Biorbyt exosome isolation kit (for Cell Culture Media)

Cat #: orb1566759 (manual)

Size: 50 ml

Product Composition

Reagent name	Specifications
Biorbyt exosome isolation kit (for Cell Culture Media)	50 mL
Manual	1 copy

Product Introduction

Exosomes are small membrane vacuoles (30-150 nm) containing complex RNAs and proteins that are secreted by different types of cells during culture and are abundant in body fluids, including blood, saliva, urine, and breast milk. Exosomes are considered cell-to-cell messengers that deliver effectors or signaling molecules between specific cells. However, the formation and composition of exosomes and the biological pathways involved in them are still not fully understood.

Biological studies such as exosome function and transport require the isolation of intact exosomes, and the Biorbyt exosome isolation kit (for Cell Culture Media) is a reagent specifically designed to isolate exosomes from supernatant samples from extracted cell culture media. This product provides a simple and reliable way to concentrate intact exosomes from cell culture media supernatant samples by bundling water molecules and forcing hard-to-dissolve components (i.e., exosomes) out of solution, and a large number of exosomes can be isolated from the sample by simple slower centrifugation. Compared with traditional ultra-high-speed centrifugation, the exosomes in the sample are under less pressure and can maintain a more intact form. At the same time, the extraction process takes less time, requires a lower sample input, and is more efficient. Exosomes obtained with this product can be used for a variety of downstream experiments, such as RNA analysis, high-throughput sequencing, cell co-culture, etc.

This kit is sufficient to isolate exosomes from the supernatant of 100 mL of cell culture medium.

Operation Procedure

Sample Preparation:

1. Collect the cell culture medium.
2. Centrifuge the cell culture medium at 3,000×g for 20 minutes to remove cells and debris.
3. Carefully transfer the cell culture medium supernatant obtained from step 2 to a new tube.

Exosome isolation:

1. Transfer the desired volume of cell-free medium to a new tube and add 0.5 volumes of exosome extraction reagent. (You can refer to the table below)

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Volume of cell culture medium	Extraction reagents
1 mL	500 μL
10 mL	5 mL

2. Mix the medium and exosome extraction reagent mixture by vortexing or inverting until a homogeneous solution is obtained.
3. Incubate overnight at 4 °C.
4. After incubation, centrifuge at 2~8°C, 10000×g for 1 hour.
5. Aspirate and discard the supernatant, while exosomes are contained in the pellet at the bottom of the tube (sometimes difficult to distinguish with the naked eye).
6. Resuspend exosomes using 1× PBS or appropriate buffer, refer to the table below.

Volume of cell culture medium	The volume resuspended
1 mL	25~100 μL
10 mL	100μL ~1 mL

7. Once the exosome pellet has been resuspended, it can be analyzed downstream by affinity or further purified.
8. Store isolated exosomes at 2°C to 8°C for up to 1 week, or ⩽-20°C for long-term storage.

Precautions

1. This product is suitable for the extraction of exosomes from the supernatant of various types of cell culture mediums. In order to prevent contamination caused by more bovine exosomes in FBS, the serum-free medium or exosome-free serum medium can be replaced when the cell culture is about 50%-70%, and the supernatant can be collected and used after continuing to culture for about 12~48h. Cells can also be cultured directly using exosome-free serum medium, followed by direct collection of cell culture supernatant for exosome extraction.
2. The volume of cell culture medium required for an experiment depends on the amount of exosomes in the supernatant, which may be affected by the type, state, and number of cells.
3. When centrifuging with a fixed-angle centrifuge, mark the direction in which the tube is placed. Generally, the sample has a small amount of exosomes, and the precipitate may not be visible to the naked eye after centrifugation, so after marking the direction, 1× PBS can be repeatedly pipetted towards the inner wall of the outer side of the centrifuge tube during resuspension to elut.
4. The centrifuged pellet can be resuspended in 100-200 μL of 1× PBS, or the pellet can be processed directly using the reagents used in the next experiment. For example, the lysate can be used directly to resuspend the pellet by pipetting and then entering the RNA and protein extraction process.

Storage conditions

Store at 4℃, valid for one year.

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