



DNAbiotech
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RNA Viruses Extraction Kit

Catalog no.: DB9864FAST
(100 prep)

Intended for research use only

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General description

“**DNABioTech DB9864FAST Viral RNA extraction kit**” is optimized for viral RNA extraction from **respiratory specimens**. **Lysmaster vials** lead to shorter time and more safe extractions and reduce personal errors. Appropriate conditions for binding of RNA to the silica membrane of the corresponding **DNABioTech Columns** are optimized. The binding process is reversible and specific to nucleic acids. Only one washing step efficiently remove contaminations with the “**DNABioTech DB9864FAST Viral RNA extraction kit**” washing buffer.

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 RT-PCR, PCR, 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 DB9864FAST Viral RNA extraction kit**” is tested against predetermined specifications to ensure consistent product quality.

Safety Note

The buffers included in “**DNABioTech DB9864FAST 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 #: DB982264-100 rxn
1	<i>Handbook protocol</i>	1
2	Columns (pcs)	100
3	Collection Tubes	200
4	Lysmaster	100 vials into freezer box
5	VB2 Solution	33 ml
6	WB (Ready to use)	60 ml
7	Elution buffer (EB)	10 ml

Storage condition:

Shipping: RT

Storage: RT, all kit components can be stored at room temperature (18–25 °C) except LysMaster vials box that should be kept at 4 °C .

Protocols of Extraction

Before experiment notes:

- *Preheat Elution Buffer BE to 65 °C.
- * Centrifuge speed: 8000- 12000 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:

-Nasopharyngeal and oropharyngeal swab in standard VTM.

Procedure

1. Add 200 μ l of sample to a Lysmaster vial and **vortex vigorously for 30 seconds.**
2. Add 300 μ l of VB2 solution and **vortex vigorously for 20 seconds.**
3. Transfer the mixture from last step into the column placed in a collection tube. Centrifuge for 1 min at 8000 \times *g*. Discard flow-through and collection tube.
4. Place the column into a new collection tube, add 500 μ l Wash Buffer and centrifuge for 1 min at 8000 \times *g*. Discard flow-through.
5. Place back the column into the collection tube, then centrifuge at full speed (>12,000 \times *g*) for **2-3 min to dry the membrane completely.** Discard flow-through and collection tube.

Note: the filter of spin column should be dry completely otherwise it may have resulted in “invalid” response errors.

6. Place the column into a nuclease-free 1.5 ml microcentrifuge tube. Carefully open the lid of the column and apply 40-50 μ l preheat Elution Buffer (EB) to the center of the membrane.
7. Close the lid and incubate 1 min 65 °C. Centrifuge at full speed (>12,000 \times g) for 1 min. The microcentrifuge tube now contains the eluted RNA.
8. Either use 5-10 μ l (should be set up according to the RT-PCR kit) 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	<i>Incomplete lysis of cells</i>	Extend the incubation time of digestion or reduce the amount of sample volume used for lysis.
	<i>The RNA elution is incomplete</i>	Ensure that centrifugation at 14,000 \times g is performed, to ensure that all the RNA is eluted.
The genomic RNA is sheared	<i>The genomic RNA was handled improperly</i>	Pipetting steps should be handled as gently as possible. Reduce vortexing times during mixing steps (no more than 10-15 seconds).
	<i>Improper storage of sample</i>	Repeated freezing and thawing of stored samples should be avoided as this may lead to decreased RNA size.
	<i>The sample is old</i>	Fresh samples are recommended for maximum genomic RNA yield
No band in PCR	<i>Optimize the concentration of template</i>	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.

Related products:	Some other products
Splin columns for Extraction kits	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....