

Kit Content

	2rxn	30rxn	
LV Module (with 50ml tube)	2	30	set
Spin Column	2	30	pcs
Collection Tubes (2 ml)	4	60	pcs
Buffer AE	0.5	2	ml
Carrier RNA	12	140, 12x2	µg
Proteinase K	1x2	11x3	mg
TVL Buffer	2.4	36	ml
Buffer RW1 (concentrated)	4.5	66	ml
Buffer RW2 (concentrated)	0.34	5	ml
RNase-Free H ₂ O	0.48	7.2	ml

Kit Storage

Upon arrival,
 1. Please store **Proteinase K and Carrier RNA** at **-20 °C** for long term storage.
 Buffer, solvent and consumables, please store at 15-25 °C.

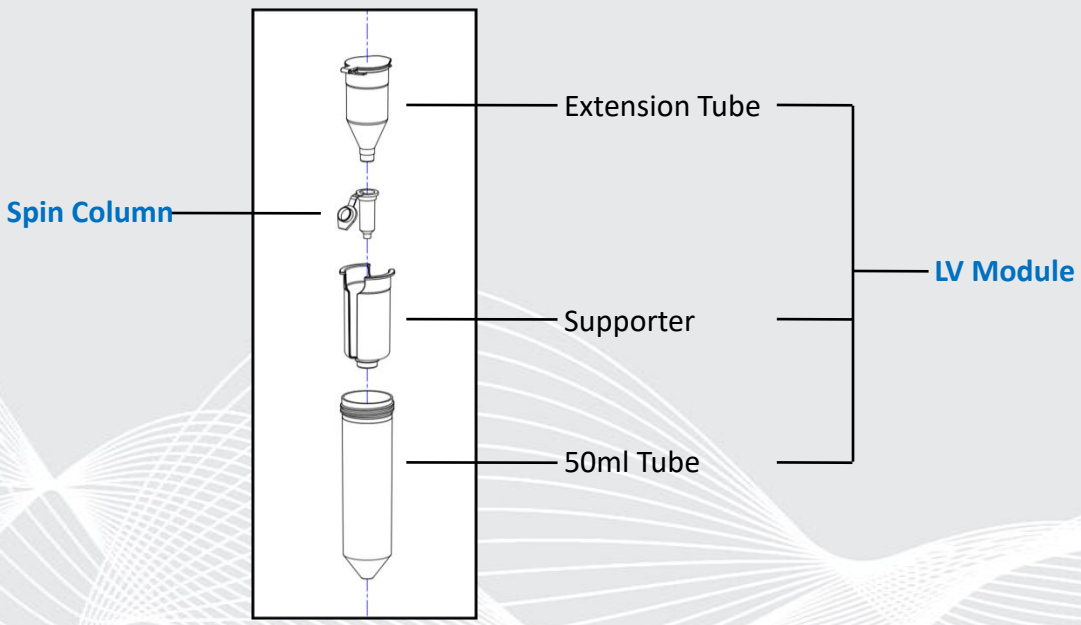
Kit Preparation

- 1. Prepare 20 mg/ml Proteinase K**
 For 1 mg Proteinase K, please add 50 µl AE Buffer into tube and vortex thoroughly for dissolving.
 For 11 mg Proteinase K, please add 550 µl AE Buffer into tube and vortex thoroughly for dissolving.
 After dissolving into the solvent, please store at 4°C for 6 month or -20°C for 1 year.
- 2. Prepare 1 µg/µl Carrier RNA**
 For 12 µg Carrier RNA, please add 12 µl AE Buffer into the bottom of tube and mix thoroughly for dissolving.
 For 140 µg Carrier RNA, please add 140 µl AE Buffer 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.
- 3. 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.
- 4. 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.

