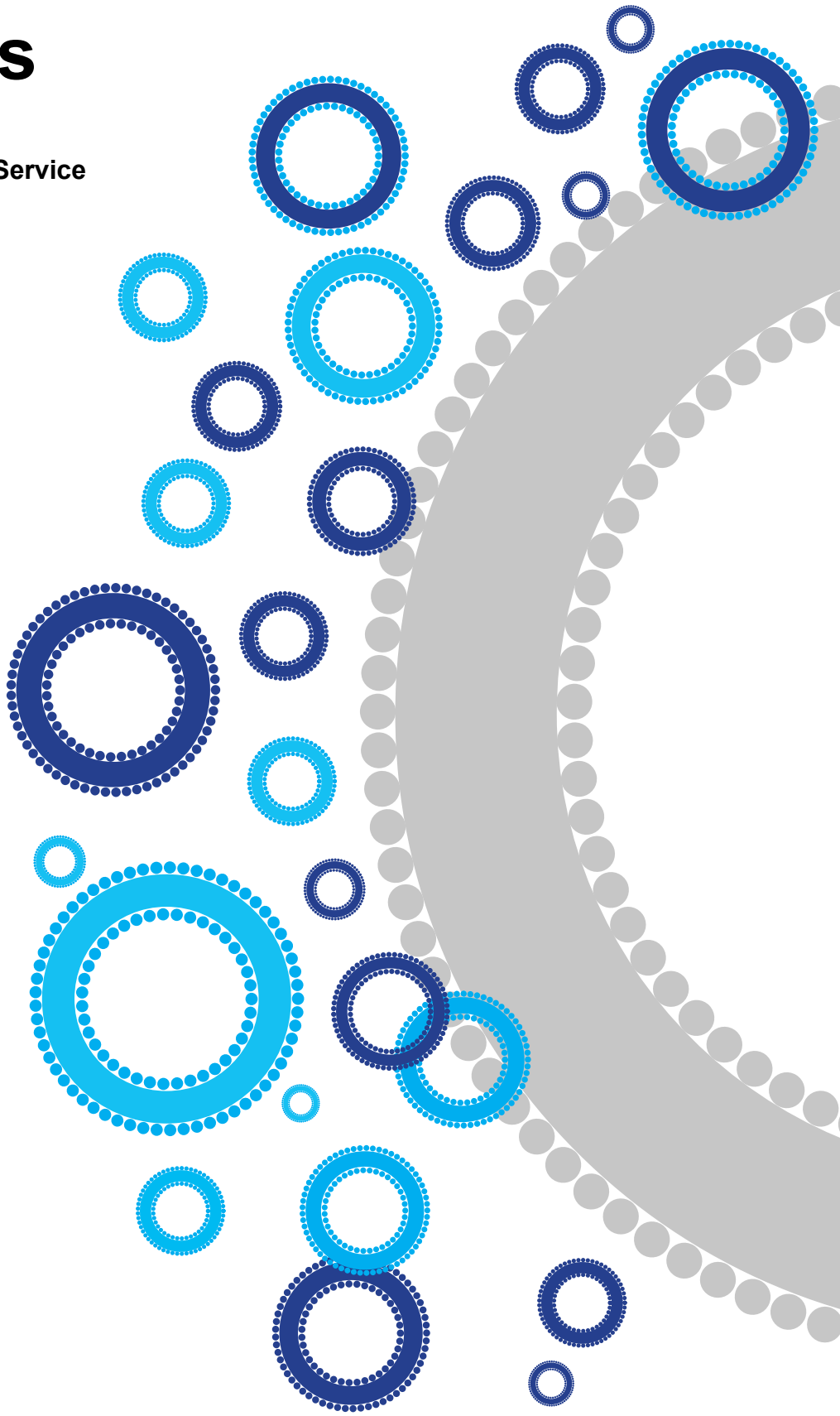
CELL guidance systems

Exosomes

Purification I Detection
Purified Exosomes I NTA Service



Exosome Experts

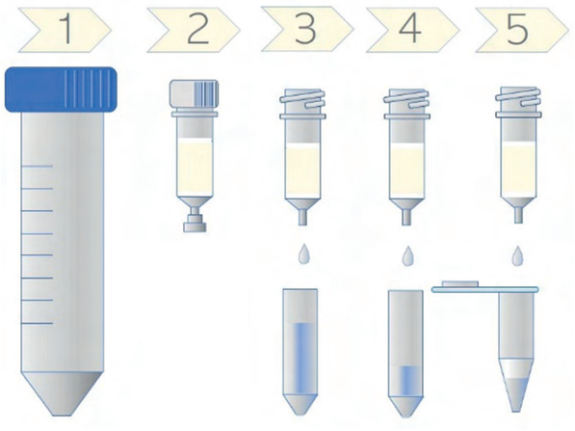
Purification

Exo-spin™ Exosome Purification: Overview

A flexible range of bench-top products for quick and easy purification of exosomes from a variety of sources

- **Excellent yields and high levels of purity**
  Exosomes with ultra-low protein and rRNA contamination.
- **No ultracentrifugation required**
  Protocol provides consistent results every time.
- **Simple and reliable**
  Isolate intact whole exosomes for functional studies.

Exo-spin™ Exosome Isolation Workflow



Step 1: Remove cells and cellular debris.

Step 2: If required, use Exo-spin™ Exosome Precipitation Buffer to precipitate exosomes.

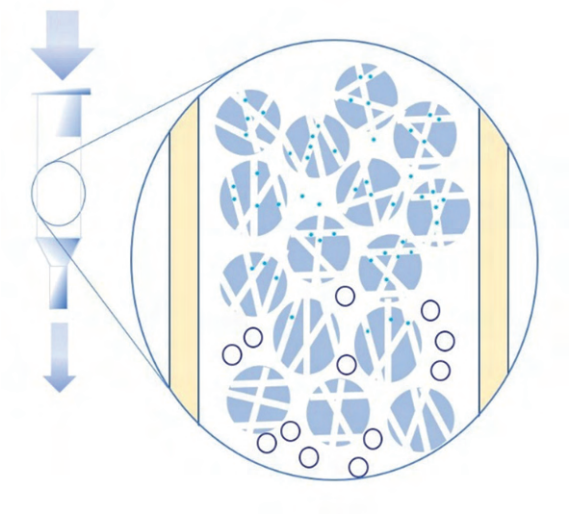
Step 3: Prepare the column by equilibrating with PBS.

Step 4: Add the pre-cleared sample or the precipitated exosome pellet resuspended in PBS to the Exo-spin™ column, elute and discard the flow-through.

