

## RNA HS Assay kit

Product Number: RA100s

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### Shipping and Storage

1. The complete reagent kit is stored in a dark place at 2-8°C. After initial use, it is recommended to store RNA HS Reagent at room temperature and avoid light; RNA HS Buffer storage at room temperature; RNA HS Standard 1 and 2 were stored at 2-8°C.
2. Adjust transportation methods according to different destinations.

### Components

| Component                                 | RA100s   | RA100s   |
|---|----------|----------|
|   | 100preps | 500preps |
| RNA HS Reagent (200 × in DMSO)            | 250µl    | 1.25ml   |
| RNA HS Buffer                             | 50ml     | 250ml    |
| RNA HS Standard 1 (0 ng/µl in TE buffer)  | 1ml      | 5ml      |
| RNA HS Standard 2 (10 ng/µl in TE buffer) | 4×250µl  | 10×500µl |

### Description

RNA HS (High Sensitivity) Assay Kit is a simple, sensitive, and accurate RNA fluorescence quantitative detection kit. This reagent kit includes fluorescence detection reagents, buffer solutions, and RNA standards. This reagent kit has high selectivity for RNA, is not affected by dsDNA, and has excellent linear relationships with RNA samples in the 5-100 ng range. It can accurately quantify total RNA, rRNA, and mRNA samples with concentrations ranging from 250pg/µl to 100ng/µl, and has excellent tolerance to some conventional pollutants such as salt, free nucleotides, proteins, solvents, and detergents. This product is easy to operate and can be performed at room temperature. Before use, dilute the fluorescence detection reagent with buffer to a working solution, then add the RNA sample to be tested and detect it using a Qubit fluorescence meter.

### Application

Total RNA、rRNA、mRNA from 250 pg/µl to 100ng/µl

### Note

1. Fluorescent dyes have quenching issues, please try to avoid light as much as possible.
2. For detection reagents and RNA standards, mix them upside down before each use, and briefly centrifuge for 1-2 seconds to collect the reagents at the bottom of the tube.
3. To avoid degradation of RNA standards, please use RNA free consumables for the experiment and store the standards at 2-8°C after the experiment is completed.
4. To ensure the accuracy of quantitative results, please use a calibrated pipette for operation.
5. Please perform quantitative testing at room temperature. Before use, place the components in the reagent kit at room temperature. During the experiment, please do not hold the PCR tube with your hands for a long time.
6. Please make sure to complete the testing of all samples within 3 hours of preparing the working solution to avoid deviation in the results caused by fluorescence quenching.

### Protocol

The following steps apply to Qubit2.0, Qubit3.0, and Qubit4.0 fluorometers

1. Before use, balance the components in the reagent kit to room temperature.
2. Take a sufficient quantity of 0.5 ml PCR tubes for the detection of samples and standards.

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**Note: Only 0.5 ml PCR tubes can be used.**

3. Mark each PCR tube cap. **Do not** label on the side wall of the PCR tube to avoid affecting the collection of fluorescence signals.
4. Prepare working solution. Take RNA HS Reagent from the reagent kit and dilute it with RNA HS Buffer in a ratio of 1:200 to prepare the detection working solution. Prepare it for use immediately. **It is prohibited to use glass containers to prepare working solution.**

**Note: Adequate working solution should be prepared. For example, if there are 7 RNA samples to be tested and two standard samples are added, 2ml of working solution needs to be prepared. Take 10 $\mu$ l of RNA HS Reagent and add it to 1990 $\mu$ l of RNA HS Buffer. Invert and mix well, and set aside.**

5. Prepare testing standards. Take 190 $\mu$ l of the detection working solution into the standard PCR tube, add 10 $\mu$ l of Standard 1 and Standard 2 to the corresponding standard PCR tube, gently vortex and shake for 2-3 seconds to avoid the formation of bubbles as much as possible. Please ensure that the amount added to the pipette is accurate in this step.
6. Prepare testing samples. Take 180-199 $\mu$ l of working solution into the PCR tube, add 1-20 $\mu$ l of RNA samples to be tested, so that the final volume of each test sample is 200 $\mu$ l. Gently vortex for 2-3 seconds to avoid the formation of bubbles as much as possible.

**Note: The volume range of the RNA sample to be tested is 1-20 $\mu$ l; The volume range of the working solution added for testing is 180-199 $\mu$ l, with a total volume of 200 $\mu$ l.**

7. Incubate all PCR tubes at room temperature in a dark environment for 2 minutes.
8. According to the operating instructions of the Qubit fluorometer, select the RNA high-sensitivity detection program to measure the concentration.

