

TransExo[®] Cell Media Exosome Kit

Please read the data sheet carefully prior to use.

Cat. No. FE401

Storage: at 2-8°C for one year.

Description

TransExo[®] Cell Media Exosome Kit is designed to extract and purify exosomes from the cell supernatant. The obtained high-purity and high-activity exosomes can be used in Western Blot, qPCR, particle size, transmission electron microscopy, and other detection methods.

Highlights

- Easy-to-use. No ultracentrifugation is required.
- High sensitivity.

Kit Contents

Component	FE401-01
Exosome Precipitation Solution-Cell Media (EPS-C)	25 ml
Exosome Resuspension Solution-Cell Media (ERS-C)	10 ml

Procedures

1. Centrifuge the cell supernatant sample at 3,000×g for 30 minutes at 2-8°C to remove cells and debris. Collect the supernatant.
2. Mix the sample and EPS-C upside down according to the recommended dosage and ratio in the table below, and let it stand overnight at 2-8°C.

Sample Volume	EPS-C	Recommended Centrifuge Tube Specification
1 ml	0.5 ml	1.5 ml
10 ml	5 ml	15 ml

3. Mark the position of the pellet on the centrifuge tube containing the mixture that has been allowed to stand overnight. Place the centrifuge tube in the centrifuge according to the mark, and align the mark on the outside of the centrifuge rotor. Centrifuge the cell supernatant sample at 3,000×g for 30 minutes at 2-8°C to pellet exosomes.
4. Discard the supernatant and collect the pellet. Centrifuge at 10,000×g for 5 minutes. Carefully discard the remaining supernatant with a 200 μl pipette tip, avoiding the pellet position. It is recommended to centrifuge again for 5 minutes. And carefully discard the remaining supernatant with a 10 μl pipette tip, avoiding the pellet position.
5. Add 30 μl ERS-U solution to the exosome pellet. Resuspend the pellet by gently pipetting to obtain exosomes. If the exosomes precipitate too much during the experiment, the resuspension volume can be appropriately increased until the precipitate is completely dissolved.

Notes

- Fresh sample preparation is recommended. Avoid repeated freezing and thawing. If the collected cell supernatant cannot be used for the next experiment immediately, it is recommended to store it at -80°C and complete the exosome extraction within one week.
- Different types of cells secrete different amounts of exosomes. For the initial extraction, it is recommended to set up a pre-experiment to explore the optimal amount of sample used.
- To extract exosomes from a large number of cell supernatant samples, they can be concentrated with a 100 kDa ultrafiltration tube and extracted according to the above steps.



- The dissolved volume of ERS-C solution after exosome extraction is added proportionally according to the volume of cell supernatant before concentration. For example, 300 μ l of ERS-C solution is added to the exosomes extracted after 10 ml of cell supernatant is concentrated and resuspended. Excessive concentration of exosomes may reduce the solubility, and the dissolution volume can also be adjusted according to the needs of the reagent. It is recommended to design a pre-experiment to determine the optimal volume.
- When using an angle rotor centrifuge, the exosomes pellet will adhere to the tube wall, which should be resuspended the exosomes on the tube wall with care. Some samples have a small amount of exosomes, so precipitation is invisible during the extraction process. The precipitation location can be marked before centrifugation to facilitate subsequent experiments.
- It is not recommended to use a large volume centrifuge tube for centrifugation. The large wall area will lead to insufficient resuspending of exosomes, which may easily cause losses and reduce the extraction efficiency.
- If the laboratory does not have a centrifuge of the corresponding specifications, it is recommended to add EPS-C according to the proportion, then divide the mixture into 1.5 ml centrifuge tubes for centrifugation, and finally combine the extracted exosomes from each tube. For example, extract 5 ml of cell supernatant exosomes. 2.5 ml of EPS-C can be added according to the proportion and incubated overnight into 5 tubes for centrifugation extraction, and finally combined into one tube, or centrifugation can be repeated in the same EP tube until all centrifugation is complete.
- It is recommended to remove the residual EPS-C after centrifugation, otherwise it may affect the Western Blotting.
- If the extracted exosomes are subsequently applied to Western Blot or qPCR, it is recommended to directly use the lysis buffer to lyse the exosomes precipitation after the end of step 4. The extracted exosomes can be aliquoted and stored at -80°C to avoid repeated freezing and thawing.
- If the follow-up application involves the detection of morphological structure such as activity, particle size or integrity after the exosomes are extracted, the freshly extracted exosomes should be used as soon as possible. The exosomes mentioned are recommended to be filtered with a 0.45 μm filter and stored at $2-8^{\circ}\text{C}$, and tested within 24 hours.

FOR RESEARCH USE ONLY