Step 5: Add PBS to the Exo-spin™ column and elute your purified exosomes.

Cat code	Product name
EX01	Exo-spin™
EX02	Exo-spin™ blood
EX03	Exo-spin™ mini columns
EX04	Exo-spin™ midi columns
EX05	Exo-spin™ miniHD
EX06	Exo-spin™ buffer
EX07	Exo-spin™ 96
EX10	Exo-rack

Size exclusion chromatography (SEC) Technology



Principles of size exclusion chromotography

Exosomes run outside the beads, so elute first

Small particles and free proteins are trapped in beads

Purification

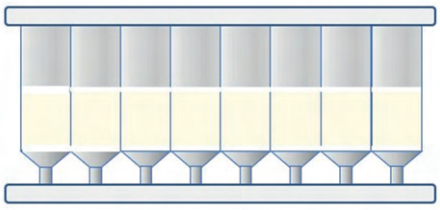
Exo-spin™ Exosome Purification: Product Range

A comprehensive range of products for isolation of exosomes from various sample types and volumes

Exo-spin™ mini columns

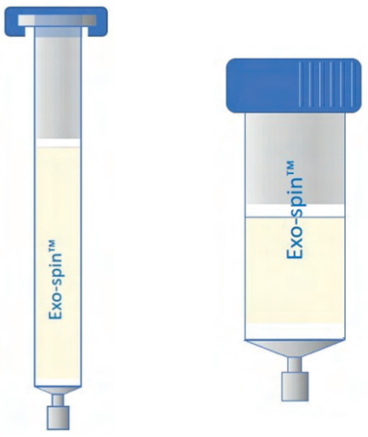
- **A simple, two-step protocol** Allows consistent and reliable purification of samples in under 2 hours.
- **Purify from blood** Isolate directly from both sera and plasma samples.
- **Process up to 50 ml per column** Combine with Exo-spin™ Exosome Precipitation Buffer to isolate from up to 50 ml of low protein biofluids.

