Minute™ Hi-Efficiency Exosome Isolation Reagent

Cat. No. EI-027

Description

Exosomes are vesicles secreted by cells. They are present in a variety of body fluids such as serum, ascites, spinal cord fluid, urine, and saliva. Cultured cells also secrete significant number of exosomes. The size distribution of exosome is ranging from 30-120 nm. The biological function of exosomes is believed to serve as intercellular messengers. Filtration and ultracentrifugation are classical ways of isolating and enriching exosomes. This is a tedious process and requires special equipment. Another common way for enrichment of exosome is through precipitation. Currently major commercial kits are PEG-based. EI-027 is designed to precipitate total exosomes from biofluids using a high efficacy, non-PEG based reagent for exosome precipitation. This kit is suitable for routine biofluid samples using the same reagent and similar protocol.

Packaging: Exosome Precipitation Reagent: 20 ml

Shipping and Storage: This product is shipped and stored at ambient temperature

Important: This kit can be used for enrichment of total exosomes from samples such as serum, ascites, plasma, spinal cord fluid, saliva and urine. However, there are some variations in specific steps for sample preparation, sample pre-treatment, and centrifugation force used for each specific sample. The following protocol is for multiple sample types in general. Refer to **Table 1** for specific method and g force recommended for different samples.

Protocol

Important: Shake the reagent bottle a few times to mix the contents well prior to use

1. Place your sample in a test tube and centrifuge at 2000 X g for 10 min to remove large debris. See table below for pre-treatment prior to step 1.
2. Transfer the supernatant to a fresh tube and add ½ volume of exosome precipitation reagent to the same tube (for example add 50 µl reagent to 100 µl sample)
3. After incubation, centrifuge at 4°C for a period of time specified in table 1 below. Remove the supernatant and spin at 10,000 X g for 30 seconds to 1 min to bring down liquid that may adhere to the wall of the tube. Carefully remove the residue liquid completely. Resuspend the pellet in 1x PBS (pH 7.2-7.4) or other buffer of your choice. The amount of buffer used depends on the size of the pellet (for serum sample, the amount of resuspension buffer is about 1-2 volumes of starting sample volume). In some cases, precipitated exosomes are not visible and could be attached to side wall of a test tube. Be sure to wash the wall of the tube with resuspension buffer if the exosome pellet is not visible. Resuspended exosome is now ready for downstream experiments such as isolation of RNA, Western blot and other analysis.

Table 1. Experimental conditions for different samples

Sample types	Pre-treatment	Vol.	Incubation Time step 2	X g in step 3
Serum	No	>10 µl	30 min-1h	10,000 X g 15 min
Plasma	Dilute 1:2 with PBS	>10 µl	30 min-1h	10,000X g 15 min
Ascites	No	>50 µl	30 min-1h	10,000X g 15 min
Urine and spinal cord fluid	No	>1 ml	overnight	10,000 X g 1h

Note: Plasma contains significant amount of blood coagulation related proteins **that may** interfere with exosome precipitation**.** An alternate is to treat plasma sample with proteinase K but this may result in partial loss of exosome surface proteins.