

## Virus Concentration Kit

### Introduction

The Virus Concentration Kit is a kit that can quickly and efficiently concentrate common viruses. This kit is suitable for lentivirus, adeno-associated virus, adenovirus, baculovirus and some other viruses, and only needs to mix the virus concentration reagent in proportion to the collected viral supernatant, incubate for a certain period, and then resuspend with the virus resuspension solution. Compared with ultracentrifugation and column concentration, this kit is more convenient to use, and the virus recovery rate can reach up to 90%, and the concentrated virus concentration can be increased by 10-100 times. At the same time, this kit can effectively (but not sufficiently) remove large amounts of serum proteins, cell debris, and genomic DNA from the collected viral supernatant. In addition, the kit provides a virus preservation reagent for better protection of the concentrated virus.

The reagent provided in this kit can concentrate 150 mL of viral supernatant as recommended in this instruction.

### Components and Storage

Components	K2706-1 Kit
Virus Concentration Reagent (4X)	50 mL
Virus Resuspension Solution	15 mL
Virus Preservation Reagent (10X)	1.5 mL
Store the kit at -20°C, stable for 1 year.	

### Protocol

1. After virus packaging is complete, collect the virus-containing supernatant and centrifuge at 8000 g at 4°C for 30 min to fully pellet the cell debris. The clarified supernatant is taken for subsequent concentration. Virus-containing supernatants can also be filtered with the 0.45 µm, polyethersulfone (PES) filters. Do not use nitrocellulose filters as they may adsorb viruses.
2. Pre-thaw the Virus Concentration Reagent (4X) at 4°C, and mix it with the clarified viral supernatant in a 1:3 ratio. After mixing, incubate on a shaker or mixer at low speed at 4°C for at least 4 h or overnight. The viral solution after incubation usually becomes cloudy.

**\*Note:**

1. Virus Concentration Reagent (4X) may precipitate after thawing, heat it in a 37°C water bath for 30 minutes or more to make it completely dissolved, and then pre-cool it at 4°C for later use.
2. Virus Concentration Reagent (4X) is very viscous, so pipette slowly and make sure it is well mixed with the viral supernatant.
3. Longer incubation time appropriately can help improve virus recovery, but it is recommended that the incubation time should not exceed 24 h.
4. When mixing on a shaker or mixer, it is necessary to ensure that the rotation speed allows the solution to mix well.
5. The mixing process needs to be performed at 4°C, where the virus will be best concentrated. If the temperature is too high, the concentration will not be sufficient.
6. If the sample volume is too large, it is recommended to extend the incubation time appropriately.

3. After incubation, centrifuge at 4°C, 8000 g for 0.5-1 h. A white precipitate can be seen at the bottom of the tube (sometimes the pellet is not visible). Carefully discard the supernatant. Do not touch the pellet during the aspiration process and do not shake the centrifuge tube vigorously.
4. Centrifuge at 4°C, 8000 g for 1 min, then carefully discard the remaining supernatant without touching the pellet.
5. (Optional): Add Virus Preservation Reagent to the Virus Resuspension Solution. Mix Virus Preservation Reagent (10X) and Virus Resuspension Solution in a ratio of 1:9.

**\*Note:** The addition of the Virus Preservation Reagent (10X) is not required. It is recommended if the virus is subsequently used to culture cells or tissues, not if the virus is subsequently used in animals. If it is subsequently used for extraction of the virus's DNA or RNA, this step is optional.

6. Add 1-10% of the original viral supernatant volume of Virus Resuspension Solution (with or without Virus Preservation Reagent as needed), stand for 10 minutes, and then gently pipette 20-30 times to resuspend the virus pellet. Bubbles need to be avoided when blowing, and the blowing needs to be gentle, as vigorous blowing may cause the virus inactivation.

**\*Note:**

7. If you can't see the white precipitate, you can use the Virus Resuspension Solution to blow the area where the precipitate may form.
8. The amount of white precipitate does not represent the amount of virus. In addition to viral particles, there may be small amounts of serum proteins, cell debris, and genomic DNA in the white pellet.

7. Centrifuge at 4°C, 12000 g for 3-5 min, and the supernatant is concentrated virus. It can be stored at -80°C in appropriate aliquots as needed.

**\*Note:** The centrifugation precipitate contains serum proteins, cell debris, and genomic DNA.

## Note

1. Virus Concentration Reagent (4X) may precipitate after thawing, heat it in a 37°C water bath for 30 minutes or more to make it completely dissolved, and then pre-cool it at 4°C for later use.

2. In order to avoid virus inactivation, repeated freeze-thaw cycles should be avoided both before and after concentration.
3. Serum in cell culture media is useful for virus precipitation and concentration. If the medium used for virus packaging is low serum or serum-free, or if the virus needs to be concentrated from a solution with very low protein content, it is recommended to add a certain amount of sterile BSA (final concentration of 3%) during virus concentration to significantly improve the concentration effect.
4. For your safety and health, please wear lab coats and gloves during the experiment.
5. For research use only. Not to be used in clinical diagnostic or clinical trials.



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