Exo-spin™ 96



- **96-well format** Incorporates the technology of Exo-spin™ mini columns into a high-throughput system.
- **Process 8-96 samples at once** Detachable strips of 8 columns gives flexibility to your isolations.
- **Gravity protocol** Purification achieved without the need for specialist instrumentation.

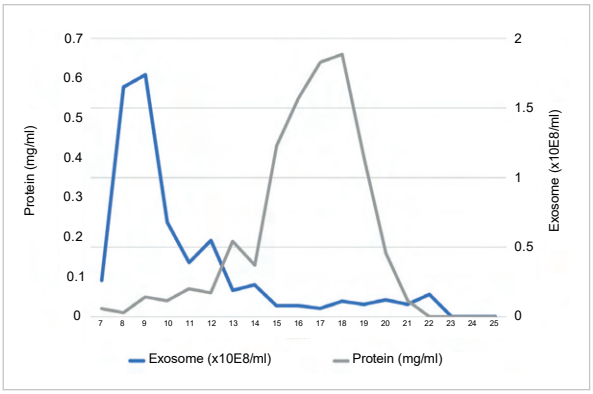
Exo-spin™ mini-HD and midi columns



- **High-resolution protocol** Optimizes yield and purity and enables finer fraction collections.
- **Process large and intermediate volumes** Isolate from up to 500 ml of low protein biofluids and 1 ml of blood.

Exosome Extraction Profiles

Exo-spin™ Midi Columns allow for gravity assisted fractionation, where exosomes are separated from the vast majority of proteins.



Exosomes were isolated using Exo-spin™ Midi Columns (Cat Code EX04) from 60 ml of conditioned medium generated by a human breast carcinoma cell line. Fractions (each of 500 µl) were collected and analysed to (1) evaluate particle numbers and (2) measure absorbance at 280 nm to evaluate protein concentration.

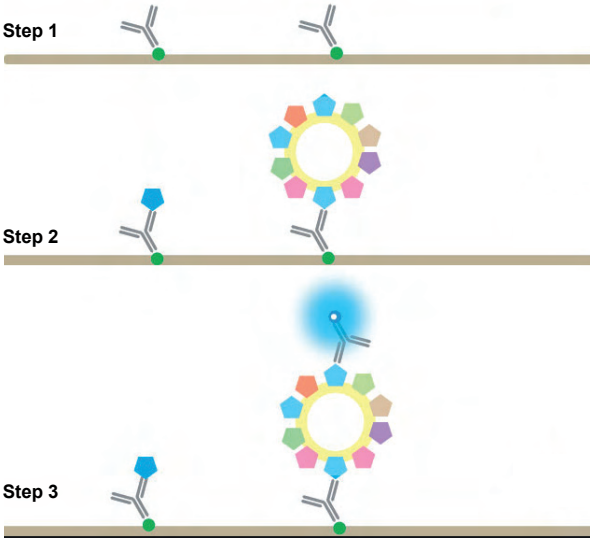
Detection

ExoLISA™ Exosome Detection Assay

An exquisitely sensitive Europium Time-Resolved Immunofluorescence assay for exosome markers

- **Simplicity**
  Assay is clear and simple allowing for high reproducibility.
- **Sensitivity**
  Europium-labelled antibodies are used for detection enabling the use of TRF.
- **Specificity**
  Only antigens displayed in multiple copies are detected.

ExoLISA™ Exosome Detection Assay



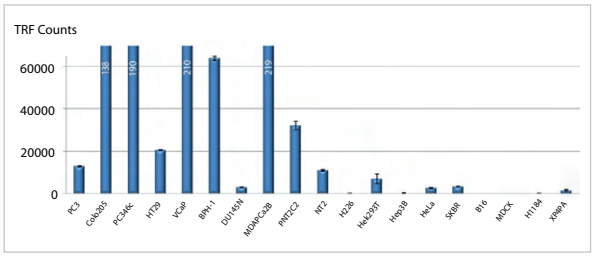
Step 1: Biotinylated antibody is bound to streptavidin coated assay plates.

