EpiQuik™ Viral RNA Isolation Fast Kit

Base Catalog # P-9107

PLEASE READ THIS ENTIRE USER GUIDE BEFORE USE

Uses: The EpiQuik™ Viral RNA Isolation Fast Kit is suitable for a quick preparation of viral RNA from cell-free liquid specimens (excluding plasma and serum), specifically from saliva and nasal or nasopharyngeal swabs.

Input Amount: The amount of starting materials can be up to 400 µl of liquid volume, with the best volume of 200 µl. A total of 50 standard extractions (using 200 µl of sample) can be performed with this kit.

Binding Capacity and Yield: The column binding capacity can be up to 5 µg. RNA spiking tests show that the yield is >95%. However, the yield from different samples may vary depending on the sample type.

Purity: Purified RNA is ready for most downstream applications and specifically for RT-PCR.

Precautions: To avoid cross-contamination, carefully pipette the sample or solution using aerosol-barrier pipette tips and always change pipette tips between liquid transfers. Wear gloves throughout the entire procedure. In case of contact between gloves and sample, change gloves immediately.

KIT CONTENTS

Components	50 samples P-9107-050	Shipping Temperature	Storage Upon Receipt	Storage Checklist
RLB (RNA Lysis Buffer)	10 ml	RT	RT	
RPB (RNA Purification Buffer)	7 ml	RT	RT	
RWB (RNA Wash Buffer)	6 ml	RT	RT	
EB (Elution Buffer)	2 ml	RT	RT	
F-Spin Column	50 pcs	RT	RT	
F-Collection Tubes	50 pcs	RT	RT	

SHIPPING & STORAGE

The kit is shipped at ambient room temperature. Upon receipt, store the components according to the temperatures in the table above away from light. The kit can be stable for up to 6-months from the date of shipment when stored properly.

All components of the kit are stable for 6 months from the date of shipment, when stored properly.

Note: *Check all buffers for salt precipitation prior to use. Re-dissolve any precipitate by warming up to 37°C.*

MATERIALS REQUIRED BUT NOT SUPPLIED

- ☐ Vortex mixer
- ☐ Desktop centrifuge (up to 14,000 rpm)
- ☐ Pipettes and RNase-free pipet tips
- ☐ 1.5 ml microcentrifuge tubes (RNase-free)
- ☐ Isopropanol (100%)
- ☐ Ethanol (100%)
- ☐ Physiological saline

GENERAL PRODUCT INFORMATION

Quality Control: Each lot of the EpiQuik™ Viral RNA Isolation Fast Kit is tested against predetermined specifications to ensure consistent product quality. EpigenTek guarantees the performance of all products in the manner described in our product instructions.

Product Warranty: If this product does not meet your expectations, simply contact our technical support unit or your regional distributor. We also encourage you to contact us if you have any suggestions about product performance or new applications and techniques.

Safety: Suitable lab coat, disposable gloves, and proper eye protection are required when working with this product.

Product Updates: EpigenTek reserves the right to change or modify any product to enhance its performance and design. The information in this User Guide is subject to change at any time without notice. Be sure to use the latest User Guide for this kit which can be accessed online at www.epigentek.com/datasheet.

Usage Limitation: The EpiQuik™ Viral RNA Isolation Fast Kit is for research use only and is not intended for diagnostic or therapeutic application.

A BRIEF OVERVIEW

The *EpiQuik™ Viral RNA Isolation Fast Kit* provides a fast, simple, and cost-effective method for the isolation of viral RNA from cell-free liquid specimens (excluding plasma and serum), specifically from saliva and nasal or nasopharyngeal swabs. The specialized buffering system allows RNA to bind to the glass fiber matrix of the spin column while contaminants pass through the column. Impurities are efficiently washed away, and pure RNA is eluted. The RNA purified with the EpiQuik™ Viral RNA Isolation Fast Kit is suitable for a variety of routine applications, specifically for RT-PCR.

The kit has the following features:

- Fast procedure delivers high-quality total RNA in total 10 minutes
- Ready-to-use RNA for high performance in any downstream application
- Consistent RNA yield from a small amount of starting material

PRINCIPLE & PROCEDURE

The EpiQuik™ Viral RNA Isolation Fast Kit contains all the reagents required for successfully performing RNA isolation directly from cell-free samples (excluding plasma and serum). After lysis, binding, and wash, RNA is easily recovered in quantities of up to 5 µg using specially designed columns. Total RNA is then ready to be used for a variety of downstream applications.

