Nanotrap Extracellular Vesicle A Particles Protocol

This protocol describes a method using non-magnetic Nanotrap® Extracellular Vesicle A Particles to concentrate intact extracellular vesicles that were isolated using size exclusion columns, such as the qEV columns from Izon Science Limited.

Contents and Equipment Lists

Download the Contents and Equipment Lists PDF at https://www.ceresnano.com/ev/ev-user-guide.

Troubleshooting

- Ensure that all reagents are stored at the indicated temperatures.
- Make sure refrigerated reagents and samples (if applicable) are warmed to room temperature before starting the procedure.
- Ensure that the centrifuge tubes used can withstand the centrifugal forces required.
- If you are interested in exploring different applications for the Nanotrap® Extracellular Vesicle Particles, please contact support@ceresnano.com for technical assistance.
- Nanotrap Extracellular Vesicle A Particles must be adequately vortexed immediately before each use.
- Nanotrap Extracellular Vesicle A Particles are not suitable for use in conjunction with RNA extraction kits that use alcohols in the lysis buffer, as it decreases the lysis efficiency of extracellular vesicles that are bound to the Nanotrap Extracellular Vesicle A Particles. Please ensure any RNA extraction kit used downstream is compatible with this kit before use. RNA extraction kits can be used if the lysis buffer is substituted for one that does not contain alcohols, such as MagMAX™ Microbiome Lysis Solution, (part number A42361). This is a guanidine thiocyanate based lysis solution that does not contain alcohol.
- Once the Nanotrap Extracellular Vesicle A Particles have been pelleted, the lysate can be removed and used in any RNA extraction protocol.
- Note that extracellular vesicles bind irreversibly to Nanotrap Extracellular Vesicle A Particles. This is an important consideration for extracellular quantification analysis and functional studies downstream.

Guidelines

After the isolation of extracellular vesicles using a size exclusion column, follow the steps below to concentrate the extracellular vesicles. The volume of Nanotrap Extracellular Vesicle A Particles required to concentrate extracellular vesicle-containing samples is dependent on the volume of the purified extracellular vesicle sample. See **Table 1** for suggested volumes of Nanotrap Particles to be used for different volumes of purified extracellular vesicles. If you are unsure of how to prepare your sample or what volume of Nanotrap Particles to add, please contact support@ceresnano.com for assistance.

Table 1: Recommended volume of Nanotrap Extracellular Vesicle A Particles to be added to purified extracellular vesicle samples

Volume of Purified Extracellular Vesicles	Volume of Nanotrap Extracellular Vesicle A Particles
0.68 mL	50 µL
1.6 – 2.8 mL	100 µL
8 mL	150 µL
20 mL	200 µL

Concentrating Extracellular Vesicles

In a clean lo-bind tube of the appropriate size, complete the following steps, in order.

1. Add the sample containing the purified extracellular vesicles to the sample tube.
2. Vortex Nanotrap Extracellular Vesicle A Particles for 30 seconds to resuspend.
3. Add the appropriate volume of Nanotrap Extracellular Vesicle A Particles **(see Table 1)** to the sample tube.
4. Vortex sample tube for 30 seconds to ensure that the Nanotrap Extracellular Vesicle A Particles are fully resuspended and incubate for one hour at room on a tube roller or inverter.
5. Centrifuge the mixture at 16,800 x g for 10 minutes to pellet the extracellular vesicles bound to Nanotrap Extracellular Vesicle A Particles.
6. Remove the supernatant, being careful not to disturb the pellet containing extracellular vesicles bound to Nanotrap Extracellular Vesicle A Particles.
   Note: *If the pellet is disturbed, the sample may need to be re-centrifuged.*
7. The extracellular vesicle pellet is now ready for downstream applications. The pellet can be resuspended in a desired buffer volume or used directly with method-appropriate lysis buffer to avoid further dilution of concentrated extracellular vesicles.

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