# **Equalbit RNA HS Assay Kit**



Version 22.1



# **Product Description**

Equalbit RNA HS (High Sensitivity) Assay Kit is a simple, sensitive, and accurate RNA fluorescence quantitative detection kit. This kit contains fluorescence detection reagents, buffers, and RNA standards. This kit is highly selective for RNA and is not affected by dsDNA. It has an excellent linearity between 5 ng and 100 ng for RNA samples, providing accurate quantification of total RNA, rRNA, mRNA samples from 250 pg/µl to 100 ng/µl. This kit has excellent resistance for most conventional contamination, including salt, free nucleotides, proteins, solvents, and detergents. It is easy to operate, and the assay can be performed at room temperature. Before use, please dilute the fluorescence detection reagent with buffer into a working solution, and then add the RNA sample for detection by a Qubit fluorometer.

# Components

Components	EQ211-01 (100 assays)	EQ211-02 (500 assays)
Equalbit RNA HS Reagent (200 × in DMSO)	250 µl	1.25 ml
Equalbit RNA HS Buffer	50 ml	250 ml
Equalbit RNA HS Standard # 1 (0 ng/µl in TE buffer)	1 ml	5 ml
Equalbit RNA HS Standard # 2 (10 ng/µl in TE buffer)	4 × 250 μl	10 × 500 μl

## **Storage**

Store at 2 ~ 8°C and protect from light.

After the first use, it is recommended to store the Equalbit RNA HS Reagent at room temperature and protect from light, store the Equalbit RNA HS Buffer at room temperature, and store the Equalbit RNA HS Standard # 1 and # 2 at 2 ~ 8°C.

Adjust the shipping method according to different destinations.

## **Applications**

It is applicable for detection of 250 pg/µl - 100 ng/µl of total RNA, rRNA and mRNA samples.

#### **Notes**

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

- 1. Be sure to protect from light due to the fluorescent dye may quench.
- 2. Mix detection reagents and RNA standards by inversion before use and centrifuge briefly to collect the reagent to the bottom of the tube for 1 2 sec.
- 3. In order to avoid the degradation of RNA standards, please use RNA-free consumables for the experiment, and store the standards at 2 ~ 8℃ after the experiment.
- 4. Please use the calibrated pipette to ensure the accuracy of quantitative results.
- 5. Please conduct quantitative test at room temperature. Before use, place put each component in the kit at room temperature. During the experiment, do not hold the detected PCR tube with your hand for a long time.
- 6. Please complete the detection within 3 h of working solution preparation to avoid fluorescence quenching that could lead to biased results.

#### **Machanism & Workflow**

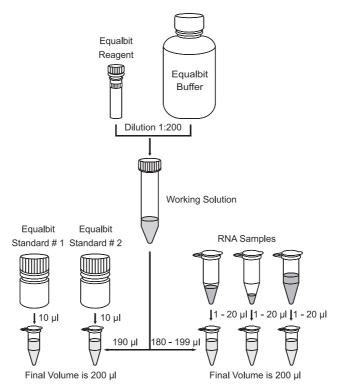


Fig 1. Workflow of Equalbit RNA HS Assay Kit

# **Experiment Process**

This protocol is only suitable for Qubit 2.0, Qubit 3.0 and Qubit 4.0 fluorimeters.

- 1. Equilibrate all the components to room temperature before use.
- 2. Prepare sufficient 0.5 ml PCR tubes to accommodate all samples and standards.
  - ▲ Only 0.5 ml PCR tubes are suitable for detection. It is recommended to use Qubit assay tubes (Thermo #Q32856) or Axygen PCR-05-C tubes (Axygen #10011-830).
- 3. Label the lid of each PCR tube. Do not label the side walls of the PCR tubes as this may interfere with the acquisition of fluorescent signals.
- 4. Prepare fresh working solution of Equalbit RNA HS Reagent, by diluting it in Equalbit RNA HS Buffer according to a ratio of 1:200. **DO NOT** use glass containers for the preparation of working solution.
  - A sufficient amount of working solution should be prepared to accommodate all samples and standards. For example, to accommodate 7 RNA samples and 2 standards, it is needed to prepare 2 ml of working solution by adding 10 μl of Equalbit RNA HS Reagent to 1,990 μl of Equalbit RNA HS Buffer. Mix thoroughly by vortexing.
- 5. Prepare standards. Load 190 μl of working solution to each tube used for standards, then carefully add exactly 10 μl of Standard # 1 and Standard # 2 to the corresponding tube, respectively. Gently vortex for 2 3 sec to avoid bubbles. Make sure the exact pipetting of 10 μl.
- 6. Prepare samples. For each tube used for samples, add 180 199 μl of working solution, and then carefully add 1 20 ul of RNA sample. Make sure the final volume in each tube is 200 μl. Gently vortex for 2 3 sec to avoid bubbles.
  - ▲ The volume range of RNA sample to be tested is 1 20 μl; The volume range of the test working solution is 180 199 μl, and the total volume is 200 μl.
- 7. Incubate at room temperature for 2 min. Protect from light.
- 8. Load the sample tubes into a Qubit Fluorometer and test the sample concentration by performing the RNA High Sensitivity Detection Program.