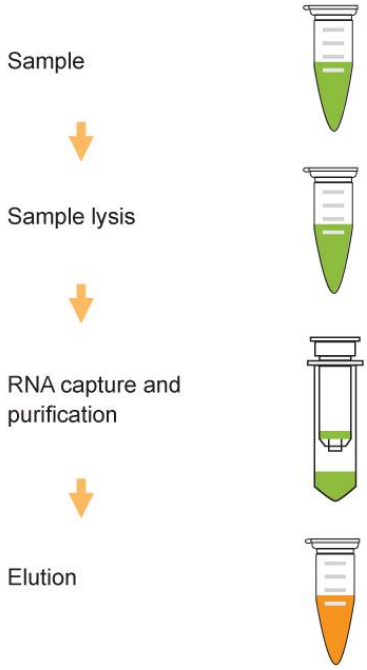

Fig 1. Schematic Procedure for the EpiQuik™ Viral RNA Isolation Fast Kit.

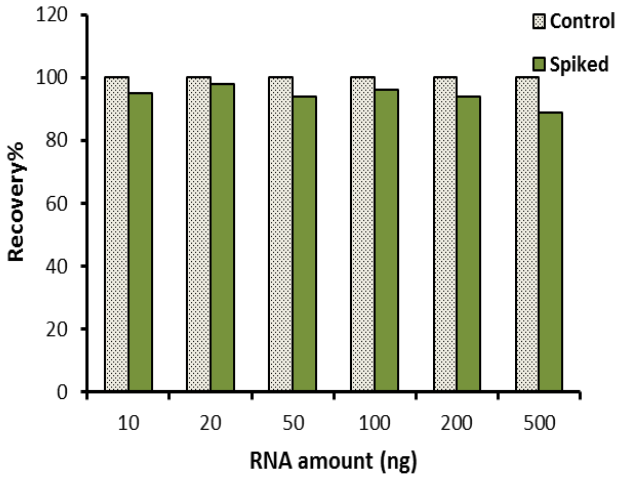

Fig 2. Quantification analysis of the isolated RNA. 200 nt RNA fragment at different concentrations was spiked into normal saline and then isolated with the EpiQuik™ Viral RNA Isolation Fast Kit. Isolated RNA was then fluorescently quantified using a fluorescent method for RNA/ssDNA measurement.

A: Positive Control Sample

B: Negative Control Sample

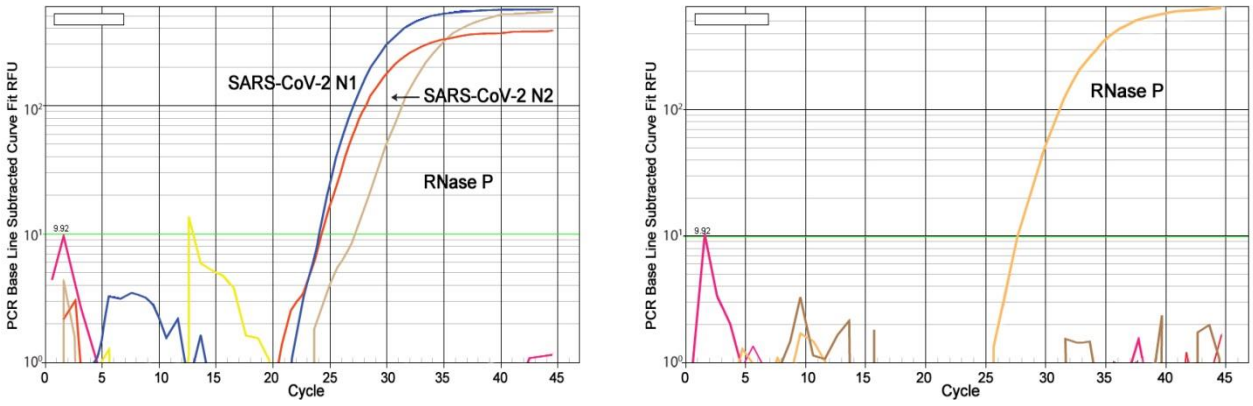

Fig 3. Total RNA isolated from nasal swab samples with EpiQuik™ Viral RNA Isolation Fast Kit and was used for RT-PCR analysis. A: Nasopharyngeal swab samples were spiked with SARS-CoV-2-N positive control; B: Nasopharyngeal swab samples were not spiked with SARS-CoV-2-N positive control. PCR was processed with use of primers and probes against SARS-CoV-2 N1/N2 and human RNase P gene (internal control).

PROTOCOL

For the best results, please read the protocol in its entirety prior to starting your experiment.

Starting Materials

Input Amount: The sample amount can be up to 400 µl volume.

Working Buffer Preparation

Working RWB (RNA Wash Buffer): Add 24 ml of 100% ethanol to 6 ml of **RWB (RNA Wash Buffer)**

1. Sample Harvesting

Liquid viral sample:

1.1a Directly collect 200 µl of liquid samples into a 1.5 ml vial.

