Huwel Nucleic Acid Extraction Kit – Version 2.0

HL-NAX-100: 100 Exts

PI/HLNAX-05

Intended use

Huwel Nucleic Acid Extraction Kits are intended for molecular biology applications such as PCR and RT-PCR. These kits are not intended for the diagnosis, prevention, or treatment of a disease.

Background Information

The Huwel Nucleic Acid Extraction Kit is a fast, economical and easy isolation method of high purity viral RNA/DNA from Blood EDTA, nasopharyngeal or throat swabs in VTM/MTM, plasma, sputum and body fluids. The buffer system provided in the kit allows efficient lysis followed by selective binding of RNA/DNA to the spin column and recovers good yield.

Kit components

Component	Quantity	Storage
Spin Column	100	RT
Collection tube	200	RT
Huwel Lysis Buffer	40 mL	RT
Huwel Wash Buffer 1	50 mL	RT
Huwel Wash Buffer 2	2 x 50 mL	RT
Huwel Elution Buffer	20 mL	RT
Lyophilized Poly A	500 µg	-15°C to -25°C
Proteinase K	100 mg	-15°C to -25°C
Kit Protocol	1	NA

Note: Huwel Wash Buffer 1 and Huwel Wash Buffer 2 are provided with Ethanol. No need to add additional Ethanol.

Storage

- Huwel Nucleic Acid Extraction Kits are stable at room temperature (15–25°C) until the expiration date mentioned on the kit.
- **Store the Lyophilized Poly A at 4°C and Lyophilized Proteinase K at -15°C to -25°C upon receipt of the kit.**
- Reconstitute the Lyophilized Poly A in 500 µL of Huwel Elution Buffer. Prepare 50 µL aliquots in sterile vials and store at -15°C to -25°C; stable for 12 months.
- Reconstitute the Lyophilized Proteinase K in 5 mL of Huwel Elution Buffer. Prepare small aliquots according to the user requirement and store at -15°C to -25°C; stable for 12 months.

Huwel Nucleic Acid Extraction Kit

Procedure

1. To 200 µL of Huwel Lysis Buffer, add 5 µL of **Poly A** and 50 µL of **Proteinase K** in a microfuge tube
2. Add 200 µL of VTM/MTM/Plasma sample
3. Mix well and incubate for 10 min at 72°C
4. Add 100 µL of **Huwel Lysis Buffer**, vortex the tube for 5 sec and briefly spin to collect the sample droplets inside the cap
5. Load the entire sample lysate (555 µL) into Huwel spin column-collection tube assembly
6. Centrifuge at 8,000 x g for 1 min. Discard the flowthrough
7. Change the collection tube, add 500 µL of **Huwel Wash Buffer 1**
8. Centrifuge 8,000 x g for 1 min. Discard the flowthrough
9. Add 450 µL of **Huwel Wash Buffer 2**
10. Centrifuge 8,000 x g for 1 min and discard the flowthrough
11. Add 450 µL of **Huwel Wash Buffer 2**
12. Centrifuge 8,000 x g for 1 min. Discard the flowthrough
13. Change the collection tube (microfuge tube not provided), give empty spin by centrifuging at 13,000 x g for 1 min
14. Place the spin column into a fresh microfuge tube
15. Add 50/100 µL of **Huwel Elution Buffer** into the spin column
16. Incubate the spin column at RT for 5 min
17. Centrifuge 8,000 x g for 1 min
18. Store the eluate at -20°C

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Troubleshooting guide

Problem	Possible cause	Recommendation
Low nucleic acid quality or yield	Buffers stored at sub-optimal conditions	Store the kit at room temperature (15–25°C). Close reagent bottles tightly to avoid contamination and maintain stability. Store the reconstituted Poly A and Proteinase K at -15°C to -25°C.
	Incomplete mixing of sample and reagents	Mix the sample tube well after adding each reagent.
Poor elution of Nucleic acids	Drift in pH of water	If an alternate source of water or buffer is used for elution, make sure that the pH matches with the Elution buffer provided in the kit.
Low RNA yield	RNase contamination	Work in RNase-free environment. Use fresh samples or store the samples at -80°C until they can be processed. Use eluted RNA immediately for downstream applications or store at -80°C.

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