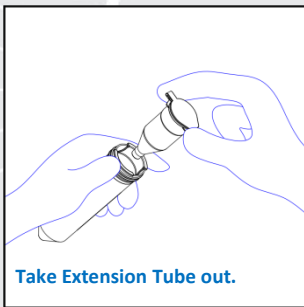
General Protocol

1. Pipette 1000 µl serum/plasma sample into 1.5 ml micro-centrifuge tube (not provided) and add 300 µl Buffer RCL1. Pulse-vortexing for 10 sec, brief spin down then incubate at 25°C (room temperature) for 3 min.
2. Add 100 µl Buffer RCL2, pulse-vortexing for 10 sec, brief spin down then incubate at 25°C (room temperature) for 1 min.
3. Centrifuge at 11,000 x g for 3 min.
4. Transfer clear supernatant to a new 15 ml centrifuge tube, add 1400 µl Isopropanol (not provided), pulse-vortexing for 10 sec then briefly spin down.
5. Connect LV Module with MC21 Column to become LV Column Module. Please refer to the illustration in next page. Transfer all lysate into LV Column Module, incubate at 25°C (room temperature) for 2 min.
6. Centrifuge at 2,700 x g for 3 min, discard the flow-through.
7. Add 3ml CRW1 Buffer into LV Column Module, centrifuge at 2,700 x g for 3 min, discard the flow-through.
8. Take LV Column Module out of 50 ml tube. Disconnect the MC21 Column from the LV Module, then place the MC21 Column on a 2 ml Collection Tube. Please refer to the illustration in next page.
9. Add 700 µl CRW2 Buffer into spin column, centrifuge at 11,000 x g for 1 min, discard the flow-through.
10. Add 700 µl CRW2 Buffer 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 30-50 µl RNase-Free H₂O and incubate at 25°C (room temperature) for 2 min.
13. Centrifuge at 11,000 x g for 1 min for elution.

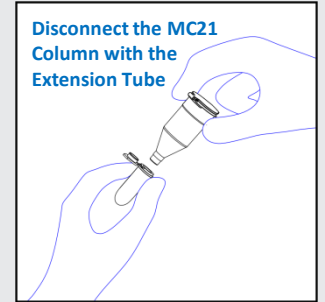
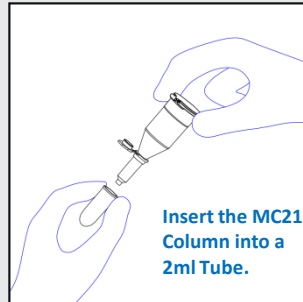
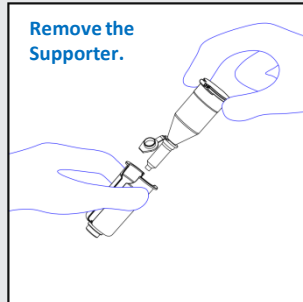
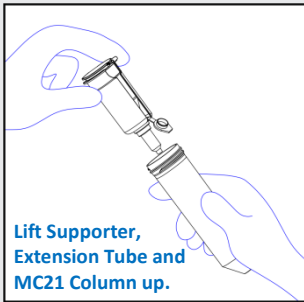
FOR RESEARCH USE ONLY



Connect LV Module with MC21 Column



Disconnect MC21 Column from LV Column Module



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