



PRODUCT INFORMATION

One-Tube Viral DNA Isolation Buffer

Product information for VT4785/VT4786:

Kit Components:

Components	VT4785, 50 Preps	VT4786, 100 Preps
Lysis Buffer-V	30 ml	60 ml
Protocol	1	1

Storage and stability

Transportation at ambient temperature. Store at 4°C. Valid for 1 year.

Introduction

The Lysis Buffer-V Reagent is used for rapid isolation of viral DNA from broad range of cell-free clinical samples including serum, urine and plasma for clinical research and life science applications. The one-tube procedure is simple and fast, no transfer of sample between tubes is needed, which minimizes cross-contamination between samples. No phenol extraction is required. Purified viral DNA can be used for PCR, Real-Time PCR and other applications.



Features

- ü High yield. The recovery is generally >85%.
- ü Simple procedure. No transfer of sample between tubes is need, which minimizes the cross-contamination between samples.
- ü Easy to scale up.
- ü Compatible with many downstream applications including PCR and Real Time PCR.

Procedures

1. Sample Preparation. Add 0.2 ml liquid virus to a 1.5 ml micro-centrifuge tube.

Note 1: 1.5 ml conical microtube with screw cap and rubber-O ring are recommended.

Note 2: Do positive and negative controls.

Note 3: This protocol is suitable for viral DNA extraction from various cell-free liquid specimens including fresh, frozen and anti-coagulated blood samples. If anti-coagulated blood samples are used, anti-coagulating reagents must be EDTA or ACD. This procedure is not suitable for the blood sample with heparin as anti-coagulating reagent.

Note 4: Preparation of plasma. Add 1.5 ml of whole blood to a 1.5 ml eppendorf tube, spin at 1,000 g for 20 minutes. Transfer the supernatant plasma to a clean 1.5 ml eppendorf tube. Do not use any whole blood sample that was kept at 20-25°C longer than six hours. Plasma is stable at room temperature for 24 hours, at 2-8°C for 5 days and at -20°C for six months. Aliquot 0.2 ml in each 1.5 ml microtube.

Note 5: Enrichment of virus. If necessary, add up to 1.5 ml of liquid viral sample to a 1.5 ml vial and centrifuge at 4°C and 24,000 g for 60 minutes, discard most of the supernatant, keep approx 0.2 ml solution in the tube.



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2. Add 0.6 ml of Lysis Buffer-V Reagent, vortex vigorously for 30 seconds; keep at room temperature for 10 minutes.
3. Add 0.7 ml of isopropanol, mix well by inverting 5 times, and centrifuge at 13,000-15,000 g for 15 minutes at room temperature.
4. Carefully discard the supernatant.
5. Add 1.0 ml of pre-cooled 75% ethanol, mix well, centrifuge at 13,000-15,000 g for 5 minutes.
6. Discard the supernatant, avoid touching the pellet.
7. Air-dry for 2 minutes, add 200 µl dd-water or TE buffer to dissolve the pellet. The turbid solution can directly used for PCR, or kept at -20°C for long term storage.

Note 1: It is normal if the solution become turbid; don't spin the turbid solution.

Note 2: For PCR, a ratio of 1:5 or 1:10 of volume of DNA solution to volume of PCR system is recommended.



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