Exosomes Affinity-based Isolation Kit (EVlent™)

JOT-EV-02-05-01

Product Overview

This kit is improved and optimized based on the magnetic beads capture method, adding three types of magnetic beads, CD9/CD81/CD63, which can specifically capture the marker proteins on the surface of EVs. Taking CD9 magnetic beads as an example. The functionalized magnetic beads in EVlent™ can efficiently capture EVs onto modified beads assembled with EVs characteristic protein CD9 antibodies. And these beads have a unique affinity for the CD9 protein on the surface of EVs. It can meet the needs of a variety of downstream experiments such as exosome-cell co-culture experiments, proteomics studies, NGS high-throughput sequencing and WB detection.

Components and Storage Conditions

Reagents	Specifications	Storage Conditions
EVlent™ magnetic beads	1 mL	4℃
Incubation buffer Ⅰ	50 mL	RT
Incubation buffer Ⅱ	50 mL	RT
Washing Buffer	50 mL	4℃
Elution Buffer	20 mL	RT

Protocol

1. Centrifuge the plasma sample (2000g, 5-10 minutes) to obtain the supernatant for further use and store it for a long time at -80℃ to avoid repeated freeze-thaw.
2. Put 200μL of plasma into the microcentrifuge tube and add 800μL of precooled Incubation buffer I, then mix well at room temperature.
3. Add 40μL of EVlent™ magnetic beads, place it on vertical mixer to shake thoroughly at room temperature and incubate for 2-3 hours.

4. Place the microcentrifuge tube in the magnetic stand for 3min. Separate and remove the supernatant. Then add 1mL of Incubation buffer II. Flip it upside down 20 times and place the microcentrifuge tube in the magnetic stand for 3min and remove the supernatant.
5. Add 1mL of Washing Buffer, flip it upside down 20 times, and place the microcentrifuge tube in the magnetic stand for 3min and remove the supernatant.
   Repeat this step 2 times.

The following steps need to be selected according to the downstream experimental needs.
If the downstream is for proteomics research (preferred recommendation)

a. Add 50µL of Lysis buffer (JOT-EV01-03-01) to the beads obtained in step 5.
b. Follow the routine pre-treatment steps in the laboratory, or use the Proteomics preprocessing kits (JOT-EV01-03-01) for fast and efficient processing.

If the downstream is for WB characterization study

a. Add 4×LDS Sampling Buffer to the beads obtained in step 5 (B0007, Invitrogen).
b. 5min incubation at 95°C, then magnetically separate the beads and aspirate the supernatant for gel electrophoresis.

If downstream is for NTA and TEM detection

a. Add 100µL of Elution Buffer, vortex for 10min, and then place the microcentrifuge tube in the magnetic stand for 3min and collect the supernatant.
b. Add 100µL of Elution Buffer again, vortex for 10min, and then place the microcentrifuge tube in the magnetic stand for 3min, collect and consolidate the supernatant.

Note: If NTA/TEM is used for downstream, increase the volume of plasma sample as appropriate, 500 µL is recommended for better results.

Precautions and Disclaimer

Magnetic beads will settle when left to sit. Please shake the beads gently and well before each use to keep the beads evenly suspended.

Elution Buffer has an irritating odour, please try to use it in a fume hood.

During the storage and use of magnetic beads, operations such as freezing, drying, and high-speed centrifugation should be avoided, as this can damage the structure of the magnetic beads and affect its protein binding ability.

This product is limited to scientific research use by professionals and should not be used for clinical diagnosis or treatment, nor for food or medicine.

Life Science Production

The Science Centre, 1 Blaydon Road, Sandy, Bedfordshire, SG19 1RZ

Tel: +44 (0) 1234 889180 Email: sales@lifesciencegroup.co.uk Web: www.lifesciencegroup.co.uk