DNAbioTech
Life Science

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RNA Viruses Extraction Kit

Catalog no.: DB9864

(50 and 100 prep)

Intended for research use only

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Www.dnabiotch.ir

Diba Noavaran Azma Company

Customer and technical support

If you have any question, do not hesitate to ask! DNAbiotech would be highly appreciated for any comment(s).

About this user manual

It is recommended for the first time users to read the detailed protocol sections of the user manual before using these products. Experienced users, however, may refer to the Protocol-at-a-glance instead. The Protocol-at-glance is designed to be used only as a supplemental tool for quick referencing while performing the purification procedure.

Contact us at
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Contents

General description

"**DNAbiotech DB9864 Viral RNA extraction kit"** is optimized for viral RNA extraction (Specifically for **COVID-19 virus**) from **respiratory specimens**. Appropriate conditions for binding of RNA to the silica membrane of the corresponding **RNAbiotech Columns** are achieved by addition of ethanol to the lysate. The binding process is reversible and specific to nucleic acids. Washing steps efficiently remove contaminations with the "**DNAbiotech DB9864 Viral RNA extraction kit"** washing buffers.

Kit specifications

-This kit is designed for the rapid isolation of highly pure genomic Viral RNA from **respiratory specimens**.

-The kits allow purification of highly pure genomic RNA with a typical concentration of 20–40 ng per µL.

-The obtained RNA is ready-to-use for subsequent reactions like PCR, rtPCR, Southern blotting, or any kind of enzymatic reactions.

Warranty: the 1st reaction of this product may be used as free sample and if desired results do not obtained the product could be returned**.**

Quality Control

In accordance with DNAbiotech Co. Management System, each part of the "**DNAbiotech DB9864 Viral RNA extraction kit"** is tested against predetermined specifications to ensure consistent product quality**.**

Safety Note

The buffers included in "**DNAbiotech DB9864 Viral RNA extraction kit"** contain irritant which is harmful when in contact with skin or eyes, or

when inhaled or swallowed. Care should be taken during handling. Always wear gloves and eye protector, and follow standard safety precautions.

Buffer VL1 contain chaotropes agents. It can form highly reactive compounds when combined with bleach. Do NOT add bleach or acidic solutions directly to the sample-preparation waste**.**

Kit Components

No.	Name	cat #: DB9864-50rxn	cat #: DB982264-100 rxn
1	*Handbook protocol*	1	1
2	Columns and Collection Tubes (pcs)	50	100
3	Extra Collection Tubes	50	100
4	VL1 Buffer (Lysis Buffer)	12.5 ml	25 ml
5	VB2 Solution	17 ml	33 ml
7	WB1 (Concentrate)**	17 ml (12 ml ethanol should be added)	33 ml (24 ml ethanol should be added)
8	WB2 (Concentrate)**	10 ml (40 ml ethanol should be added)	20 ml (80 ml ethanol should be added)
9	Elution buffer (EB)	5 ml	10 ml

Storage condition:

Shipping: RT

Storage: RT, All kit components can be stored at room temperature (18–25 °C) and are stable up to one year.

Protocols of Extraction

Before experiment notes:

*Set an incubator, thermo block or water bath to 65 °C.
*Preheat Elution Buffer EB to 65-70 °C.
* Centrifuge speed: 8000- 14000 *x g*

Specimens to be collected

(Recommended): At minimum, **respiratory material** should be collected:

- Lower respiratory specimens:

Sputum (if produced) and/or endotracheal aspirate or bronchoalveolar lavage in patients with more severe respiratory disease.

(Note high risk of aerosolization; adhere strictly to infection prevention and control procedures).

- or **wash (5 ml Sterile Water for Injection)** in ambulatory patients

- **And/or upper respiratory specimens:**

(Not Recommended): Nasopharyngeal and oropharyngeal swab in (300µl deionized Water)

Procedure

1. Add 250 µl of VL1 Buffer to a nuclease-free 1.5 or 2 ml microcentrifuge tube. Add 200 µl nasopharyngeal and oropharyngeal swab or wash in ambulatory patients. **Mix by vigorous vortexing for 30 seconds**. (**Optional**: Incubate 10 min at 65 - 70° C and vortex every 5 min. Spin down briefly to remove any drops from inside of the lid).

2. Add 300 µl Buffer VB2 to the sample, and **mix thoroughly by vortexing for 20 seconds**. Spin down briefly to remove any drops from inside of the lid.

3. Transfer the mixture from step 2 into the column placed in a collection tube. Centrifuge for 1 min at 8000 *x g*. Discard flow-through.

