

SARS-CoV
Cat. No. C02-01-1112R

One-step real-time reverse transcription PCR for
detection of SARS Coronavirus

Includes main components for 50 reactions for Roche LightCycler®
Real-Time PCR System



Rev. 1
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HAI KANG LIFE CORPORATION LIMITED

PLEASE READ THROUGH THE ENTIRE PROTOCOL BEFORE
STARTING.

1. KIT COMPONENTS

Amplification Reagents

- 2 x 400 µl SARS-CoV Mastermix (store at -20°C)
- 2 x 25 µl Positive Control (store at -20°C)
- 1 x 20 µl Taq Polymerase (store at -20°C)

Storage Conditions

Store Mastermix, Positive Control and Taq polymerase at -20°C.
Thaw frozen reagents just before use. Mix reagents thoroughly (do
not vortex Mastermix containing enzyme).

Note:

Please aliquot the Mastermix into appropriate volume according to
your test frequency, in order to minimize repeated freeze and thaw
cycles. Frequent thawing and freezing may inactivate some kit
components.

2. PROCEDURES

Set-up

- In addition to the RNA obtained from the test samples, each
experiment requires a positive and negative (water) control.
- Set up real-time PCR components according to the table below:

| Components | Volume per reaction |
|----------------|---------------------|
| Mastermix | 15.6 µl |
| Taq Polymerase | 0.4 µl |
| Sample RNA | 4 µl |
| Total Volume | 20 µl |

Note:

- Use real-time PCR tubes and consumable recommended for
your real-time PCR equipment.
- Keep RNA samples on ice throughout experiment.
- 4 µl of provided Positive Control can be used to monitor the
success of amplification.
- It is advisable to run samples in duplicate to ensure reliability of
results.
- Spiking Control: 1 µl of Positive Control can be spiked into test
sample to check whether the test sample contains PCR
inhibitory substances.

Cycling conditions:

| | | | Data Collection Points (FAM Filter / 530nm) |
|-----------|------|------------|--|
| 1 Cycle | 42°C | 30 Minutes | - |
| | 95°C | 10 Minutes | - |
| 5 Cycles | 95°C | 10 Seconds | - |
| | 45°C | 30 Seconds | - |
| 40 Cycles | 72°C | 1 Minute | - |
| | 95°C | 15 Seconds | - |
| | 58°C | 1 Minute | ☐ |

(Take readings at point ☐)

3. DATA ANALYSIS AND INTERPRETATION

Detailed explanations of the basic and advanced operating
procedures should be provided with your Roche LightCycler®
Real-Time PCR System. This kit is optimised using the Roche
LightCycler® 2.0 Real-Time PCR System.

Spiking Control

Negative real-time PCR result may be due to a few scenarios: 1.
absence of detected sequence in the sample; 2. presence of
detected sequence below limit of detection; 3. presence of PCR
inhibitory substances. The purpose of spiking control is to verify
whether the test sample contains substances, which may affect PCR
reactions. If Ct equals 40 (or similar to Ct given by negative control)
when the test sample is spiked with the positive control, the test
sample is highly likely to contain PCR inhibitory substances, and the
result should NOT be taken as negative. Repeated extraction and
real-time PCR of the sample will be required.

If you require more detailed analysis information, please contact Hai
Kang Life Corporation Limited for technical assistance.

4. TECHNICAL ASSISTANCE

Our technical staff will provide technical assistance you may need in
using this kit. Simply call +(852) 2111 2123 during office hours:

Monday – Friday: 9:00am to 5:30pm
Saturday: 9:00am to 1:00pm

A recorded message (in English, Cantonese or Putonghua) may be
left outside office hours.

Alternatively, you may contact our technical staff by fax or email.

Fax: +(852) 2111 9762
Email: technical@haikanglife.com

5. WARRANTIES AND LIABILITIES

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