

INTENDED USE

AZUL Stool RNA Extraction Kit is an easy and efficient system for the isolation of high-quality RNA (human and bacterial) from fresh, frozen stool or from stool samples stored in stabilization solution.

SUMMARY AND EXPLANATION

This kit uses a silica-based spin column technology for isolating RNA from biological samples, thereby eliminating toxic phenol-chloroform extractions. The eluted RNA is suitable for all sensitive downstream applications such as qPCR and Next-Generation sequencing.

PRODUCT FEATURES

- Rapid purification of high-quality, ready-to-use RNA.
- No organic extraction or alcohol precipitation.
- Consistent and high yields.
- Complete removal of contaminants and inhibitors for reliable results.
- Kit formats for low- to high-throughput – options for automation of all kits.

PRECAUTIONS

- AZUL Stool RNA Extraction kits are intended for use as supplied. Do not dilute or add other components to the AZUL Stool RNA Extraction kit.
- Dispose of used reagents, debris, and consumables as hazardous waste according to established laboratory procedures.
- Use RNase-free plasticware/reagents and aerosol-barrier pipet tips for better yields.

DIRECTIONS FOR USE

1. Collect and weigh >200 mg of fresh, frozen stool samples or transfer >200 µL stool samples stored in stabilization solution to a clean microfuge tube and add 1 mL of Stabilization Buffer (SB).
2. Centrifuge the tube at 10,000 rpm for 5 mins.
3. Discard the supernatant, to the pellet add 500 µL-700 µL of Extraction Buffer (EB), and 25 µL of Lysis Buffer 1 (LB1).
4. Mix briefly by vortexing for 30 secs.
5. Add 20 µL of Lysis Buffer 2 (LB2) to the mixture and mix well. Incubate the tubes at 37°C for 10-15 mins or until the solution turns clear.
6. Add 25 µL of Proteinase K to the tube and incubate at 56°C for 10 mins with intermittent vortexing every 5 mins.
7. Centrifuge the tube at 15,000 rpm for 15 mins at RT. Transfer the clear supernatant to a new microfuge tube.
8. Add 600 µL Binding Buffer (BB) to this suspension and mix briefly by inverting the tube a few times.
9. Transfer the suspension to a spin column and centrifuge the tube at 15,000 rpm for 2 mins at RT.
10. 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.
11. Wash the spin column with 600 µL Wash Buffer 1 (WB1) at 15,000 rpm for 1 min and discard the flow through.
12. Add 500 µL of Wash Buffer 2 (WB2) to the column and centrifuge at 15,000 rpm for 1 min to completely remove salts and impurities.
13. Keep the purification column in a clean, sterile 1.5 mL microfuge tube and add 30 µL- 50 µL of Elution Buffer or DNase/RNase-free water to the center of the column.
14. Centrifuge the column for 15,000 rpm for 2 mins.
15. Discard the purification column and store the eluted RNA at -20°C or -80°C until use.

KIT COMPONENTS

Components	For 50 preps	For 25 preps
Extraction Buffer	35 mL	20 mL
Lysis Buffer 1 (LB1)	2 mL	1 mL
Lysis Buffer 2 (LB2)	1 mL	0.5 mL
Stabilization Buffer (SB)	50 mL	25 mL
Proteinase K	1.3 mL	750 µL
Binding Buffer (BB)	30 mL	15 mL
Wash Buffer 1 (WB1)	30 mL	15 mL
Wash Buffer 2 (WB2)	25 mL	15 mL
Elution Buffer (EB)	4 mL	2 mL
Spin Column	50 (Pouch pack)	25 (Pouch pack)

CAUTION

- Check the Binding Buffer and Lysis 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.
- During operation, always wear a lab coat, disposable gloves, protective goggles and mask.

KIT STORAGE AND STABILITY

- Store the kit at room temperature, Proteinase K and Lysis Buffer 2 at -20°C.
- Viable for 1 year if stored at appropriate conditions.

ORDERING INFORMATION

Please call us at +91 8088747968 or mail at hello@azooka.life for any queries or assistance.

Additional information can be found online at www.azooka.life

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