4. Place the column into a new collection tube, add 500 µl Wash Buffer1, and centrifuge for 1 min at 8000 *x g*. Discard flow-through.

5. Add 600 µl Wash Buffer2, and centrifuge for 1 min at 8000 *x g*. Discard flow-through.

6. Centrifuge spin-collection at full speed (>14,000 *x g*) for 2 min to **dry the membrane completely**. Discard flow-through and collection tube.

7. Place the column into a nuclease-free 1.5 ml microcentrifuge tube. Carefully open the lid of the column and apply 40-50 µl Elution Buffer to **the center of the membrane**.

8. Close the lid and incubate 1 min (preferred at 65 - 70° C). Centrifuge at full speed (>14,000 *x g*) for 1 min. The microcentrifuge tube now contains the eluted RNA.

9. Either use 5-10 µl of the eluted RNA directly in One-step RT-PCR or store the eluted RNA at -70°C for later analysis.

Troubleshooting

Problem	Possible cause	suggestions
The yield of genomic RNA is low	*Incomplete lysis of cells*	Extend the incubation time of digestion or reduce the amount of sample volume used for lysis.
	The RNA elution is incomplete	Ensure that centrifugation at 14,000 x g is performed, to ensure that all the RNA is eluted.
The genomic RNA is sheared	*The genomic RNA was handled improperly*	Pipetting steps should be handled as gently as possible. Reduce vortexing times during mixing steps (no more than 10-15 seconds).
	Improper storage of sample	Repeated freezing and thawing of stored samples should be avoided as this may lead to decreased RNA size.
	The sample is old	Fresh samples are recommended for maximum genomic RNA yield
No band in PCR	*Optimize the concentration of template*	If the template be less or more than usual, PCR reaction won't be able to amplify the RNA target. Use a nanodrop and find the exact concentration of the RNA and apply the template as it is indicated in your PCR procedure.

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Related products:	***Some other products***
LBC kit (Liquid based cytology)	SDS-PAGE preparation kit
50 X TAE buffer	Bradford Protein assay kit
10 X TBE buffer	MTT Assay kit
5 X TBE buffer	TMB ELISA kit (3 reagents)
Ready to use PBS buffer for 500 ml and 1 liter	BCA kit (different reaction and volumes)
ECO DNA safe stain for gel (gel stain)	Ethidium bromide dropper 5ml, 10 mgr / ml solution, molecular
Load safe DNA stain (for sample)	Gel buffer stacking gel
6 X DNA loading buffer	Gel buffer Separation
Agarose (10, 25, 50 and 100 gr)	Sample bufferSDS-PAGE
DNA extraction kit from whole blood	Acryl amid/bis acryl Amid Stock Solution (30/0.8 %)
DNA extraction kit from tissue	DEPC treated water
Plasmid DNA extraction kit	PMSF
DNA extraction kit from bacteria	IPTG
DNA extraction kit from stool	RIPA buffer
Total DNA extraction kit	TrizolEX (25,50 and 100 ml)
Total RNA extraction kit	Saturated phenol
DNA extraction kit from plant	Gram staining kit
Spin columns (different types)	Diff quick kit
Collection tube	Trypcin-EDTA 0.25 100 ml
RNase A solution (10 mgr/ml)	100 X L glutamine Solution 100 ml
Proteinase K solution (20 mgr/ml)	Penicillin-Streptomycin 100X
Absolute Ethanol	DMSO for cell stock
Isopropanol (2-propanol)	DMSO for MTT assay
Taq DNA polymerase master mix	RNA stabilization solution
PFU master mix	LB agar and LB broth media
And….	*And….*

DNAbiotech is the 1st Iranian Biotechnology Company producing wide ranges of ready to use **molecular buffers**. These products are produced from the high quality ingredients and help researchers run their projects faster.

Other products & services:

- ✓ Cloning and expression of different recombinant peptides
- ✓ Gene, Primer and peptide synthesizing
- ✓ Column based DNA extraction kits from different samples
- ✓ RNA extraction kit
- ✓ Bioinformatics services
- ✓ Production of secondary antibodies (goat anti mouse, anti rabbit and anti human antibodies, HRP conjugated).
- ✓ Taq polymerase and PFU master mix
- ✓ Molecular grade buffers (TAE, TBE, RIPA and….)
- ✓ Column based DNA/RNA extraction kits.
- ✓ And ….

For more information visit us at “www.dnabiotech.ir”

More Products Launch Coming Soon!