

# MagicPure<sup>TM</sup> Viral DNA/RNA Kit

Cat. No. EC301

Storage: Carrier RNA at -20°C for one year; Proteinase K solution at -20°C for two years; Magnetic Virus Beads at 2-8°C for one year (protect from freezing); others at room temperature (15°C-25°C) for one year

## Description

*MagicPure*<sup>TM</sup> Viral DNA/RNA Kit provides a simple and fast method to isolate viral DNA/RNA from up to 200 μl of plasma, serum, body fluid and mammalian cell culture supernatant. Samples are lysed by proteinase K and unique lysis buffer. Viral DNA/RNA is enriched by carrier RNA and bound to highly efficient magnetic beads. After washing, high quality DNA/RNA is eluted from the beads. Resulting DNA/RNA is free of protein contamination, and is suitable for PCR, RT-PCR, qPCR and qRT-PCR. The Kit is suitable for automatic magnetuc bead separation instruments.

- · Simple and fast, no centrifugation required
- · High quality, free of contaminants and inhibitors

## Kit Contents

| Component               | EC301-01/11 (50 rxns) |
|-------------------------|-----------------------|
| Binding Buffer 19(BB19) | 12 ml                 |
| Clean Buffer 19(CB19)   | 18 ml                 |
| Wash Buffer 19(WB19)    | 12 ml                 |
| RNase-free Water        | 10 ml                 |
| Carrier RNA(1 µg/µl)    | 150 µl                |
| Proteinase K (20 mg/ml) | 1 ml                  |
| Magnetic Virus Beads    | 1 ml                  |
| Magnetic Stand(16 hole) | 1/-                   |

# Sample requirements

- Stored at 2-8°C for 4 hours; at -20°C or -80°C for long term storage.
- Avoid repeated freezing and thawing for plasm and serum (no more than once)

## Procedures

Before starting, add 4 ml isopropanol to BB19, 12 ml 100% ethanol to CB19, and 48 ml 100% ethanol to WB 19, mix thoroughly.

- 1. Add 20 µl of Proteinase K to a sterile 1.5 ml microcentrifuge tube.
- 2. Add 200 µl of sample to tube.
  - (if the sample volume is less than 200  $\mu$ l, use 1×PBS or 0.9% NaCl to make the final volume of 200  $\mu$ l).
- 3. Add 300  $\mu$ l of fresh prepared BB19 with carrier RNA (prepare BB19 with carrier RNA according to the table given in next pageand use it within 2 hours).
- 4. Add 20 μl of mixed Magnetic Beads to the lysate, mix thoroughly by vortexing for 30 seconds. Incubate at room temperature for 10 minutes. During the incubation, invert the tube 3-5 times.
- 5. Place the microcentrifuge tube onto the magnetic stand until the beads are pelleted against the magnet. Remove the supernatant.
- 6. Remove the microcentrifuge tube from the magnetic stand, add 500 µl of CB19 (make sure ethanol has been added) to the microcentrifuge tube, and vortex the microcentrifuge tubes for 15 seconds. Place the microcentrifuge tube onto the magnetic stand until the beads are pelleted against the magnet. Remove the supernatant.
- 7. Remove the microcentrifuge tube from the magnetic stand, add 500  $\mu$ l of WB19 (make sure ethanol has been added) to the microcentrifuge tube, and vortex the microcentrifuge tubes for 15 seconds.
  - Place the microcentrifuge tube onto the magnetic stand until the beads are pelleted against the magnet. Remove the supernatant.





- 8. Repeat Step 7 once.
- 9. Airdry the beads (not more than 10 minutes).
- 10. Remove the microcentrifuge tubes from the magnetic stand, add 100-200 µl of RNase-free Water to the microcentrifuge tube. Mix gently by vortexing or pipetting up and down for 1 minute to resuspend the beads and incubate at 65oC for 5 minutes. Mix gently by vortexing once or twice during incubation.
- 11. Place the microcentrifuge tube onto the magnetic stand , gently turn the tube left and right to attach the beads to the magnet. Carefully transfer the supernatant into a clean 1.5 ml microcentrifuge tube. The purified DNA can be stored at  $-20^{\circ}$ C and the purified RNA can be stored at  $-80^{\circ}$ C.

### Addition of carrier RNA to BB19

Calculate the volume of BB19 with carrier RNA needed by selecting the number of samples to be processed from Table 1.

Gently mix by inverting the tube 10 times. To avoid foaming, do not vortex.

For larger numbers of samples, volumes can be calculated using the following sample calculation:

 $N \times 0.31 \text{ ml} = A \text{ ml}$ 

 $A ml \times 2.8 \mu l/ml = B \mu l$ 

where: N = number of samples to be processed simultaneously

A = volume of BB19 needed

B = volume of Carrier RNA to be added into BB19

Table 1. Volumes of BB19 and carrier RNA required for different number of samples.

| No. of  | Vol. of | Vol. of     | No. of  | Vol. of | Vol. of     |
|---------|---------|-------------|---------|---------|-------------|
| samples | BB 19   | Carrier RNA | samples | BB 19   | Carrier RNA |
| 1       | 0.31 ml | 2.8 μl      | 6       | 1.86 ml | 2.8 µl      |
| 2       | 0.62 ml | 5.6 µl      | 7       | 2.17 ml | 5.6 μl      |
| 3       | 0.93 ml | 8.4 µl      | 8       | 2.48 ml | 8.4 µl      |
| 4       | 1.24 ml | 11.2 μl     | 9       | 2.79 ml | 11.2 μl     |
| 5       | 1.55 ml | 14 μl       | 10      | 3.10 ml | 14 μl       |

#### Notes

- Avoid repeated thawing and freezing samples.
- Aliquot the Carrier RNA into RNase-free microcentrifuge tubes and store at -20°C. Avoid repeated freezing and thawing (not more than three times) for Carrier RNA.
- Beads must be mixed well before using.
- Use sterile tubes and pipette tips to avoid DNase and RNase contamination.
- Thoroughly dry beads before elution to avoid residual ethanol interfering downstream applications.