

PEG Virus Precipitation Kit

(Catalog #K904-50/200, 50/200 Preps, Store kit at +4°C or -20°C)

I. Introduction:

Virus was usually produced at a low titer. It often needs to be concentrated for storage or further applications. A quick, easy and inexpensive method is desired to concentrate virus and remove impurities. BioVision's PEG Virus Precipitation Kit provides an easy, convenient and time-saving method to concentrate virus without ultra-centrifugation. 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. 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 sources. The whole process uses non-toxic reagents. The concentrated virus can be used for infection, viral DNA or RNA purification, etc.

II. Kit Contents:

Components	K904-50	K904-200	Part No.
PEG Solution (5X)	125 ml	4 X 125 ml	K904-xx(x)-1
Virus Re-suspension Solution (1X)	10 ml	4 X 10 ml	K904- xx(x)-2

III. Reagent Storage Conditions:

The concentrated virus solutions are ready to use and stable for 12 months at +4°C or at -20°C for long term storage.

IV. Virus Precipitation Protocol:

The following protocol is designed for 10 ml virus solution. You can proportionally adjust the volumes according to your sample volume. The kit is also available for larger volume samples. Please inquire.

1. Infect cells or transfect and allow maximum virus accumulation.
2. For mammalian cell - virus or insect - baculovirus, centrifuge culture at 3,200 x g for 15 min at 4°C to remove cells debris. For bacterial phage, centrifuge at 16,000 x g for 15 min 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 min 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 Resuspension Solution. Aliquot the virus and store at -70°C for future use.

Notes:

- 1) For 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 x g for 15 min at 4°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:
 - i) 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.
 - ii) Alternatively, add solid KCl into the virus suspension to a final concentration of 1 M.
 - iii) Let stand on ice for 15 - 30 min.
 - iv) Spin at 12,000 x g for 10 min at 4°C to remove the precipitate.
 - v) Carefully collect the virus supernatant. Aliquot and store at -70°C for future use.

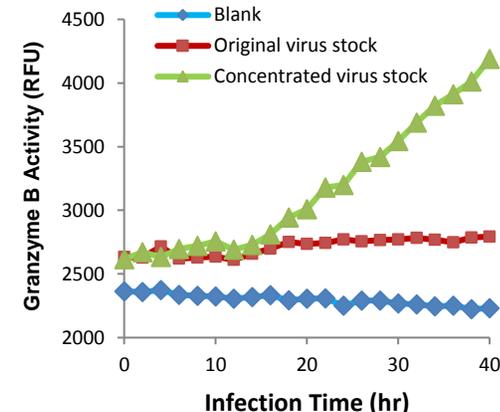


Figure: Concentration of baculovirus: Low titer baculovirus (10 ml) expressing human Granzyme B was precipitated following the kit protocol and the precipitate was suspended in 1ml of Virus Re-suspension Solution. Both low titer and precipitated baculovirus were subsequently used to infect SF9 Insect 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 Fluorometric Assay Kit (K168-100).

V. References:

1. Lech K. *Current Protocols in Molecular biology* (1990) 1.13.1-10
2. Kimlton C.P, Corbitt G, Morris D.J. *Journal of Virological Merthods.* (1990) 28: 141-146.
3. Colombet J. Robin A, Lavie L, et al. *Journal of Virological Merthods.* (2007) 71: 212-219.

FOR RESEARCH USE ONLY! Not to be used on humans.