Spin Column Exosome Isolation Kit

Cat. No.: P-CA-504

Size: 20Tests / 50Tests

Product Description

This kit employs a centrifugal column-based method for exosome isolation. The column matrix efficiently adsorbs impurities while excluding exosomes, enabling ultra-fast isolation with high recovery rates. The isolated exosomes are suitable for downstream applications such as Western blot (WB) analysis, nanoparticle tracking analysis (NTA), nano-flow cytometry, electron microscopy, omics research, and functional studies in cellular and animal models. Additionally, this kit effectively removes free exosome-labeling dyes, ensuring enhanced purification quality.

Product Composition

Component	20 Tests	50 Tests	Storage Conditions
Spin Column 01	20 EA	50 EA	2-8°C, Shading Light
Collection Tubes (2 mL)	20 EA	50 EA	2-30°C, Shading Light
Solution A	25 mL	100 mL	2-8°C, Shading Light

Storage Conditions

Store Spin Column 01 and Solution A at 2-8°C. The shelf life is 18 months.

Applicable Samples

This kit is suitable for exosome isolation from various sample types. For small-volume, precious samples such as cerebrospinal fluid, saliva, bile, or seminal plasma, please consult our technical support team for guidance.

Required Instruments, Reagents, and Consumables (Not Included)

- High-speed refrigerated centrifuge
- Benchtop Orbital Shaker
- 2 mL / 1.5 mL Centrifuge tubes
- Magnetic Rack
- Ultrafiltration tubes (MWCO: 50 kDa)
- Ultrafiltration tubes (MWCO: 10 kDa)

Protocol

(一) Exosome Isolation Protocol

1. Sample Processing

1) **Cell Removal:** Centrifuge the sample at 300 × g for 5 minutes at 4°C. Carefully transfer the supernatant to a new centrifuge tube.
   Note: This step can be skipped for cell-free samples.
2) **Removal of Cellular Debris:** Centrifuge the supernatant obtained from Step 1 at 2,000 × g for 10 minutes at 4°C. Transfer the supernatant to a new centrifuge tube.
3) **Removal of Large Particles:** Centrifuge the supernatant obtained from Step 2 at 14,000 × g for 30 minutes at 4°C. Carefully transfer the resulting supernatant to a new centrifuge tube.

2. Spin Column 01 Pre-treatment

1) Based on the number of exosome samples to be isolated, take an equal number of Spin Column 01 and Collection Tubes (2 mL) from the kit. Remove the bottom cap of the Spin Column 01 and place it into the Collection Tube (2 mL). Use a pipette to aspirate and discard the protective liquid from the upper portion of the Spin Column 01.
2) Add 500 µL of Solution A to the Spin Column 01, centrifuge at 300 g for 1 minute, and discard the waste liquid in the Collection Tube.
3) Repeat step 2) once;
4) Place the Spin Column 01 into a new 2 mL centrifuge tube, ready for the next step in exosome isolation.

Note: If the sample volume does not exceed 0.2 mL, a 1.5 mL centrifuge tube can also be used.

3. Exosome Isolation

1) Add the pretreated sample (50-500 µL) to the previously prepared Spin Column 01, and centrifuge at 4°C, 300 g for 1 minute.
   a. If the sample volume exceeds 0.5 mL, it is recommended to concentrate the sample using a 50 kDa ultrafiltration tube to 0.5 mL, ensuring the concentration factor does not exceed 20 times.
   b. For high-viscosity samples, such as plasma, serum, or pleural and peritoneal fluids, dilute with an equal volume of deionized water before adding to Spin Column 01.
   c. For samples with high levels of impurities, such as plasma, serum, pleural and peritoneal fluids, and milk, it is recommended to dilute with PBS 10 times, then concentrate using a 50 kDa ultrafiltration tube by a factor of 10. Afterward, add PBS in a 10-fold volume relative to the remaining liquid, concentrate again by 10 times, and then proceed to separate exosomes with Spin Column 01.
   d. During centrifugation, maintain a centrifugal force of 300-500 g (increasing the centrifugation force appropriately for more viscous samples) to achieve effective exosome separation.
2) Remove Spin Column 01, and the liquid in the centrifuge tube contains the separated exosomes. Take a portion for analysis and store the remaining solution at -80°C.

   Note: Depending on the impact of the exosome solution's properties on subsequent experiments, consider using an ultrafiltration tube (MWCO: 10 kDa) to exchange the solution with an alternative buffer.

(二) Removal of Free Dye

1. Spin Column 01 Preparation

1) Based on the number of samples requiring dye removal, take an equal number of Spin Column 01 units and 2 mL Collection Tubes from the kit. Remove the bottom cap of each Spin Column 01, place it into a 2 mL Collection Tube, and aspirate the protective liquid from the top of the Spin Column 01 using a pipette.
2) Add 500 µL of Solution A to the Spin Column 01, centrifuge at 300 g for 1 minute, and discard the waste liquid in Collection Tube.
3) Repeat step 2) once.
4) Place the Spin Column 01 into a new 1.5 mL centrifuge tube, preparing it for the subsequent exosome isolation step.

2. Removal of Free Dye

1) Add the fluorescent dye-labeled exosome sample (50–200 µL) into the pre-treated Spin Column 01. Centrifuge at 300 g for 1 minute at 4°C.
2) Remove Spin Column 01. The liquid in the centrifuge tube contains the purified exosomes, ready for subsequent experiments.