

Kit Content

	2rxn	50rxn	
LV Module (with 50ml tube)	2	50	set
Spin Column	2	50	pcs
Collection Tubes (2 ml)	4	100	pcs
Buffer RC	30	185x4	ml
Buffer RL	3	75	ml
Buffer RB	3	40x2	ml
Buffer RW1 (concentrated)	4	100	ml
Buffer RW2 (concentrated)	0.3	7.5	ml
RNase-Free H ₂ O	0.5	8	ml

Kit Storage

Upon arrival,
 1. Please store **Buffer RC** at 4 °C for long term storage.

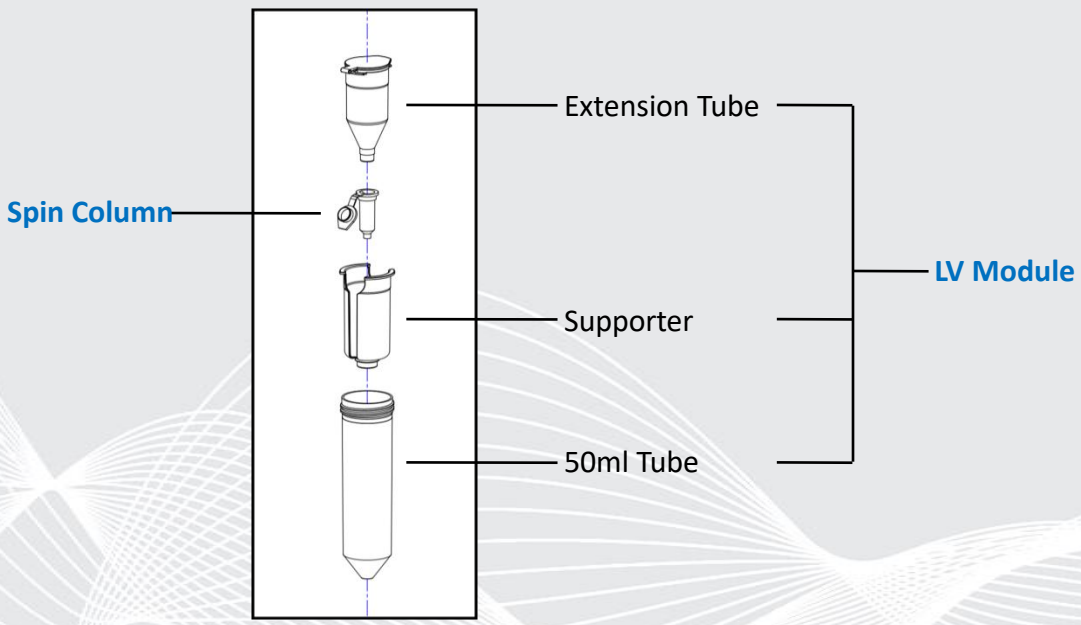
Other buffer, solvent and consumables, please store at 15-25 °C.

Kit Preparation

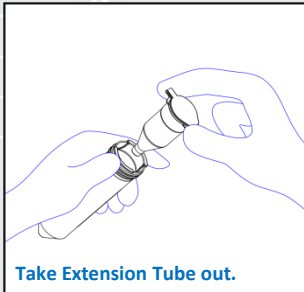
- 1. Prepare Buffer RB**
 Add 2.4 volume of 100% EtOH into Buffer RB and vortex thoroughly. After adding 100% EtOH, please tick the sticker on the bottle and close the cap tightly.
- 2. Prepare Buffer RW1**
 Add equal volume of 100% EtOH into Buffer RW1 (concentrated) to get Buffer RW1. After adding 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.