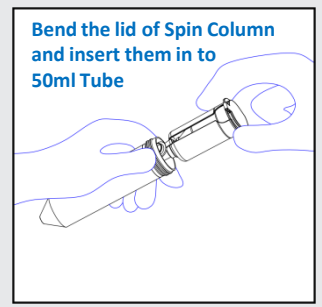
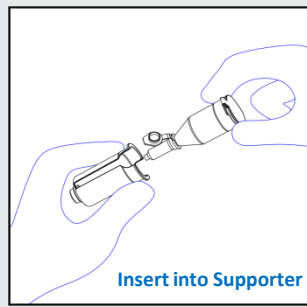
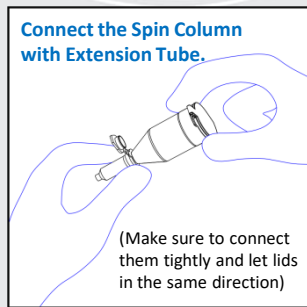
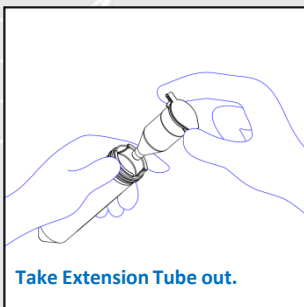
General Protocol

1. Add 50 µl Proteinase K (20 mg/ml) into a 15 ml tube (not provided).
2. Add 5 µl Carrier RNA (1 µg/µl) into the 15 ml tube.
3. Transfer 1000 µl serum or plasma sample into the tube. Close the cap, vortex for 5 sec then brief spin down.
4. Add 1000 µl TVL Buffer into the tube. Close the cap, vortex vigorously for 15 sec then brief spin down.
5. Incubate at 56 °C for 15 min, then cool down to room temperature.
6. Add 1000 µl of 100% EtOH, close the cap and mix thoroughly by vortex for 15 sec, brief spin down.
7. Connect LV Module with the Spin Column to become LV Column Module. Please refer to the illustration in next page.
8. Transfer all lysate into LV Column Module, centrifuge at 2,700 x g for 2 min, discard the flow-through.
9. Add 3ml RW1 Buffer into LV Column Module, centrifuge at 2,700 x g for 2 min, discard the flow-through.
10. 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.
11. Add 700 µl Buffer RW1 into spin column, centrifuge at 11,000 x g for 1 min, discard the flow-through.
12. Add 700 µl Buffer RW2 into spin column, centrifuge at 11,000 x g for 1 min, discard the flow-through.
13. Place spin column on a new 2 ml Collection Tube, centrifuge at 11,000 x g for 3 min to eliminate any remaining EtOH.
14. 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.

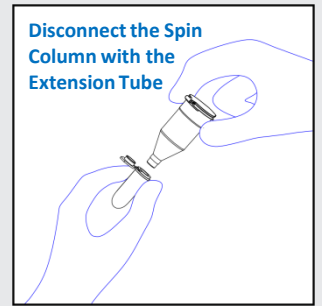
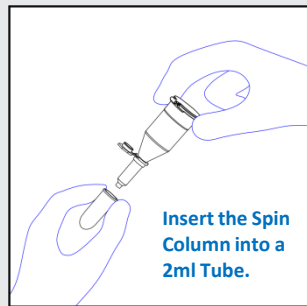
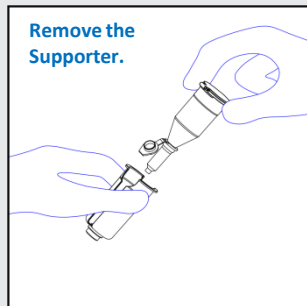
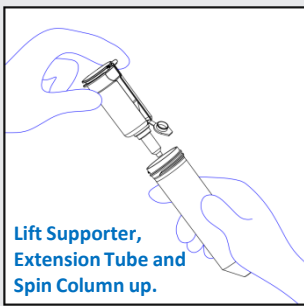
FOR RESEARCH USE ONLY



Connect LV Module with the Spin Column



Disconnect Spin Column from LV Column Module



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