### Lyophilized Exosome HansaBioMed Standards

The best standards for exosome research

Highly pure lyophilized Exosome Standards from human biofluids (plasma, serum, urine, saliva) and different cell culture media.

### Save time and get pure exosomes for your research

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).

Cat Code	Source		
Lyophilized Exosome Standards from Human Biofluids			
HBM-PEP	Lyophilized Exosome from Human Plasma of healthy donors		
HBM-PES	Lyophilized Exosome from Human Serum of healthy donors		
HBM-PEU	Lyophilized Exosome from Human Urine of healthy donors		
HBM-PESL	Lyophilized Exosome from Human Saliva of healthy donors		
Lyophilized Exosome Standards from Cell Culture Media			
HBM-COLO1	Lyophilized Exosomes from COLO1 cell line (Human colon carcinoma)		
HBM-MM1	Lyophilized Exosomes from MM1 cell line (Human melanoma)		
HBM-BLCL	Lyophilized Exosomes from BLCL21 cell line (EBV transformed lymphoblastoid B cells)		
НВМ-НСТ	Lyophilized Exosomes from HCT116 cell line (Human colon carcinoma)		
HBM-U87	Lyophilized Exosomes from U87 MG cell line (Human glioblastoma astrocytoma)		
HBM-SK	Lyophilized Exosomes from SK-N-SH cell line (Human neuroblastoma)		
НВМ-РС3	Lyophilized Exosomes from PC3 cell line (Human Prostate adenocarcinoma grade IV)		
НВМ-ВРН	Lyophilized Exosomes from BPH-1 cell line (Human being prostatic hyperplasia)		
HBM-DAUDI	Lyophilized Exosomes from DAUDI cell line (Human Burkitt Lymphoma)		
HBM-A549	Lyophilized Exosomes from A549 cell line (Lung carcinoma)		
НВМ-К562	Lyophilized Exosomes from K-562 cell line (Pleural effusion, leukemia chronic myelogenous)		
HBM-B16F10	Lyophilized Exosomes from B16F10 mouse cell line (mouse melanoma cell line)		
Lyophilized Exosome Standards are sold in vials containing 100 µg or 30 µg of total protein (Particles/ml > 1x10^10) HBM can provide, upon request, Exosome Standards and Cell Lysates from more than 100 different tumor cell lines. Contact us at info@hansabiomed.eu			

### **Applications**

- Assay calibration
- Control (spike-in) for
  exosome quantification.
- Protein marker analysis using different techniques
- Extraction and analysis of exosome nucleic acid
- Standardized positive controls for immunocapture performance evaluation
- Flow cytometry
- Electron microscopy

### Advantages

- High quality purified exosomes
- Easy to reconstitute
- Easy to ship and store (+4°C)
- Long term storage stability (36 months)
- Exosomes available from a large cell line bank (over 100 cell lines).

## Lyophilization is the ideal method for preparing and preserving exosome stability for long-term storage at +4°C $\,$



markers on fresh (F), frozen (-20°C) and

lyophilized exosomes (L)



2. Comparative NanoSight analysis of freshly purified (right panel) and lyophilized plasma exosomes (left panel).

Figures 1 and 2 compare the effect of lyophilization vs freezing at -20°C, showing no difference in protein expression at WB and exosome integrity at NanoSight respectively.

Sitar, Simona, et al. "Size characterization and quantification of exosomes by asymmetrical-flow field-flow fractionation." Analytical chemistry (2015).

Gardiner, Chris, et al. "Measurement of refractive index by nanoparticle tracking analysis reveals heterogeneity in extracellular vesicles." Journal of extracellular vesicles 3 (2014). Ferrante, Sarah C., et al. "Adipocyte-derived exosomal miRNAs: a novel mechanism for obesity-related disease." Pediatric research 77.3 (2014): 447-454.

### HBM-Exosome Standards are suitable for many applications in exosome research

Figure 3 shows a profile of common protein markers in exosomes purified from human plasma. HBM-PEP-30 standards are used for detection of each marker in ELISA assay, by using HBM immunoplate for Overall exosome capture from human plasma. HBM Exosome Standards are also suitable for exosomal nucleic acid extraction and downstream analyses. Figure 4 shows amplification of β-Actin transcript from total RNA obtained from HBM-COLO-100, HBM-MM1-100 and HBM-BLCL21-100 standards. RNA extraction was performed using the HBM-RNA extraction kit.



3. Expression of common exosome protein markers on plasma purified exosomes (HBM-PEP-30)



4.  $\beta$ -Actin transcript amplification from total RNA extracted from HBM-Exosome Standard COLO-100, MM1-100, BLCL-100

# HBM Exosome Standards guarantee higher purity and better performances over competitors

HBM Exosome Standard from human serum (HBM-PES) were compared to human serum exosome standard from a Competitor.Exosomal markers CD9 and CD63 from 50  $\mu$ g of both serum exosome standards were detected using an ELISA assay (fig 5). HBM Standard (50  $\mu$ g) generated the highest signal, whereas the signal from Competitor Standard (50  $\mu$ g) was lower than 30  $\mu$ g of HBM Standard. In a second test, 50  $\mu$ g of HBM and Competitor's Standards were serially diluted to design a standard curve for exosome quantification (fig 6). The standard curve generated with HBM Standard was linear (R<sup>2</sup> = 0,9876) within the concentration range and suitable for exosome quantification.

Characteristics	HansaBioMed Exosome Standard	Competitor's Exosome Standards
Amount per vial	100 µg	50 µg
Method of isolation	Ultracentrifuge	Precipitation Reagent
Nanoparticles/ml (average)	> 1x10^10	>1x10^6
Nanoparticles/ml in human serum exosomes (50 µg)	3,75x10^10 p/ml	2,29x10^9 p/ml
Final form	Lyophilized	Frozen
Storage temperature	4°C (lyophilized)	-20°C
Expire time	36 months	24 months
Price (1 vial)	••	••••



5. ELISA quantification of HBM Exosome Standard vs Competitor's standard (human serum) for exosomal markers CD9 and CD63



6. Standard curve for exosome quantification: HBM Exosome Standard vs. Competitor's standard

HBM Exosome Standards are purified using a protocol combining **ultracentrifugation** and **microfiltration steps** that guarantees the highest purity/integrity and the lowest contamination with other microvesicles. Competitor's Exosomes are isolated by chemical precipitation, therefore other microvesicles, protein-protein or protein-RNA complexes, and cell debris, regardless of their origin, contaminate the sample affecting its purity.

#### Lyophilization and Purification Service

In addition to the products described in this leaflet HansaBioMed also provides services for exosome purification, quantification at NanoSight, protein profiling by ExoTEST and miRNA or DNA profiling. Our professional services are performed by scientists experienced in the exosome field and using state of art equipment.

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