

Porcine epidemic diarrhea virus nucleic acid detection Kit

(MIRA-exo) Guide Manual

(Part number: WLP9233KIT)

Product description

This kit designs specific primers and probes for the highly conserved regions of Porcine epidemic diarrhea virus (PEDV). It enables detection of the Porcine epidemic diarrhea virus in swine blood, serum, or tissues in 20 minutes by MIRA technology.

Overview of the MIRA technology

Multienzyme isothermal rapid amplification(MIRA) is based on a combination of polymerases and DNA recombination/repair proteins: Recombinases bind to single-stranded nucleic acid and stimulate the resulting protein-DNA complex to search for homologous sequences in duplex DNA with Single Strand Binding protein. Once homology is located, a strand-switching reaction is performed and the oligonucleotide is paired to its complement permitting a polymerase to begin synthesis from the 3' end. The amplification process is very rapid when optimised and can reach detectable levels of product in a few minutes.

Materials provided

Materials provided	Content
E Buffer	2 tube × 1 mL
B Buffer	1 tube × 150 μL
Positive control DNA template	1 tube × 100 μL
Steel bead reagent	48
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Storage conditions

- Transport temperature: ≤ 20 °C.
- Store at -20 °C without light, no repeated freezing and thawing.
- Full activity is guaranteed for 14 months.

Set up

1. Thaw reagents at room temperature.
2. Shake the lyophilized powder tube to confirm that each tube has a small steel bead. Thoroughly mix the contents of each tube by vortexing, then briefly centrifuge.
3. Add 37.5 μL E buffer (supplied) to a sample response MIRA exo reaction.
4. Add 10 μL sample RNA.

5. Add 2.5 μ l B buffer (supplied) and mix well and spin briefly to start reaction (For multiple reactions, it is recommended to add B buffer to the inner side of the reaction tube cover, and turn the reaction tube upside down to mix well) .
6. Turn on the Amp Future WL-16-II Constant Temperature Fluorescence Detector, put the reaction tube into the device and record the sample placement sequence, select RNA mode, 16-well synchronization, and FAM channel. The amplification temperature of this program was 39 - 42 $^{\circ}$ C, 30 s/cycle, 20 minutes, and a total of 40 cycles.

Reaction mix

E Buffer	37.5 μ L
RNA sample	10 μ L
B Buffer	2.5 μ L
Total volume	50 μ L

Interpretation of results

1. Positive: the amplification curve is S-like, and the instrument is judged as "+".
2. Negative: the instrument is judged as "-".

Note: When affected by the color of the sample or other factors, the machine shows the result as "+" but there is no "S"-shaped amplification curve, the result is judged as negative.

Note

1. Reactions start as soon as B buffer is added.
2. Set up a negative control for each experiment.
3. For research and development use only.

Ordering information and technical support

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