

Product Information

PEG Virus Precipitation Kit

Catalog Number **MAK343**
Storage Temperature $-20\text{ }^{\circ}\text{C}$

TECHNICAL BULLETIN

Product Description

The PEG Virus Precipitation Kit provides an easy, convenient, and time-saving method to concentrate viruses and remove impurities without ultracentrifugation. The kit can be used for small lab samples, in which the virus is typically produced at a low titer. It often needs to be concentrated for storage and/or for use in further applications. The kit can also be used with large-scale virus preparations with high yield and high viral titer.

The kit will concentrate retroviruses, baculoviruses, lentiviruses, and phages etc. in cell culture medium or environmental samples. Virus can be concentrated over 100-fold. An optimized Virus Resuspension Solution is provided to maximize viral recovery by 40–100 % depending on the virus type and source. The entire process uses non-toxic reagents. The concentrated virus can be used for infection, viral DNA, or RNA purification, etc.

Components

The kit is sufficient for 200 preparations.

PEG Solution (5×) 4 × 125 mL
Catalog Number MAK343A

Virus Resuspension Solution (1×) 4 × 10 mL
Catalog Number MAK343B

Reagents and Equipment Required but Not Provided.

- Refrigerated microcentrifuge capable of RCF $\geq 16,000 \times g$
- Microcentrifuge tubes
- Pipetting devices and accessories

Precautions and Disclaimer

For R&D use only. Not for drug, household, or other uses. Please consult the Safety Data Sheet for information regarding hazards and safe handling practices.

Storage/Stability

The kit is shipped on wet ice. Store components at $-20\text{ }^{\circ}\text{C}$ upon receiving.

Procedure

Note: The following procedure is designed for 10 mL of virus solution. One can proportionally adjust the volumes according to the sample volume.

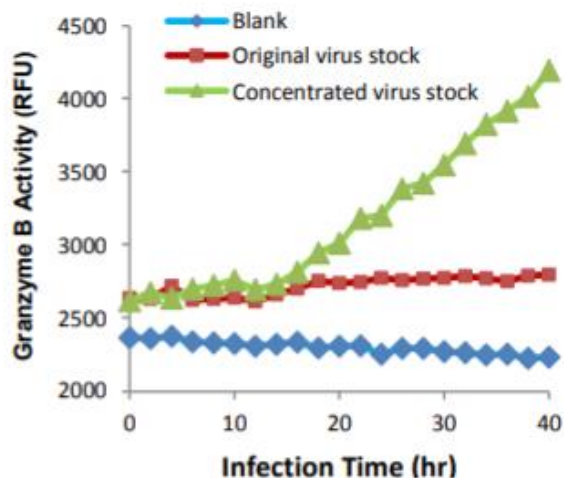
1. Infect cells or transfect, and allow maximum virus accumulation.
2. For mammalian cell-virus or insect-baculovirus, centrifuge culture at $3,200 \times g$ for 15 minutes at $4\text{ }^{\circ}\text{C}$ to remove cell debris. For bacterial phage, centrifuge at $16,000 \times g$ for 15 minutes at $4\text{ }^{\circ}\text{C}$ to remove cell debris.
3. Collect supernatant.
4. Add 2.5 mL of PEG solution (5×) to 10 mL of virus supernatant.
5. Refrigerate overnight (stable up to 2 days at $4\text{ }^{\circ}\text{C}$).
6. Centrifuge at $3,200 \times g$ for 30 minutes at $4\text{ }^{\circ}\text{C}$.
7. Carefully remove and discard supernatant by aspiration. The beige or white pellet is the virus.
8. Resuspend the virus pellet in 100 μL of Virus Resuspension Solution.
9. Aliquot the virus and store at $-70\text{ }^{\circ}\text{C}$ for future use.

Notes:

1. For a high titer virus preparation, the resuspension volume should be limited to about three times the volume of the white pellet, usually 1/10 to 1/100 volume of original sample. If insoluble material is present in the viral suspension, it can be removed by centrifuge at $3,200 \times g$ for 15 minutes at $4\text{ }^{\circ}\text{C}$.
2. Avoid freeze/thaw cycles to maximize virus recovery.
3. Trace amounts of PEG in the virus suspension will not affect the use of the concentrated virus. In some cases, PEG may increase virus infection efficiency. However, if it is desired, the trace amount of PEG can be removed by the following procedure:
 - a. Add 1 volume of solution containing 4 M KCl and 50 mM Tris-HCl, pH 7.2 (not provided), to 3 volumes of the concentrated virus suspension.
 - b. Alternatively, add solid KCl into the virus suspension to a final concentration of 1 M.
 - c. Let stand on ice for 15–30 minutes.
 - d. Spin at $12,000 \times g$ for 10 minutes at $4\text{ }^{\circ}\text{C}$ to remove the precipitate.
 - e. Carefully collect the virus supernatant.
 - f. Aliquot and store at $-70\text{ }^{\circ}\text{C}$ for future use.

Results**Figure 1.**

Concentration of Baculovirus



Low titer baculovirus (10 mL) expressing human Granzyme B was precipitated following the kit protocol and the precipitate was suspended in 1 mL of Virus Resuspension Solution. Both low titer and precipitated baculovirus were subsequently used to infect *Sf9* Insect cells at different infection times. The activity of recombinant Granzyme B secreted into the culture medium by low titer and precipitated baculovirus infected insect cells was monitored using Granzyme B Activity Assay Kit (MAK176).

References

1. Lech, K., *Current Protocols in Molecular Biology*, (1990) 1.13.1-10.
2. Kimlton, C.P. et al., *Journal of Virological Methods*. **28**, 141-146 (1990).
3. Colombet, J. et al., *Journal of Virological Methods*. **71**, 212-219 (2007).

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