# HELINI Purefast Viral RNA Mini spin prep kit

Instructions for use

For use with: VTM





2002



25/50/100/250 Prep



HELINI Biomolecules, Chennai, INDIA

# Contents

Intended use	5
Kit components	5
Storage	6
Material and Instruments required	6
Product use limitations	7
Wash buffers - Preparation	9
Sample – pre requirement preparation	11
Purification protocol	12
Quality control	15

Version 1.0Page 3Version 1.0Page 4

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### **Intended Use**

The HELINI Purefast viral RNA mini spin prep kit is a spin column based rapid and cost-effective small-scale preparation of high-quality Viral RNA from Viral transport media. Purified viral RNA can be used directly in RT-PCR/PCR.

# Kit components

Components	Volume Per reaction	25 tests	50 tests	100 tests	250 tests
Carrier RNA	5µl	125μ1	250μ1	0.5ml	1.25ml
Lysis buffer	200μ1	5ml	10ml	20ml	50ml
Elution Buffer	60µl	2ml	4ml	8ml	15ml
Wash Buffer-1*	600μ1	13ml	26ml	52ml	130ml
Wash Buffer-2*	600µl	6ml	12ml	24ml	48ml
Spin columns with collection tube	1	25	50	100	250
Collection tubes	3	75	150	300	750

<sup>\*</sup>Wash buffers supplied as a concentrate. Working buffers needs to prepare before use. Please refer page.9

### **Storage**

- The kit is shipped in room temperature.
- Upon arrival, Carrier RNA should be stored in -20°C.
- Remaining consumables store at room temperature.
- They are stable until the expiration date stated on the label.
- Repeated thawing and freezing should be avoided, as this might affect the performance of the assay.

### Material and instruments required

- Ethanol [96 100%]
- Desktop centrifuge having 13000rpm or above with a rotor for
   1.5/2 ml reaction tubes
- Micro Pipettes (variables)
- Micro Pipette tips with filters (disposable)
- Powder-free gloves (disposable)

[Please ensure that all instruments used have been installed, calibrated, checked and maintained according to the manufacturer's instructions and recommendations.]

Version 1.0 Page 5 Version 1.0 Page 6

### **Product Use Limitations**

- All reagents may exclusively be used in molecular biology DNA/RNA applications.
- The product is to be used by personnel specially instructed and trained in Molecular biology experiments.
- Strict compliance with the user manual is required for optimal PCR results.
- Attention should be paid to expiration dates printed on the box and labels of all components. Do not use expired components.
- Wear protective disposable powder-free gloves, a laboratory coat and eye protection when handling specimens and kit components.
- Avoid microbial and nuclease (DNase/RNase) contamination of the specimens and the components of the kit.
- Always use DNase/RNase-free disposable pipette tips with aerosol barriers.
- Use separated and segregated working areas for sample preparation, reaction setup and amplification/detection activities.
- The workflow in the laboratory should proceed in unidirectional manner. Always wear disposable gloves in each area and change them before entering a different area.
- Store positive and/or potentially positive material separated from all other components of the kit.
- Discard sample and assay waste according to your local safety regulations.

### **Technical Assistance**

For technical assistance and more information, please contact;

0091-9382810333

0091-44-244490433

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Version 1.0 Page 7 Version 1.0 Page 8

# Wash buffers - Preparation

Add the indicated volume of ethanol (96-100%) to Wash Buffer I (concentrated) and Wash Buffer II (concentrated) prior to first use:

	Cat.No:2002– 25 prep		
	Wash buffer-1	Wash Buffer-2	
Concentrated Buffer	13ml	6ml	
Ethanol [96 – 100%] to add	9ml	16ml	
Total volume	22ml	22ml	

	Cat.No:2002 – 50 prep		
	Wash buffer-1	Wash Buffer-2	
Concentrated Buffer	26ml	12ml	
Ethanol [96 – 100%] to add	18ml	32ml	
Total volume	44ml	44ml	

	Cat.No:2002 – 100 prep		
	Wash buffer-1	Wash Buffer-2	
Concentrated Buffer	52ml	24ml	
Ethanol [96 – 100%] to add	36ml	64ml	
Total volume	88ml	88ml	

	Cat.No:2002 – 250 prep		
	Wash buffer-1	Wash Buffer-2	
Concentrated Buffer	130ml	60ml	
Ethanol [96 – 100%] to add	90ml	160ml	
Total volume	220ml	130ml	

# **Important Notes:**

All purification steps should be carried out at room temperature.

All centrifugations should be carried out in a table-top microcentrifuge at >13000 x g (12000-14000 rpm, depending on the rotor type).

# Adjustment of sample volume:

If sample volume to be used more, Scale up buffers volume accordingly.

Version 1.0 Page 9 Version 1.0 Page 10

### **Procedure:**

- 1. Transfer 200μl of Lysis buffer into sterile 1.5ml centrifuge tube.
- 2. Pipette mix 5µl of carrier RNA into lysis buffer.
- 3. Add 200µl of VTM. Mix well by pulse vortex for 15 seconds. [Option: If you are using Internal control template to monitor extraction efficiency, please add 5µl of Internal control template]
- 4. Centrifuge few seconds to bring down drops to the bottom of the tube.
- 5. Incubate in room temperature for 10min.
- 6. Add 200μl of 100% Ethanol and mix well by vortex for 20seconds. Spin down few seconds to bring down drops to bottom of the tube.
- 7. Transfer entire sample-lysis mix into the Purefast® spin column. Centrifuge at 8000rpm for 1 min. Discard the collection tube containing filtrate. Place spin column into fresh collection tube.

- 8. Add 600µl of Wash buffer-1 [Ethanol added] to the Purefast® spin column. Centrifuge at 8000rpm for 1min Discard the collection tube containing filtrate. Place spin column into fresh collection tube.
- 9. Add 600µl of Wash buffer-2 [Ethanol added] to the Purefast® spin column. Centrifuge at 10000rpm for 1min. Discard the collection tube containing filtrate. Place spin column into fresh collection tube.
- 10. Centrifuge at **13000rpm** for **2 min** [Empty spin]. This step is essential to avoid residual ethanol. Discard the collection tube with flow-through.
- 11. Transfer the Purefast® spin column into a fresh 1.5 ml microcentrifuge tube [Not provided with the kit].
- 12. Add 60μl of Elution Buffer to the centre of Purefast® spin column membrane. Incubate 2 minute at room temperature.
- 13. Centrifuge at 8000rpm for 1 min and discard the Purefast spin column. Micro centrifuge tube now contains the eluted nucleic acid. Either use the directly in PCR or store at -80°C for later analysis.

### **Recommendation for Real-time PCR:**

Use 5 -  $20\mu$ l of elute

Version 1.0 Page 11 Version 1.0 Page 12

# **Quality Control**

In accordance with the HELINI Biomolecules in house Quality Management System, each lot of HELINI Purefast viral RNA mini spin prep kit is tested against predetermined specifications to ensure consistent product quality.

# **Explanations of symbols**



In vitro diagnostic medical device



Catalogue number



Pack size – number of tests



Manufacturer

Manufactured by

## HELINI Biomolecules,

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Version 1.0 Page 13 Version 1.0 Page 14

 Version 1.0
 Page 15
 Version 1.0
 Page 16

 $\begin{tabular}{lll} \hline Version 1.0 & Page 17 & \hline \\ \hline \hline Version 1.0 & Page 18 \\ \hline \end{tabular}$ 

 Version 1.0
 Page 19
 Version 1.0
 Page 20