

VetScreen PCV2

User Manual
Cat. No. V03-01-1127

One-step reverse transcription PCR for gel-based
detection of Porcine Circovirus Type 2
in veterinary and environmental samples

Includes main components for 50 reactions



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HAI KANG LIFE CORPORATION LIMITED

PLEASE READ THROUGH THE ENTIRE PROTOCOL BEFORE
STARTING.

1. KIT COMPONENTS

DNA Amplification Reagents

- 25 Tubes Reagent Spheres (store at 4°C)
- 2 x 500 µl PCV2 Sphere Diluent (store at -20°C)
- 2 x 25 µl Positive Control (store at -20°C)

Storage Conditions

Store Reagent Spheres at 4°C with silica gel desiccant. Store Sphere Diluent, Positive Control, and reconstituted sphere at -20°C. Thaw frozen reagents just before use. Mix reagents thoroughly (do not vortex reconstituted sphere containing enzyme).

2. PROCEDURES

Mastermix Preparation

- Determine required number of reactions (n).
- Number of Reagent Sphere(s) required = 0.5 x n.
- Add 40 µl Sphere Diluent for each Reagent Sphere.
- Allow Reagent Sphere to reconstitute on ice.
- Gently tap tubes to mix reagents. Do not vortex enzyme containing reagents.

Set-up

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

| Components | Volume per reaction |
|--------------|---------------------|
| Mastermix | 20 µl |
| Sample RNA | 5 µl |
| Total Volume | 25 µl |

Note:

- 5 µl of the provided Positive Control can be used to monitor the success of amplification.
- Keep RNA samples on ice throughout experiment.
- It is advisable to run samples in duplicate to ensure reliability of results.
- Spiking Control: 1 µl Positive Control can be spiked into test sample to check whether the test sample contains PCR inhibitory substances.

Cycling conditions:

| | | |
|-----------|------|------------|
| 1 Cycle | 42°C | 30 Minutes |
| | 95°C | 10 Minutes |
| 40 Cycles | 95°C | 30 Seconds |
| | 59°C | 30 Seconds |
| | 72°C | 30 Seconds |

3. DATA ANALYSIS AND INTERPRETATION

Expected PCR product size: 149 base pairs

Spiking Control

Negative 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 no band is visible on agarose gel 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 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

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