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VIOGENE

User Bulletin

Viral RNA Extraction Miniprep System

For serum, plasma, body fluids, and cell culture supernatant.

Downstream Application

* Reverse transcription
* Northern blotting
* PCR
* RNase protection assay

Product Contents

Cat. No	GVR1001	GVR1002
Preps	50	250
RXV Buffer	35ml	190ml
WS Buffer	15ml	45ml x 2
RNA Carrier	1	1
RNase-free ddH_2O	1.5ml x 2	15ml
RNA Mini Column	50	250
Collection Tube	50	250
Elution Tube	50	250
Protocol	1	1

All buffers need to be mixed well before use.

Shipping & Storage

Viogene Viral RNA Extraction System is shipping and storage at ambient temperature up to 6 months.

If precipitate form by freezing temperature on any buffer, warm up at 37℃ to redissolve.

Protocol

❖ **Please read the following notes before starting the procedures.**

- **WARNING**, strong acids and oxidants (like for instance bleach) should not be used together with RXV buffer (because this kind of reaction would produce cyanide)!!!

Important Notes

<Note>: Preheat RNase-free ddH_2O to 80°C.

-for GVR1001- Add 60 ml of ethanol (98-100%) to the WS Buffer bottle when first open the bottle.

-for GVR1002- Add 180 ml of ethanol (98-100%) to the WS Buffer bottle when first open the bottle.

1. **Add RNA carrier to RXV Buffer.**
   Add 1 ml RXV Buffer to the RNA carrier tube, vortex to dissolve and transfer to the RXV Buffer bottle, store at 4°C.

2. **Pipet 150 μl sample (serum, plasma, body fluids, and cell culture supernatant) into a 1.5 ml tube.**

3. **Add 570 μl of carrier added RXV Buffer to the sample, mix by vortexing.**
   Through mixing is required for sample lysis. If the sample volume is larger than 150 μl, increase the amount of RXV Buffer proportionally.

4. **Incubate the vortexed sample at room temperature for 10 minutes.**

5. **Add 570 μl of ethanol (98-100%) to the sample, and mix by vortexing.**
   If the starting sample is larger than 150 μl, increase the amount of ethanol proportionally.

6. **Place a Total RNA Column in a 2 ml Collection Tube, apply 650 μl of the ethanol added sample from step 5 to the Total RNA Column, close the cap, centrifuge at 6,000 x g (8,000 rpm) for 1 minute, and discard the filtrate.**
   If the solution remains above the membrane, centrifuge again at 13,000 rpm.

7. **Repeat step 6 for rest of the sample.**

8. **Wash the column twice with 500 μl of ethanol added WS Buffer by centrifuging at full speed (13,000 rpm or 10,000 x g) for 1 minute, and discard the filtrate.**
   Add 60 ml (for GVR1001) or 180ml (for GVR1002) of ethanol (98-100%) to the WS Buffer bottle when first open the bottle.

9. **Centrifuge at full speed for 3 minutes to remove traces of WS Buffer.**
   Residual ethanol may inhibit reverse transcriptase activity.

10. **Transfer the column to a RNase-free 1.5 ml Elution Tube, add 50 μl of preheated (80°C) RNase-free ddH_2O, and centrifuge at full speed for 1 minute to elute RNA.**

11. **Store RNA at -70°C.**