1.2a Add 100 µl of the **RLB (RNA Lysis Buffer).** Mix by pipetting.

1.3a Incubate at room temperature for 5 min.

Swab sample:

1.1b Correctly collect nasal or nasopharyngeal swab samples according to the established swab sample collection instruction.

1.2b Place the swab into a clean 1.5 ml microtube, and snap off the handle. Add 200 µl of physiological saline, rotate the swab in saline for 30 seconds.

1.3b Remove the swab and discard. Add 100 µl of the **RLB (RNA Lysis Buffer)** to the sample.

1.4b If the transport of the swab sample is needed, the swab sample should be eluted in 200-300 µl of the transport media. And 200 µl of the transport media containing swab sample into a 1.5 ml microtube and then add 100 µl of the **RLB (RNA Lysis Buffer)** to the sample. Mix by pipetting.

1.5b Incubate at room temperature for 5 min.

2. RNA Isolation

2.1 After incubation, add 70 µl of **RPB (RNA Purification Buffer)** followed by adding 300 µl of isopropanol and mix the sample by inverting gently the tube.

2.2 Place a F-spin column into a 2 ml collection tube. Transfer the mixture to a F-spin column. Centrifuge at 14,000 rpm for 45 seconds. Remove the column from the collection tube and discard the flowthrough.

2.3 Replace column to the collection tube. Add 250 µl of working **RWB (RNA Wash Buffer)** to the column and centrifuge at 14,000 rpm for 45 seconds. Discard the flowthrough.

2.4 Replace column to the collection tube. Add 250 µl of working **RWB** (RNA Wash Buffer) to the column and centrifuge at 14,000 rpm for 1 min. Discard the flowthrough.

Note: *If more than 200 µl of sample are used, then step 1 and 2 should be repeated in increments of 200 µl of the sample and add buffers/reagents proportionally (i.e., if 400 µl of sample is being used,*

*200 µl of RLB (RNA Lysis Buffer) should be added at step 1.2a, 1.3b, or 1.4b, 140 µl of **RPB (RNA Purification Buffer)** and 600 µl of isopropanol should be added at Step 2.1. Then add half of the mixture (670 µl) to the column first and process the remaining 670 µl by repeating Step 2.2 again.*

3. Elution

Place the column in a new 1.5 ml tube (RNase-free). Add 15 µl of **EB** (Elution Buffer), depending on the amount of starting materials, directly to the column filter. Centrifuge at 14,000 rpm for 1 min to elute purified RNA.

***Note**: For high concentrated RNA, use 8 µl to elute.*

Purified RNA is now ready for use or storage at –20°C for up to 2 months and at -80°C long term.

TROUBLESHOOTING

Problem	Possible Cause	Suggestion
Degraded RNA/low integrity	RNase contaminant	Clean everything, use barrier tips, wear gloves and a lab coat, and use RNase-free tubes.
Low yields of RNA	Poor binding	Ensure sufficient volume of RPB (RNA Purification Buffer) and isopropanol is added. And ensure **working RWB (RNA Wash Buffer)** is properly prepared.
	Incorrect elution conditions	Add 8-15 µl of the **EB (Elution Buffer)** to the center of each **Column**, let it stand for 2 minutes, and centrifuge at 14,000 rpm for 2 minutes.
Inhibition of downstream enzymatic reactions	Presence of ethanol in the purified RNA	Repeat the wash step: Centrifuge at 14,000 rpm again for 2 minutes to remove the residual **RWB (RNA Wash Buffer)**.

RELATED PRODUCTS

P-9003	Methylamp™ RNA Bisulfite Conversion Kit
P-9005	EpiQuik™ m6A RNA Methylation Quantification Kit (Colorimetric)
P-9007	EpiNext™ 5-mC RNA Bisulfite-Seq Easy Kit (Illumina)
P-9008	EpiQuik™ m6A RNA Methylation Quantification Kit (Fluorometric)
P-9009	MethylFlash™ 5-mC RNA Methylation ELISA Easy Kit (Fluorometric)
P-9013	Epigenase™ m6A Demethylase Activity/Inhibition Assay Kit
P-9015	MethylFlash™ Urine N6-methyladenosine (m6A) Quantification Kit (Colorimetric)
P-9016	EpiQuik™ CUT&RUN M6A-Seq Kit
P-9018	EpiQuik™ CUT&RUN M6A Enrichment Kit
P-9108	EpiMag™ Viral RNA Isolation Kit (Magnetic Beads)
P-9109	EpiMag™ 96-Well Viral RNA Extraction Kit (High Throughput)