Step 2: Biological samples are added. Exosomes and any free antigen are captured by the antibody.

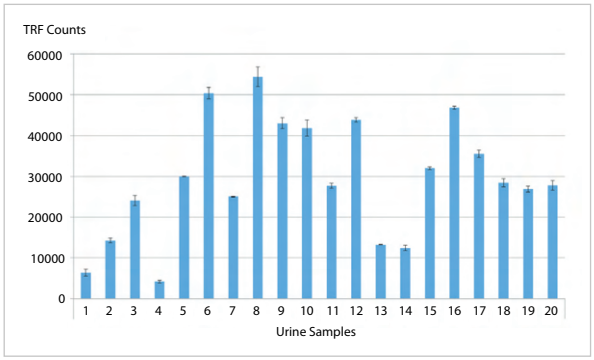
Step 3: Europium-labelled antibody of the same clone as the one used in Step 1 is added and binds specifically to exosome antigen. The epitopes of bound monomers are already occupied and not detected. Samples are read on a time-resolved fluorescence plate reader.

Cat code	Product name
EX501	ExoLISA™ CD9 Exosome Assay, 96 wells
EX502	ExoLISA™ CD63 Exosome Assay, 96 wells
EX503	ExoLISA™ CD81 Exosome Assay, 96 wells
EX-P31	Wash Buffer (Concentrate 25x), 20ml

Sample profiling



CD9 ExoLISA™ exosome assay performed for 19 different cell lines. Off-the-scale readings indicated (x1000) on individual bars. Samples generating vastly different signal intensities can be measured due to the broad signal range covered by the assay.



ExoLISA™ exosome assay analysis of 20 urine samples shows great variation in CD9 content between samples.

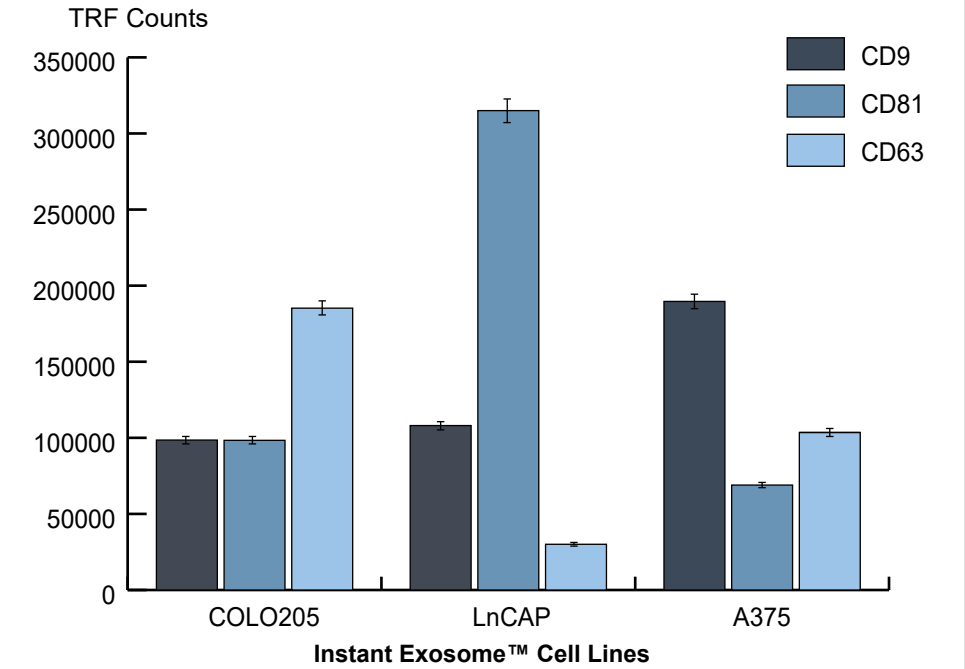
Purified Exosomes

Instant Exosomes™ to accelerate your research

A range of freeze-dried, characterized exosomes

- **High quality**
  Exosomes are isolated using a combination of size exclusion chromatography (SEC) and precipitation.
- **Stable**
  Instant Exosomes™ are stable for long-term storage.
- **Characterized**
  Each lot is characterized by ExoLISA™ exosome detection assay, nanoparticle tracking analysis (NTA) and bicinchoninic acid (BCA) assay.

