

Exosome Isolation and Purification Kit (Biofluids)

Cat. No.: E1526241 | Pack size: 3T | Storage: Room temperature

Overview

Exosomes are small vesicles (30–150 nm) secreted by cells, containing RNA and proteins, and are abundantly present in body fluids such as blood, saliva, urine and milk. Exosomes act as intercellular messengers to deliver effector molecules or signaling molecules between specific cells. Nevertheless, their structure, composition of effector substances and the biological pathways they participate in remain poorly characterized. Intact exosome particles are indispensable for research on the biological functions of exosomes. Conventional ultracentrifugation is plagued by complicated workflows, high requirements for specialized equipment and cumbersome operation. This exosome isolation and purification kit adopts optimized formulations, which is applicable to exosome extraction from body fluids including cerebrospinal fluid, amniotic fluid, milk, saliva, bronchoalveolar lavage fluid and nasal lavage fluid. Equipped with a purification filter device, the kit enables rapid and efficient acquisition of high-purity exosome pellets. The purified products can be applied to transmission electron microscopy (TEM), nanoparticle tracking analysis (NTA), nucleic acid analysis, protein analysis, cell-based assays and animal experiments.

Product Components

Component No.	Component Name	Appearance	3T	Storage
E1526241A	Exosome Concentration Solution	Liquid	1.8 mL	RT
E1526241B	Exosome Purification Filter	—	3 EA	RT

Required Materials

High-speed centrifuge, vortex mixer, 50 mL centrifuge tubes, 1.5 mL centrifuge tubes, sterile 1×PBS buffer.

Operating Protocol

I. Sample Pretreatment

1. Sample handling and thawing: For frozen samples, take them out from the refrigerator and thaw in a 25 °C water bath, then place fully thawed samples on ice. Fresh samples shall be placed on ice immediately after collection.

2. Recommended initial sample volume: For single isolation, the minimum recommended volume for bronchoalveolar lavage fluid, ascites and other body fluids is 1 mL; 2–3 mL is preferred per isolation run.

3. Centrifugation to remove cell debris: Transfer samples into centrifuge tubes and centrifuge at 3,000 $\times g$ (~6,200 rpm*) for 10 min at 4 °C to remove cell debris. Note: If abundant pellets are observed, repeat centrifugation at 3,000 $\times g$ for 10 min until no obvious precipitate exists, and retain the supernatant after each round of centrifugation.

*Calculated for small centrifuges with an effective radius of approximately 7 cm (for tubes ≤ 2 mL), the same applies hereinafter.

4. Centrifugation to remove impurity fragments: Transfer the supernatant to a new centrifuge tube and centrifuge at 12,000 $\times g$ (~12,400 rpm) for 10 min at 4 °C to eliminate residual impurities and debris.

5. Supernatant transfer: Transfer the clarified impurity-free supernatant into a new centrifuge tube.

II. Exosome Isolation

1. Supernatant pretreatment: Add Exosome Concentration Solution (ECS reagent) to the clarified supernatant. Refer to the table below for detailed dosing standards; volumes for other sample input amounts can be calculated proportionally based on the listed ratios.

Sample Name	Sample Volume	ECS Volume
Bronchoalveolar Lavage Fluid (BALF)	1 mL	0.25 mL
Ascites	1 mL	0.25 mL

2. Solution Mixing: After adding ECS reagent, tightly cap the centrifuge tube and vortex thoroughly for 1 minute on a vortex mixer. Incubate the mixture at 4 °C for no less than 8 hours. Note: Extended incubation time can improve exosome yield, but the incubation duration must not exceed 24 hours.

3. Exosome Precipitation: Take out the tube containing the mixture and centrifuge at 12,000 $\times g$ (~12,400 rpm) for 30 minutes at 4 °C. Discard the supernatant completely to the greatest extent possible; the pellet is enriched with exosome particles.

4. Secondary Short Spin: Centrifuge the tube with the pellet again at 12,000 $\times g$ (~12,400 rpm) for 2 minutes at 4 °C, then thoroughly remove all residual supernatant.

5. Exosome Resuspension: Gently pipette the pellet up and down with an appropriate volume of 1 $\times$ PBS buffer until fully dissolved, then transfer the resuspension to a new 1.5 mL centrifuge tube. Recommended resuspension volume: approximately 100 μ L of 1 $\times$ PBS for every 1 mL starting body fluid sample.

6. Harvest Crude Exosome Pellets: Centrifuge the 1.5 mL tube with resuspended sample at 12,000 ×g (~12,400 rpm) for 2 minutes at 4 °C, retain the supernatant which contains enriched crude exosomes. Note: If visible precipitate remains, repeat centrifugation at 12,000 ×g for 2 minutes until no obvious pellet is observed, and collect the supernatant after each centrifugation.

III. Exosome Purification

1. Column Purification: Combine all crude exosome suspensions derived from the same type of body fluid and load them into the upper chamber of the Exosome Purification Filter (EPF column). Centrifuge at 3,000 ×g (~6,200 rpm) for 10 minutes at 4 °C. Collect the eluate at the bottom of the EPF column, which is the final purified exosome product. Note: EPF columns are disposable and cannot be reused.

2. Exosome Storage: Aliquot the purified exosomes into appropriate volumes and store long-term in a -80 °C freezer for subsequent downstream experiments.

Note:

This product is for life science research use only. It shall not be applied for medical diagnosis or any clinical purposes.

Specifications

Item	Information
Synonyms	Body Fluid-Derived Exosome Extraction & Purification Kit
Specifications & Purity	BioReagent, Sterile
Sample type	body fluids

Storage and Shipping

Item	Information
Storage	Room temperature
Shipped In	Normal
Stability And Storage	Store at RT long term (24 months).

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