

ImaBeads® Viral Nucleic Acid Extraction Kit

For purification of Viral Nucleic Acid

Precautions

I. Handling Requirements

When working with chemicals, always wear a suitable lab coat, disposable gloves, and protective goggles.

II. Equipment and Reagents to Be Supplied by User

- Ethanol (96–100 %)*
- 1.5 ml microcentrifuge tubes
- Pipet tips with aerosol barrier
- Vortexer
- Microcentrifuge (with rotor for 1.5 ml tubes) may be required for some samples

* Do not use denatured alcohol, which contains other substances such as methanol or methylethylketone.

III. Waste Handling

- Treat waste with the country, federal, state and local regulations.

IV. Important points before use

- Do not use the product if it has expired.
- Add absolute ethanol (see the bottle label for volume) to IW2 Buffer then mix by shaking for a few seconds and tick the checkbox of the label on the bottle. Be sure and close the bottle tightly after each use to avoid ethanol evaporation.

Kit Contents

ICVL Buffer

ICVB Buffer

IBW1 Buffer

IW2 Buffer (Add Ethanol)

RNase-free Water

Proteinase K (Add PK Storage Buffer)

PK Storage Buffer

ImaBeads - 01

Storage and Stability:

1. This kit should be stored at room temperature.
2. Proteinase K should be stored at 4°C upon arrival.

Description

ImaBeads® Viral Nucleic Acid Extraction Kit is designed by patented technology for purification of Viral Nucleic Acid. Nucleic acid will be bound to magnetic beads. After washing off the contaminants, the purified Nucleic acid is eluted by treated nuclease-free elution buffer. Purified nucleic acid is suitable for downstream Real-time PCR, PCR or other molecular biology applications.

ImaBeads® Viral Nucleic Acid Extraction Kit Test Data

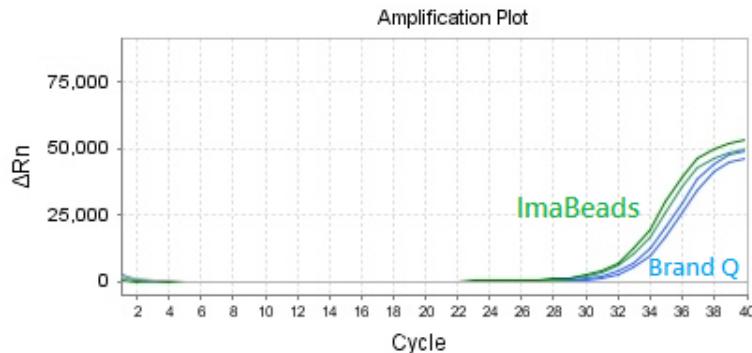


Fig 1. Viral RNA extracted from FCoV (feline coronavirus)

Viral RNA from 200 μ l of cat ascites samples was extracted using the ImaBeads® Viral Nucleic Acid Extraction Kit and & Competitive brand Q. A Real-time PCR assay was then performed with 4 μ l of synthesized cDNA as template, primers, and probe using the TaqMan Real-Time PCR system. The results confirmed that virus RNA can be successfully extracted and the performance of ImaBeads are about 1 Ct lower than competitor brand Q.

Preparation before using

Add 1.1 ml of PK Storage Bufferd to the Proteinase K tube.

Store prepared Proteinase K (10mg/ml) at 4 °C.

Protocol

1. Transfer 200 μ l of sample (serum, plasma, body fluids, and cell culture supernatant) into a microcentrifuge tube (not provided). If sample volume is less than 200 μ l, adjust sample volume to 200 μ l of with PBS (not provided).
2. Add 180 μ l of ICVL Buffer and 20 μ l of Proteinase K (10 mg/ml) to the sample, mix by vortexing.
3. Incubate at 56 °C for 10 minutes.
4. Vortex the magnetic **ImaBeads – 01** to mix completely and take 500 μ l of ImaBeads to a 1.5 ml RNase-free microcentrifuge tube.
5. Place the tube on a magnetic separator for 1 minute or until ImaBeads have pelleted then discard the cleared supernatant and remove the tube.
6. Add 380 μ l of ICVB Buffer to the sample lysate and vortex thoroughly.
7. Apply sample mixture to the 1.5 ml RNase-free microcentrifuge tube (prepared for use in step 5.) and mix by vortexing for 10 minutes.
8. Place the tube on a magnetic separator for 1 minute or until ImaBeads have pelleted then discard the cleared supernatant and remove the tube.
9. Add 800 μ l of IBW1 Buffer and mix by vortexing for 1 minute.
10. Place the tube on a magnetic separator for 1 minute or until ImaBeads have pelleted then discard the cleared supernatant and remove the tube.
11. Repeat step 9-10.
12. Add 800 μ l of IW2 Buffer and mix by vortexing for 1 minute.
13. Place the tube in a magnetic separator for 1 minute or until ImaBeads have pelleted then remove and discard the cleared supernatant.
14. Add 800 μ l of IW2 Buffer and mix by vortexing for 1 minute.
15. Place the tube in a magnetic separator for 1 minute or until ImaBeads have pelleted then remove and discard the cleared supernatant.
16. Incubate the tube at room temperature for 5 minutes to dry the ImaBeads.
17. Add RNase-free Water (80 μ L) and mix by vortexing for 10 seconds.
18. Incubate the tube at room temperature for 10 minutes and mix by vortexing for 10 seconds per 3 minutes.
19. Place the tube on a magnetic separator for 1 minute or until ImaBeads have pelleted then transfer the cleared supernatant to a new 1.5 ml RNase-free microcentrifuge tube.

Troubleshooting

Problem	Possible Reasons/Solution
Low Yield	<ul style="list-style-type: none">➤ Ensure absolute ethanol was added to IW2 Buffer and close the bottle tightly after each use to avoid ethanol evaporation.➤ Following ICVB Buffer addition, break up any precipitate as much as possible prior to loading to ImaBeads.