Each sample is characterized by tetraspanin markers.



Commonly expressed exosomal markers assessed using the ExoLISA™ assay are shown to be differentially expressed across the Instant Exosome™ samples isolated from human cancer cell lines.

Instant Exosomes™ should not be used in a mass spectrometer or other instrumentation that may be sensitive to precipitants.

Cat code	Product name
EX301	Instant Exosomes™ from COLO205 cell line (Human colon carcinoma)
EX302	Instant Exosomes™ from LnCAP cell line (Human prostate adenocarcinoma)
EX303	Instant Exosomes™ from A375 cell line (Human malignant melanoma)

Characteristics of Instant Exosomes™	
Protein content per vial	25 µg
Method of isolation	Precipitation + SEC
Characterized by	ExoLISA™, NTA + BCA
Nanoparticles/ml (average)	1 x 10^9
State	Freeze-dried
Storage temperature	-20°C
Shelf life	12 months

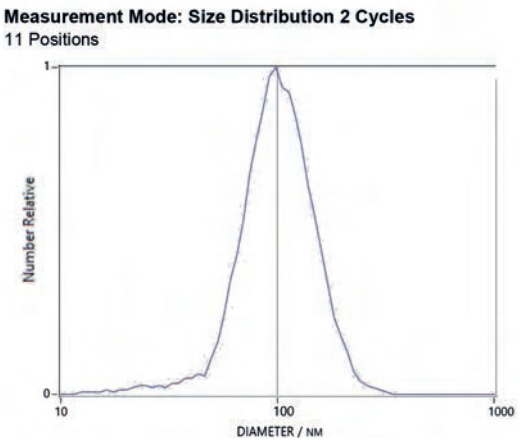
NTA Profiling

Nanoparticle Tracking Analysis (NTA) service provided using ZetaView® Instrument

Exosome characterization service for analysis of particle size and particle concentration

- **High quality**
  The service is performed in our labs by highly qualified scientists.
- **Quick turnaround times**
  Full reports are e-mailed within 5 to 10 business days.
- **Competitive price**
  Very low price per sample without the need of purchasing the equipment.

Example of an NTA report generated with the Zetaview®.



Result

Concentration:	1.2E+7 Particles / mL
Dilution Factor:	2000
Original Concentration:	2.4E+10 Particles / cm³

Quality
Average Counted Particles per Frame: 40
Number of Traced Particles: 368

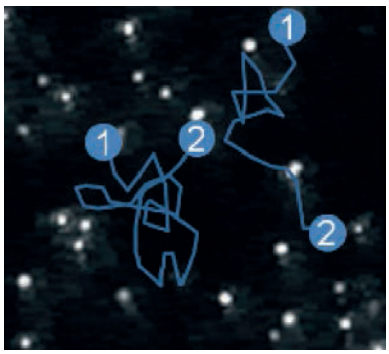
Peak Analysis (Number Absolute)

Diameter / nm	Number Absolute	FWHM / nm	Percentage
101.2	28.1	85.6	100.0

X Values

	Number	Volume
X10	61.0	89.6
X50	97.0	138.7
X90	150.5	211.6
Mean	106.0	151.2
StdDev	39.3	50.0

Zetaview® Instrument.



Individual particle movement is tracked and recorded for characterization of exosome samples.

Image provided by Particle Metrix GmbH.

Cat code	Product name
ZV-1	Analysis service set up (purchase one per order)
ZV-12	NTA analysis of a single sample

Cell Guidance Systems' reagents and services enable control, manipulation and monitoring of the cell, both *in vitro* and *in vivo*

Growth Factors
- Recombinant
- PODS® Sustained Release

Exosomes
- Purification
- Detection
- Purified Exosomes
- NTA Service

Other research products and services
- Matrix Proteins
- Small Molecules
- Cell Counting Reagent
- Lipid Quantification Assay

Cytogenetics
- Karyotype Analysis
- Array

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