

About Exosomes.

Exosomes are small endosome derived lipid nanoparticles (50-120 nm) actively secreted by exocytosis by most living cells. Exosome release occurs either constitutively or upon induction, under both normal and pathological conditions, in a dynamic, regulated and functionally relevant manner. Both amount and molecular composition of released exosomes depend on the state of a parent cell. Exosomes have been isolated from diverse cell lines (hematopoietic cells, tumor lines, primary cultures, virus infected cells) as well as from biological fluids in particular blood (e.g. serum and plasma from cancer patients) and other body fluids (bronchoalveolar lavage fluid, pleural effusions, synovial fluid, urine, amniotic fluid, semen, saliva etc). Exosomes have pleiotropic physiological and pathological functions and an emerging role in diverse pathological conditions such as cancer, infectious and neurodegenerative diseases.

Lyophilized Exosomes Standards.

HansaBioMed's purified lyophilized exosomes are isolated through a combination of ultracentrifugation and microfiltration procedures. Exosomes are subsequently quantified and validated for overall protein content and particle number by Nanoparticles Tracking Analysis (NTA, NanoSight). Lyophilization does not alter the stability of exosome proteins and nucleic acids, in comparison to other storage methods, including storage of fresh exosomes at -20°C. Lyophilized exosomes are easy to ship and stable for long term storage (up to 36 months).

Types of Exosome Standards available:

- Lyophilized exosome standards from human Biofluids (plasma, serum, urine, saliva) of healthy donors.
- Lyophilized exosomes from cell culture media (COLO1, MM1, BLCL21, HCT116, SK-N-SH, U87, PC3, BPH-1, DAUDI, A549, K562, mouse cell B16F10).
- Lyophilized Exosome Standards size available: 100 µg and 30 µg, sold in packages of 2, 4 or 6 vials.

Procedure for Exosome Standards reconstitution.

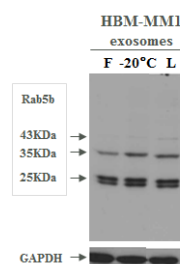
- Reconstitute Lyophilized Exosome Standard by adding deionized water, 100 µl for Lyophilized Standard 100 µg and 30 µl for Lyophilized Standard 30 µg, to get a final concentration of 1 µg/µL. Different volumes of deionized water for exosomes reconstitution can be chosen by the users in according with the desired final concentration. Resuspend exosomes pipetting the solution up and down 10-15 times, avoiding bubbles. Vortex the reconstituted standard for 60 seconds
- Briefly centrifuge the tubes containing the standard to ensure that the solution is collected at the bottom of the tube. Pipette the solution up and down 10 times, avoiding the introduction of bubbles. After this step, the standard is ready to use.

Storage.

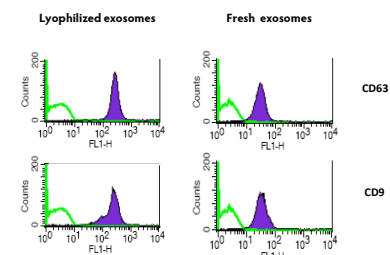
- Lyophilized Exosomes can be stored for 36 months at 4°C.
- Reconstituted Exosome Standards are not suitable for long term conservation at room temperature; use them within 2 hours after reconstitution. The remaining reconstituted solution should be aliquoted into polypropylene vials (preferably low binding) and stored at -20°C for up to one month or at -80°C for up to six months. Strictly avoid repeated freeze-and-thaw cycles.

Performance.

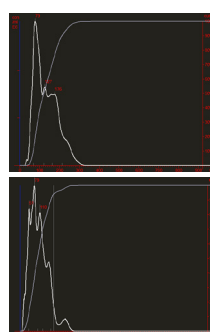
We have compared the effects of lyophilization on the preservation of exosomal proteins with respect to other storage methods such as storing fresh exosomes at -20°C and confirming their stability over 36 months at 4°C. Lyophilization do not substantially affect exosome count or biomarker expression compared to other storage methods (Fig 1, 2, 3, 4). Comparing different storage methods of exosome standards (fresh vs. frozen vs lyophilized) with an anti-CD81 ELISA assay, the loss of signal compared to fresh material is minimal when using lyophilized exosomes (CV - 15%) (Fig 4).



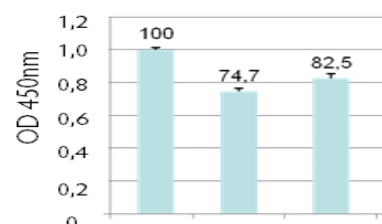
1. Western Blot comparison of exosomal markers on fresh (F), frozen (-20°C) and lyophilized exosomes (L)



2. Comparison of exosomal markers on fresh and lyophilized exosomes.



3. Comparative Nanosight analysis of freshly purified (right panel) and lyophilized plasma exosomes (left panel).



4. ExoTEST™ comparative detection of CD81 on HMB-MM1 derived exosomes:
1- Fresh exosomes; 2- Frozen exosomes (-20°C); 3- Lyophilized exosomes

Related products.

Products	Catalog Numb
ExoTEST™ Ready To Use Kit for Overall Exosome capture and quantification from Biological fluids	HBM-RTK-POF/##
ExoTEST™ Ready To Use Kit for Overall Exosome capture and quantification from human Serum	HBM-RTK-POS/##
ExoTEST™ Ready To Use Kit for Overall Exosome capture and quantification from Cell culture supernatant	HBM-RTK-POC/##
ExoTEST™ Ready To Use Kit for Tumor-derived Exosome enrichment and quantification from Biological fluids	HBM-RTK-PTF/##
Exo-Total RNA Extraction Kit	HBM-RNA-BOF-###/##
TumorExo-Total RNA Extraction Kit	HBM-RNA-BTF-###/##

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