

HELINI™
PureFast
Nucleic acid
Minispin prep Kit

[Optimized for Shrimp biological samples]

Cat. No. 2022 - 25 Preps
Cat. No: 2023 - 50 Preps
Cat. No: 2024 - 100 Preps

Handbook

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Kit components

No. of reactions	25	50	100
Catalogue Number	2022	2023	2024
Proteinase K	0.5ml	1ml	2 x 1ml
Tissue Lysis buffer	15ml	30ml	60ml
Binding buffer	10ml	20ml	40ml
Elution Buffer	3ml	6ml	12ml
Wash Buffer-1	9ml	18ml	36ml
Wash Buffer-2	6ml	12ml	24ml
Spin columns with collection tube	25	50	100
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Storage

Kits packed in two boxes. One carton box contains Proteinase K that should be stored in -20C. Another Box contains remaining consumables that has to be store in room temperature.

Technical Assistance

For technical assistance and more information, please contact;
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Reagent 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:200216 – 25 prep	
	Wash buffer-1	Wash Buffer-2
Concentrated Buffer	9ml	6ml
Ethanol [96 – 100%] to add	6ml	24ml
Total volume	15ml	30ml

	Cat.No:200349 – 50 prep	
	Wash buffer-1	Wash Buffer-2
Concentrated Buffer	18ml	12ml
Ethanol [96 – 100%] to add	12ml	48ml
Total volume	30ml	60ml

	Cat.No:200350 – 100 prep	
	Wash buffer-1	Wash Buffer-2
Concentrated Buffer	36ml	24ml
Ethanol [96 – 100%] to add	24ml	96ml
Total volume	60ml	120ml

Safety Information

When working with chemicals, always wear a suitable lab coat, disposable gloves, and protective goggles. Discard sample and assay waste according to your local safety regulations.

Introduction

HELINI Purefast Nucleic acid Minispin prep Kit is designed for rapid and cost-effective small-scale preparation of high quality nucleic acid from tissue and biological samples. The kit utilizes an exclusive silica based membrane technology in the form of a convenient spin column. The standard procedure takes less than 30 minutes following cell lysis and yields purified viral nucleic acid. Isolated Viral nucleic acid can be used directly in RT-PCR/PCR.

Principle

Cells are lysed during a short incubation with chaotropic salt, which immediately inactivates all nucleases. Cellular nucleic acids bind selectively to special glass fibres pre-packed in the purification filter tube. Bound nucleic acids are purified in a series of rapid “wash and spin” steps to remove contaminating cellular components. Finally low salt elution releases the Nucleic acids from the glass fibre. This simple method eliminates the need for organic solvent extractions and nucleic acid precipitation, allowing for rapid purification of many samples simultaneously.

Material Required

- Micro Pipettes Variable Volume 0.5-10µl, 10-100µl, and 100-1000µl
- Sterile pipette tips with aerosol barrier 2-20µl, 10-100µl, and 100-1000µl
- Disposable powder-free gloves
- Vortex mixer / Water bath
- Centrifuge with rotor for 1.5 ml reaction tubes
- 1.5ml/2ml centrifuge tubes
- 100% ethanol

Important Notes:

All purification steps should be carried out at room temperature.

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

Note: Set water bath or Dry bath to 56°C.

Procedure:

Sample preparation

Brood stock eye stalk

Rinse the cut eye stalk with clean water. Put eye stalk into a 1.5ml tube that contains 0.6 ml tissue lysis buffer

Larvae, PL or Juvenile

Place about 10 -20 mg sample into a 1.5ml tube containing 0.6 ml Tissue lysis buffer.

Pleopod, pereopod, or gill of adult shrimp

Place 2 pieces into a 1.5ml tube containing 0.6 ml Tissue lysis buffer.

Tail or muscle of adult shrimp

Place a tail or about 20 mg muscle into a 2ml tube containing 0.6 ml Tissue Lysis buffer.

1. Grind the sample in the tube with a micro pestle. Vortex well and spin down briefly.
2. Add 20µl of Proteinase K. Mix immediately by inverting several times and spin down briefly. Incubate at 56°C for 20min.
3. Centrifuge at 8000rpm for 3min. Transfer 400µl of clear supernatant into fresh 1.5ml tube. [Avoid pipetting debris]

4. Add 400µl of Binding buffer and 5ul of Internal control template [Provided in the Real-time PCR kit]. Vortex well and incubate 2min at room temperature.
5. Add 600µl of [100%] ethanol and mix well by vortex for 30seconds. Spin down few seconds to bring down drops to bottom of the tube.
6. Pipette 700µl of sample into the PureFast® spin column. Centrifuge at 8000rpm for 1 min. Discard the flow-through and place the column back into the same collection tube.
7. Pipette remaining 700µl of sample into the PureFast® spin column. Centrifuge at 8000rpm for 1 min. Discard the flow-through and place the column back into the same collection tube.
8. Add 500µl of Wash buffer-1 [Ethanol added] to the PureFast® spin column. Centrifuge at 8000rpm for 1min and discard the flow-through. Place the column back into the same collection tube.
9. Add 500µl of Wash buffer-2 [Ethanol added] to the PureFast® spin column. Centrifuge at 8000rpm for 1min and discard the flow-through. Place the column back into the same collection tube.
10. Repeat Wash buffer-2 wash once.
11. Centrifuge empty spin column attached with collection tube at **12000rpm** for an additional **1 min**. This step is essential to avoid residual ethanol. Discard collection tube.

12. Transfer the PureFast® spin column into a fresh 1.5 ml micro-centrifuge tube (not included).
13. Add 200µl of Elution Buffer to the center of PureFast® spin column membrane. Incubate 2 minute at room temperature.
14. Centrifuge at 10000rpm for 1 min and discard the PureFast spin column. 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 2.5 - 20µl of elute

Ordering information

Kit	Pack	Cat. No
HELINI™ PureFast Nucleic acid Mini spin Prep kit [Shrimps]	25 Preps 50 Preps 100 Preps	2022 2023 2024

In association with

 **Gautham Pulavar Molecular Research**

Manufactured and Marketed by

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