NIGERIAN INSTITUTE OF MEDICAL RESEARCH

DATA SHEET

Total RNA Purification Kit

For in vitro use only!

Store at room temperature

Description of Kit:

This Total RNA Purification Kit is formulated for the rapid extraction of total RNA from swab specimen and viral samples. The kit is based on silica-gel membrane adsorption for RNA purification, and the spin column allows RNA binding while removing impurities and inhibitors from the RNA. The kit does not contain phenol, hence, the use of the kit is safe and does not produce any harmful waste. The RNA extracted using this kit is suitable for several downstream applications and have been particularly tested for COVID-19 diagnosis.

Kit Content:

Lysis Buffer (before use, add 2-Mercaptoethanol as indicated in this pamphlet before use) - stable for 1 month at room temperature.

Primary Wash Buffer (provided as a concentrate. Please add absolute Ethanol as indicated in this pamphlet before use)

Secondary Wash Buffer (provided as a concentrate. Please add absolute Ethanol as indicated in this pamphlet before use)

Elution Buffer

Spin Columns with 2 ml Collection Tubes

To be provided by the user

Beta Mercaptoethanol (BME)

Absolute Ethanol

2-Propanol (Isopropanol or propan-2-ol)

1.5 ml microtubes

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NOTE:

Before use

1. **Add 10ul of Beta Mercapto Ethanol (BME) per ml of Lysis Buffer (you can add 100µl of BME to the 10ml of Lysis buffer provided**
2. **Add 2ml of absolute ethanol to the primary wash buffer**
3. **Add 8ml of ethanol to the secondary wash buffer**

PROCEDURE

1. Add the specimen into a microcentrifuge tube (For liquid specimen, add 100µl of specimen into the microcentrifuge tube)
2. Add 500µl of the Lysis Buffer (BME added).
3. Homogenize the specimen in the Lysis Buffer (you can skip this step if using a liquid specimen)
4. Vortex and incubate at room temperature for 5 minutes.
5. After incubation, add 300 µl of Isopropanol to the lysate and vortex.
6. Transfer the mixture into the spin column.
7. Centrifuge at 10,000 to 12,000 rpm for 30 sec.
8. Discard the flow-through and blot the collection tube on a tissue paper.
9. Add 700 µl of the primary wash buffer to the Spin Column.
10. Centrifuge at 10,000 to 12,000 rpm for 30 sec.
11. Discard the flow-through and blot the collection tube on a tissue paper.
12. Add 700 µl of secondary wash buffer to the Spin Column.
13. Centrifuge at 10,000 to 12,000 rpm for 30 sec.
14. Discard the flow-through and blot the collection tube on a tissue paper.
15. Centrifuge the spin column again at 12,000 to 14,000rpm for 2 mins to remove all traces of ethanol.
16. Place the Spin Column into another microcentrifuge tube.
17. Add 50 µl Elution Buffer or nuclease-free water to the centre of the column.
18. Incubate at room temperature for 1 to 2 mins
19. Centrifuge at 10,000 rpm for 1 min to elute the RNA
20. Store RNA at -20 or -80 °C.