General Protocol

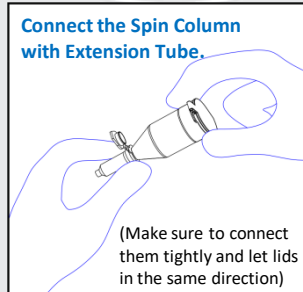
- Pipette 2 ml fresh whole blood sample into 50 ml tube and add 10 ml Buffer RC, mix well by inversion.
- Incubate on ice for 15 min. (Mix 2 times by inversion during incubation.)
- Centrifuge at 400 x g for 10 min at 4 °C to form a cell pellet and discard the supernatant completely.
- Add 4 ml of RC Buffer to res-suspend the cell pellet and mix well by inversion.
- Centrifuge at 400 x g for 10 min at 4 °C to form a cell pellet and discard the supernatant completely.
- Add 1400 µl Buffer RL (add 1% β-mercaptoethanol freshly), vortex vigorously for 30 sec, brief spin down then incubate at 25 °C (room temperature) for 5 min.
- Add 1400 µl Buffer RB, vortex 10 sec, brief spin down
- Connect LV Module with the Spin Column to become LV Column Module. Please refer to the illustration in next page.
- Transfer all lysate into LV Column Module, centrifuge at 2,700 x g for 2 min, discard the flow-through.
- Add 3ml RW1 Buffer into LV Column Module, centrifuge at 2,700 x g for 2 min, discard the flow-through.
- Take LV Column Module out of 50 ml tube. Disconnect the Spin Column from the LV Module, then place the Spin Column on a 2 ml Collection Tube. Please refer to the illustration in next page.
- (Optional) On column digest of DNA with DNase I (not provided).
- Add 700 µl Buffer RW1 into spin column, centrifuge at 11,000 x g for 1 min, discard the flow-through.
- Add 700 µl Buffer RW2 into spin column, centrifuge at 11,000 x g for 1 min, discard the flow-through.
- Add 700 µl 100% EtOH into spin column, centrifuge at 11,000 x g for 1 min, discard the flow-through.
- Place spin column on a new 2 ml Collection Tube, centrifuge at 11,000 x g for 3 min to eliminate any remaining EtOH.
- Place spin column on a new 1.5 ml micro-centrifuge tube. Add 30-100 µl RNase-Free H₂O, incubation at room temperature for 5 min, and then centrifuge at 11,000 x g for 1 min for elution.



Connect LV Module with the Spin Column

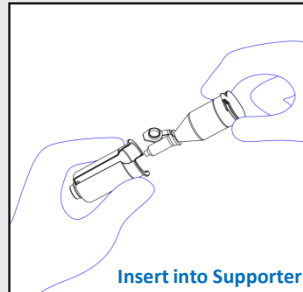


Take Extension Tube out.

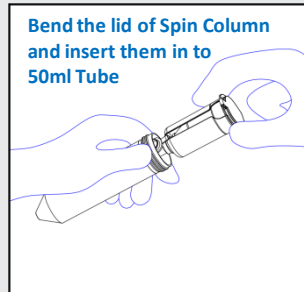


Connect the Spin Column with Extension Tube.

(Make sure to connect them tightly and let lids in the same direction)

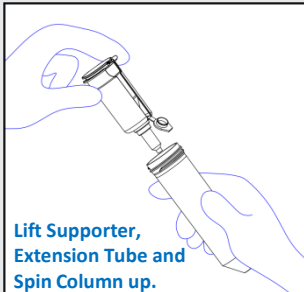


Insert into Supporter

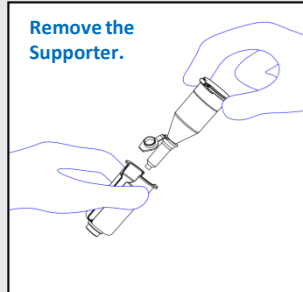


Bend the lid of Spin Column and insert them in to 50ml Tube

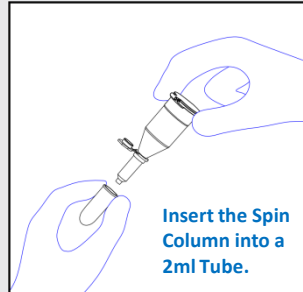
Disconnect Spin Column from LV Column Module



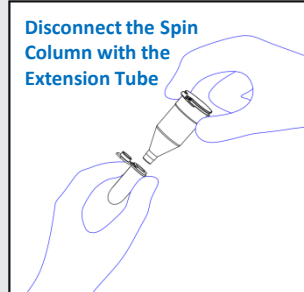
Lift Supporter, Extension Tube and Spin Column up.



Remove the Supporter.



Insert the Spin Column into a 2ml Tube.



Disconnect the Spin Column with the Extension Tube

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