



## VAHTS mRNA Capture Beads 2.0

Cat# NB-54-0338

Version 23.1

### Product Description

VAHTS mRNA Capture Beads 2.0 are 1  $\mu$ m Oligo(dT)-coupled paramagnetic microspheres intended for the isolation of poly(A)<sup>+</sup> RNA from purified total RNA. The magnetic separation technology allows for the isolation of intact mRNA in small sample volumes, eliminating the need for mRNA precipitation. The whole workflow can be completed in 1 h.

### Components

Components	NB-54-0338-01(24 rxns)	NB-54-0338-02(96 rxns)
Beads Binding Buffer 2.0	1.2 ml	4.8 ml
mRNA Capture Beads 2.0	1.2 ml	4.8 ml
Beads Wash Buffer 2.0	9.6 ml	38.4 ml
Tris Buffer 2.0	1.2 ml	4.8 ml
Nuclease-free ddH <sub>2</sub> O	1 ml	4 × 1 ml

### Storage

Store at 2 ~ 8°C and adjust the shipping method according to the destination.

### Applications

This product is intended for the isolation of poly(A)<sup>+</sup> RNA from 0.01 - 12.5  $\mu$ g of high-integrity total RNA (RIN $\geq$ 7). An intact or degraded total RNA template may lead to partial loss of poly(A)<sup>+</sup> RNA.

### Self-prepared Materials

Low-binding Nuclease-free PCR tubes and pipette tips, PCR instrument, and magnetic rack.

### Notes

For research use only. Not for use in diagnostic procedures.

1. Remove the reagent from 2 ~ 8°C storage and equilibrate to room temperature before use to ensure optimal capture efficiency.
2. Mix the beads thoroughly by inversion every time before pipetting, but avoid vigorous shaking.
3. Always wear gloves and use fresh Nuclease-free ddH<sub>2</sub>O during testing to avoid contamination.
4. Remove as much Wash Buffer as possible without disturbing the beads to ensure optimal performance.

## Experiment Process

•Perform the following steps at room temperature to ensure capture efficiency.

1. Remove the reagent from 2 - 8°C storage and equilibrate to room temperature.
2. Prepare the RNA sample: Dilute 0.01 - 12.5 pg of total RNA to 50 µl with Nuclease-free ddH<sub>2</sub>O in an Nuclease-free PCR tube and keep the tube on ice for later use.
3. Mix the mRNA Capture Beads 2.0 thoroughly by inversion. Pipette 50 µl of the beads into the total RNA sample, and mix thoroughly by pipetting up and down 6 times.
4. Load the sample into the PCR instrument. Incubate at 65°C for 5 min, 25°C for 5 min, and hold at 4°C to allow the mRNA to bind to the beads.
5. Place the sample on a magnetic rack for 5 min to isolate mRNA from total RNA. After the solution becomes clear, carefully discard remove the supernatant.
6. Remove the sample from the magnetic rack, add 200 µl of Beads Wash Buffer 2.0, and mix thoroughly by pipetting up and down 6 times. Place the sample on the magnetic rack for 5 min. After the solution becomes clear, carefully discard remove the supernatant.
7. Remove the sample from the magnetic rack, and add 50 µl of Tris Buffer 2.0 to resuspend the beads. Mix thoroughly by pipetting up and down 6 times.
8. Load the sample into the PCR instrument. Incubate at 80°C for 2 min and hold at 25°C to elute the mRNA.
9. Add 50 µl of Beads Binding Buffer 2.0 and mix thoroughly by pipetting up and down 6 times.
10. Incubate at room temperature for 5 min to allow the mRNA to bind to the beads.
11. Place the sample on a magnetic rack for 5 min to isolate mRNA from total RNA. After the solution becomes clear, carefully discard remove the supernatant.
12. Remove the sample from the magnetic rack, add 200 µl of Beads Wash Buffer 2.0, and mix thoroughly by pipetting up and down 6 times. Place the sample on the magnetic rack for 5 min. After the solution becomes clear, carefully discard remove the supernatant.
13. Choose an appropriate treatment method based on the subsequent procedure:
  - a. If the purified product is used for reverse transcription, remove the sample from the magnetic rack, add 10.5µl of Nuclease-free ddH<sub>2</sub>O, and mix well by pipetting up and down 6 times. Incubate at 80°C for 2 min and immediately place the tube on the magnetic rack for 5 min. After the solution is clear, carefully transfer 8 µl of the supernatant to a new Nuclease-free PCR tube.
  - b. If the purified product is used for RNA library preparation, for example, using VAHTS Universal V8 RNA-seq Library Prep Kit for Illumina (Neo Biotech# NB-54-0338), add the appropriate volume of Frag/Prime Buffer as specified in the Instructions for Use to prepare the library.
13. Place the sample on ice for NGS library preparation or other analytical applications (we recommend using the sample immediately for subsequent reactions), or store it at -85 - -65°C for later use.

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