Evomic Science

EXOSOME & VIRUS
We Focus On Tiny Particles

Protocol for Virus Particle Concentration (Cat#: VirC50)

1. Preparation of Crude Virus Sample

1. Collect cell culture supernatants, or cell lysis, body fluids, even environmental liquids.
2. Centrifuge the raw virus sample at 2,500×g for 15 minutes at 4˚C or room temperature to remove cells and large particles.
3. Carefully transfer supernatants into new tubes and then pass through 0.45 μm filter or centrifuge supernatants at 10,000×g for 10 minutes at 4˚C to remove most of large particles.

2. Virus Particle Concentration

1. Transfer the desired volume of the above raw virus samples to a new tube and add 0.5 volumes of Buffer V1, 0.5 volumes of Buffer V2, and 1/100th volume of Enhancer C and T, respectively. (Refer to the table below for sample volumes)

Supernatants	Buffer V1	Buffer V2	Enhancer C	Enhancer T
5 ml	2.5ml	2.5 ml	50 μl	50 μl
10 ml	5 ml	5 ml	100 μl	100 μl

2. Mix supernatants with the buffers well (Do not vortex), and then centrifuge the samples at 2,500g for 15 min at 4°C or room temperature.
3. After centrifugation, remove supernatants carefully with pipette. Left 200~500 μl of supernatants in bottom! Do not touch this soft pellet in the bottom!
4. Transfer the soft pellet with supernatants (200~500 μl) to a 2 ml dolphin microtube and spin down for 3~5 min at 2,500g.
5. Virus are concentrated on the interface and bottom phases! Remove the extra reagents / supernatants carefully with pipette! Do not touch the interface and bottom phases!
6. Suspend the concentrated virus in PBS or your desired buffer.
7. These concentrated virus are suitable for most of *in vitro* applications, such as RNA isolation (***Do not use classical TRIZOL reagent for RNA isolation***), ELISA and western blot, etc.
8. If trace contaminated proteins, endotoxins and reagents are concerned, or purer virus particles (such as for Protein Mass Spectrometer, and *in vivo* animal study, vaccine production) are desired, the concentrated virus should be further purified by the Virus Particles Purification kit (Cat# VirP300).
9. We recommend to use the fresh isolated virus particle immediately. Otherwise please store at 4˚C for overnight, or freeze at -20˚C or -80˚C for longer periods. Note that repeated thaw and freeze cycles can damage virus structure.