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17-2001: Viral RNA Extraction kit (Respiratory Specimens)

Description

The Viral DNA/RNA mini kit provides an easy and reliable method for isolating total viral RNA from plasma, serum, nasopharyngeal or oropharyngeal aspirates or washes, nasopharyngeal or oropharyngeal swabs, broncheoalveolar lavage, tracheal aspirates, and sputum. This procedure has been tested for isolating nucleic acids from COVID-19, Hepatitis A, Hepatitis C and HIV. The isolated RNA can be used for PCR, qRT-PCR and other downstream applications.

Respiratory specimens including: nasopharyngeal or oropharyngeal aspirates or washes, nasopharyngeal or oropharyngeal swabs, broncheoalveolar lavage, tracheal aspirates, and sputum. Swab specimens should be collected only on swabs with a synthetic tip with aluminum or plastic shafts. Swabs with calcium alginate or cotton tips with wooden shafts are not acceptable.

Kit Components

Catalog	17-2001-50	17-2001-250
Preps	50	250
Buffer LYE	25 ml	110 ml
Proteinase K (25 mg/mL)	1 ml	4 ml
RNA Wash Buffer *	12 ml	50 ml
Buffer RB	30 mL	130 ml
L Solution	100 µl	500 µl
DEPC-Treated ddH2O	5 ml	15 ml
Mini Column with collection tubes	50	250
Collection tubes	50	250

*Add 200 mL 100% ethanol to RNA Wash Buffer before use.

Product Info

Amount : 50 Test

Storage condition : All components can be stored at room temperature (15-25°C). All kit components are guaranteed for 1 year from the date of purchase.

Application Note

The protocol is developed for 200-300 µL samples. Small samples should be adjusted to 200-300 µl with phosphate-buffered saline (PBS) before loading.

1. Pipet **20 µL Proteinase K, 2 µL L Solution,** and **300 µL Buffer LYE** to a 1.5 mL tube.

   Calculate the number of samples to be processed and make master mix of proteinase K, L Solution and Buffer LYE.

2. Pipet **300 µL** nasopharyngeal or oropharyngeal aspirates or washes, nasopharyngeal or oropharyngeal swabs, broncheoalveolar lavage, tracheal aspirates, or plasma, serum, into the 1.5 mL tube from Step 1. Mix well by pulse-vortexting for 10 seconds. Spin briefly to collect the drops from the lid. Incubate at room temperature for 10 min to lyse the cells and virus.

3. Add **600 µL isopropanol** and mix well by pulse-vortexing for 5 seconds. Spin briefly to collect the drop from the lid.

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4. Transfer **600 µL** of the sample from step 3 into a RNA column and centrifuge at 10,000 rpm for 30 seconds. Discard the flow-through carefully to a waste container by pipetting and put the column back to the collection Transfer the remaining sample to the column and centrifuge at 10,000 rpm for 30 seconds. Discard the collection tube and transfer the RNA column to a new collection tube.
5. Add **500 µL Buffer RB** to the column and centrifuge at 10,000 rm for 30 seconds. Discard the flow-through. Put the column back to the collection tube.
6. Add **500 µL RNA Wash Buffer** to the column and centrifuge at 10,000 rpm for 30 seconds. Discard the flow-through. Put the column back to the collection tube.
7. Centrifuge the empty column at 12,000 rpm for 2 min. **It is critical to remove residual ethanol for optimal elution.**
8. Transfer the RNA column to a RNase-free 5 mL tube, add 35-50 **µL DEPC-treated water** to the column and centrifuge at 10,000 rpm for 30 seconds. The viral RNA is in the flow-through liquid.
9. Optional: Add the eluent back to the column for a second elution.

   Note: The first elution normally yields 70% of the RNA while the second elution yields another 20-30% of the RNA bound to the column.

   Note: The purified RNA should be put on ice for downstream application or store at -20°C.