CE IVDR

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AZUL FFPE RNA EXTRACTION KIT

RNA IN 3 HOURS | GOOD YIELDS FOR USE IN PCR /SEQUENCING

PRODUCT BROCHURE

Cat No-RE115

ISO 13485 CERTIFIED

PRODUCT DESCRIPTION

The AZUL FFPE RNA Extraction Kit offers a simple and efficient solution for isolating high-quality genomic RNA from archived FFPE (formalin-fixed, paraffin-embedded) tissue samples. Extracting RNA from FFPE tissues is challenging due to the formalin-induced cross-linking of RNA strands and proteins, which affects RNA quality and makes it less suitable for many downstream applications. The AZUL FFPE RNA Extraction Kit is specifically designed to overcome these challenges by partially reversing cross-links without the need for overnight digestion. The kit also includes a xylene-free deparaffinization method, providing a safer alternative to traditional approaches. Using silica-based spin column technology, the kit eliminates the need for toxic phenol-chloroform extractions. The purified RNA is suitable for sensitive downstream applications, including qPCR and Next-Generation Sequencing (NGS).

KIT COMPONENTS

Components	For 50 preps	For 25 preps
Deparaffinization Solvent	50 mL	25 mL
Lysis Buffer(LB)	20 mL	10 mL
Proteinase K	1 mL	500 µL
Binding buffer(BB)	20 mL	10 mL
Wash Buffer 1(WB1)	25 mL	13mL
Wash Buffer 2(WB2)	25 mL	13 mL
Elution Buffer(EB)	4 mL	2 mL
Spin Column	50 (Pouch pack)	25 (Pouch pack)

SPECIFICATIONS

Format	Spin column
Sample type	FFPE samples
Equipment	Microcentrifuge
Processing time	3 Hours
Sample amount	3-8 sections of 10 µm thickness
Type	Total RNA
Sample storage	Eluted RNA should be stored at ≤ -20°C
Yield	25 - 50 µg
Purity	A260/280 ≥ 1.8 - 2.1
Kit Storage	Room Temperature Proteinase K - At -20°C
Kit Validity	Viable for 1 year if stored at appropriate conditions

NOTE: *Check the Lysis Buffer and Binding buffer for any salt precipitation before every use. Re-dissolve any precipitate by warming the solution to 37°C, then cool it back to room temperature before use.*

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FFPE RNA EXTRACTION PROTOCOL

1.Trim off excess paraffin from the sides. Cut up to 3-8 sections of 10 µm thickness. Immediately transfer the sections to a clean 2mL vial and add 1 mL of deparaffinization solvent vortex vigorously for 10 seconds.

2. Centrifuge the tube at 15,000 rpm for 2 mins.

3. Discard the supernatant without disturbing the pellet.

4. Add 1 mL of 100% ethanol to the pellet and vortex briefly.

5. Centrifuge at 15,000 rpm for 2 mins.

6. Discard the supernatant without disturbing the pellet.

7. Open the tube and incubate at room temperature or 37°C for 10 mins until ethanol has evaporated.

8. To the pellet add 400 µL of Lysis buffer (LB) and vortex for 10 seconds. Add 20 µL of Proteinase K, invert and mix the tubes well.

9. Incubate at 56°C for 1 hour.

10. Follow with an incubation at 80°C for 1 hour..

11. Allow the samples to cool down and briefly centrifuge the tubes to remove drops from inside the lid.

12. Transfer the clear lysate to a clean microfuge tube and add 400 µL Binding Buffer (BB) to the lysate and mix briefly by inverting the tube a few times.

13. Transfer the suspension to a spin column and centrifuge the tube at 14,000 rpm for 1 min at RT.

14. Discard the flow-through and place the purification column back into the collection tube. Repeat this step until complete lysate has been transferred into the column and centrifuged.

15. Wash the spin column with 500µL Wash Buffer 1 (WB1) at 14,000 rpm for 1 min and discard the flow through.

16. Add 500µL of Wash Buffer 2 (WB2) to the column and centrifuge at 14,000 rpm for 1 min to completely remove salts and impurities.

