

Viral Nucleic Acid (DNA/RNA) Extraction Kit I

DR-001

For isolation of viral nuleic acid from cell-free fluid such as, serum, plasma, body fluid and cell cultured supernatant

Specific for Viral RNA extraction from buccal and nasal swabs

Kit Contents:

| | DR-001s (4 preps_sample) | DR-001 (50 preps) | DR-003 (300 preps) |
|-------------------------------|-----------------------------|----------------------|-----------------------|
| VNE Buffer | 1.8 ml x 2 | 35 ml | 200 ml |
| Wash Buffer 1 * (concentrate) | 0.9 ml x 2 | 22 ml | 132 ml |
| Wash Buffer 2 * (concentrate) | 1.5 ml | 20 ml | 50 ml x 2 |
| RNase-free Water | 0.5 ml | 6 ml | 20 ml |
| Carrier RNA | | 0.4 mg | 2.2 mg |
| VNE Column | 4 pcs | 50 pcs | 300 pcs |
| Collection Tube | 8 pcs | 100 pcs | 600 pcs |
| Elution Tube | 4 pcs | 50 pcs | 300 pcs |
| User Manual | 1 | 1 | 1 |

| Cat. No: | DR-001s (4 preps) | DR-001 (50 preps) | DR-003 (300 preps) |
|----------------------------------|----------------------|----------------------|-----------------------|
| ethanol volume for Wash Buffer 1 | 0.33 ml | 8 ml | 48 ml |
| ethanol volume for Wash Buffer 2 | 6 ml | 80 ml | 200 ml |

Specification:

Principle: spin column (silica membrane)

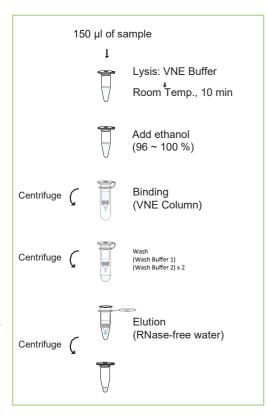
Sample: 150 µl cell-free fluid such as serum, plasma, body fluid and

cell cultured supernatant

Fragment size: 100 bp \sim 30 kb Recovery rate: 80 \sim 90 % Binding capacity: 30 ug Elution volume: 40 \sim 50 μ l Operation time: 20 min

Important Notes:

- $\hbox{1. Make sure everything is RNase-free when handling this system}.\\$
- 2. Buffers provided in this system contain irritants. Wear gloves and lab coat when handling these buffers.
- Add 1 ml of VNE Buffer to the tube of lyophilized Carrier RNA, mix well by vortexing and transfer the mixture to the VNE Buffer when first open. Store the Carrier RNA added VNE Buffer at 4 °C.
- 4. Add required ethanol (96-100%) to Wash Buffer 1 and Wash Buffer 2 before use.
- 5. Preheat RNase-free water to 70°C for elution step. (step:10)



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General Protocol:

Please Read Important Notes Before Starting Following Steps.

HINT: Preheat RNase-free water 70 °C for step 10 (elution step).

- 1. Transfer 150 µl of sample (serum, plasma, body fluids or cell cultured supernatant) into a microcentrifuge tube (not provided).
 --If the sample volume is more than 150 µl, separate it into multiple tubes.
- 2. Add 570 µl of VNE Buffer (Carrier RNA added) to the sample, mix well by vortexing, and incubate for 10 minutes at room temperature.
 - --Make sure that Carrier RNA has been added to the VNE Buffer when first use.
- 3. Add 570 µl of ethanol (96~100%) to the sample mixture, mix well by plus-vortexing.
- 4. Combine a VNE column with a Collection Tube (provided). Transfer up to 700 μl of sample mixture (ethanol added) to the VNE Column, centrifuge at 8,000 x g for 1 min then discard the flow-through. Combine the VNE Column with the used Collection Tube.
- 5. Transfer the rest of sample mixture (ethanol added) to the VNE Column, centrifuge at 8,000 x g for 1 min.

 Discard the flow-through and the Collection Tube. Combine the VNE Column with a new Collection Tube (provided).
- 6. Add 500 µl of Wash Buffer 1 (ethanol added) to the VNE Column, centrifuge at 8,000 x g for 1 min then discard the flow-through. Combine the VNE Column with the used Collection Tube.
 - --Make sure that ethanol (96~100%) has been added into Wash Buffer 1 when first open.
- 7. Add 750 µl of Wash Buffer 2 (ethanol added) to the VNE Column, centrifuge at 8,000 x g for 1 min then discard the flow-through. Combine the VNE Column with the used Collection Tube.
 - --Make sure that ethanol (96~100%) has been added into Wash Buffer 2 when first open.
- 8. Repeat step 7. Add 750 µl of Wash Buffer 2 (ethanol added) to VNE Column, centrifuge at 8,000 x g for 1 min then discard the flow-through. Combine the VNE Column with the used Collection Tube.
- 9. Centrifuge at full speed (~18,000 X g) for an additional 3 min to dry the VNE column. Discard the flow-through and the Collection Tube.
 - --Important step! This step will avoid the residual liquid to inhibit the subsequent enzymatic reactions.
- 10. Combine the VNE Column with a Elution Tube (provided). Add 50 µl of preheated RNase-free Water to the membrane center of the VNE Column. Stand VNE Column for 2 min.
 - --Important step! For effective elution, make sure that the RNase-free Water is dispensed onto the membrane center and is absorbed completely.
- 11. Centrifuge for 2 min to elute the nucleic acid.
- 12. Store nucleic acid at -70 °C.