ab102538

PEG Virus Precipitation Kit

Instructions for Use

For the rapid, sensitive and accurate purification of viruses in various samples

This product is for research use only and is not intended for diagnostic use.
Table of Contents

1. Overview 2
2. Protocol Summary 3
3. Components and Storage 4
4. Assay Protocol 5
5. Typical Data 7
1. Overview

Viruses are usually produced at a low titer. They often need to be concentrated for storage or further applications. A quick, easy and inexpensive method is desired to concentrate viruses and remove impurities.

Abcam's PEG Virus Precipitation Kit provides an easy, convenient and time-saving method to concentrate viruses without ultracentrifugation. The kit can be used for small lab samples or large scale virus preparation with high yield and high viral titer. The kit can be used to concentrate retroviruses, baculoviruses, lentiviruses, and phages etc. in cell culture medium or environmental samples. Viruses can be concentrated over 100 fold. An optimized Virus Re-suspension Solution is provided to maximize viral recovery by 40-100% depending on the virus type and sources. The whole process uses non-toxic reagents. The concentrated virus can be used for infection, viral DNA or RNA purification, etc.
2. Protocol Summary

- Infect/Transfect Cells for Virus Accumulation
- Centrifuge and Collect Supernatant
- Add PEG Solution
- Incubate Overnight
- Centrifuge and Collect Supernatant
- Re-Suspend in Virus Re-Suspension Solution
- Aliquot for Future Use
3. Components and Storage

A. Kit Components

<table>
<thead>
<tr>
<th>Item</th>
<th>Quantity</th>
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</thead>
<tbody>
<tr>
<td>PEG Solution (5X)</td>
<td>125 mL</td>
</tr>
<tr>
<td>Virus Re-suspension Solution (1X)</td>
<td>10 mL</td>
</tr>
</tbody>
</table>

* Store at +4°C or -20°C. The solutions are ready to use and stable for 12 months.

B. Additional Materials Required

- Microcentrifuge
- Pipettes and pipette tips
- 4 M KCl
- 50 mM Tris-HCl
4. Assay Protocol

Note: The following protocol is designed for 10 ml virus solution. You can proportionally adjust the volumes according to your sample volume.

Note: The PEG solution (5X) is ready to use and does not need dilution.

1. Infect or transfect cells and allow maximum virus accumulation.

2. For mammalian cell-virus or insect-baculovirus, centrifuge culture at 3,200 x g for 15 minutes at +4°C to remove cells debris. For bacterial phage, centrifuge at 16,000 x g for 15 minutes at +4°C to remove cells debris.

3. Collect supernatant and add 2.5 ml of PEG solution (5X) to 10 ml of virus supernatant. Refrigerate overnight (stable up to 2 days at 4°C).

4. Centrifuge at 3,200 x g for 30 minutes at 4°C, carefully remove supernatant by aspiration. The beige or white pellet is the virus.

5. Suspend the virus pellet in 100 μl Virus Re-suspension Solution. Aliquot the virus into small aliquots and store at -80°C for future use.
Notes:

a) For high titer virus preparation, the re-suspension 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 centrifugation at 3,200 x g for 15 min at +4°C.

b) Avoid freeze/thaw cycles to maximize virus recovery.

c) 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:

Add 1 volume of solution containing 4 M KCl and 50 mM Tris-HCl, pH7.2 (not provided) to 3 volumes of the concentrated virus suspension. Alternatively, add solid KCl into the virus suspension to a final concentration of 1 M. Sit on ice for 15-30 min. Spin at 12,000 x g for 10 min at 4°C to remove the precipitate. Carefully collect the virus supernatant. Aliquot and store at -80°C for future use.
5. Typical Data

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 was subsequently used to infect SF9 Insect cells. The activity of recombinant Granzyme B secreted into the culture medium by low titer and precipitated baculovirus infected insect cells was monitored using ab157403 - Granzyme B Activity Assay Kit (Fluorometric).
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