17. Centrifuge at 14,000 rpm for 1 min to prevent carryover of ethanol to the eluate.

18. Keep the purification column in a clean, sterile 1.5 mL microfuge tube and add 35µL- 50µL of Elution Buffer. Incubate at RT for 1 min.

19. Centrifuge the column for 14,000 rpm for 2 min.

20. Discard the purification column and store the eluted RNA at -20°C or -80°C until use.

Note: 100% Ethanol and DNAse are not provided with the kit.

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FLOW DIAGRAM OF FFPE RNA EXTRACTION PROTOCOL

Cut the tissues into 3-8 curls

+

1 mL of Deparaffinizing solvent, vortex

Centrifuge at 15,000 rpm for 2 mins

Remove supernatant and add 1 mL of ethanol (100%), vortex.

Centrifuge at 15,000 rpm for 2 mins

Add 400 µL of Lysis buffer

+

20 µL of Proteinase K

Incubate at 56°C for 1 hour

Incubate at 80°C for 1 hour

Transfer clear supernatant to new tube

Add 400 µL Binding Buffer

Mix well

Transfer lysate to Spin column

Centrifuge at 14,000 rpm for 1 min

Add 500 µL wash buffer 1

Centrifuge at 14,000 rpm for 1 min

Add 500 µL wash buffer 2

Centrifuge at 14,000 rpm for 1 min

Add 30-50 µL elution buffer

Centrifuge at 15,000 rpm for 2 mins

Eluted RNA

Store at -20°C or -80°C until use

TROUBLESHOOTING

PROBLEM	POSSIBLE CAUSES	SUGGESTED SOLUTIONS
Low RNA Yield	**Sample input:** Too much sample input or significantly less sample used	Use less input material or increase the volume of the Lysis Buffer and grind thoroughly.
	Incomplete paraffin removal or incomplete lysis can cause debris to clog or overload the column and leech salts into RNA eluate.	Deparaffinization step can be repeated to ensure complete removal of paraffin to ensure complete depataffinzation. Be sure to centrifuge and pellet any debris and transfer the supernatant while avoiding any pellet debris.
Low RNA Purity(A260/A280)	Improper sample handling results in ethanol or salt contamination.	Make sure lysate and wash buffers have passed entirely through the matrix of the column. This may require centrifuging at a higher speed or longer time.
DNA Contamination	DNase treatment was not done.	**To remove DNA:** Perform DNase A treatment post incubation (not provided in the kit.
RNA Degradation	Too much exposure to air or incubation at higher temperature for longer period.	**To prevent RNA degradation:** FFPE tissue sections should not be exposed to air. Immediately after sectioning, Deparaffinization solvent wash should be performed. Incubation at higher temperature for longer period of time should be avoided. Use DEPC-treated tips and tubes to enhance the yield and quality of RNA.

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ORDERING INFO

CATALOG NO	PRODUCT	PREP
RE101	AZUL SARS- CoV-2 Kit RNA Extraction Kit	25/50 preps
RE102	AZUL Tissue RNA Extraction Kit	25/50 preps
RE103	AZUL Bacterial RNA Extraction Kit	25/50 preps
RE104	AZUL Plant RNA Extraction Kit	25/50 preps
RE105	AZUL Soil RNA Extraction Kit	25/50 preps
RE106	AZUL Animal Cell Culture RNA Extraction Kit	25/50 preps
RE107	AZUL Blood RNA Extraction Kit	25/50 preps
RE108	AZUL Stool RNA Extraction Kit	25/50 preps
RE109	AZUL Saliva RNA Extraction Kit	25/50 preps
RE113	AZUL Microbiome RNA Extraction Kit	25/50 preps
RE114	AZUL Fungal RNA Extraction Kit	25/50 preps
RE115	AZUL FFPE RNA Extraction Kit	25/50 preps

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FEEDBACK

How did this kit perform?

Did AZUL Extraction Kit fulfill expectations required for your research?
Let us know by filling out the feedback form here

Or scan the QR code:

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