sample prep

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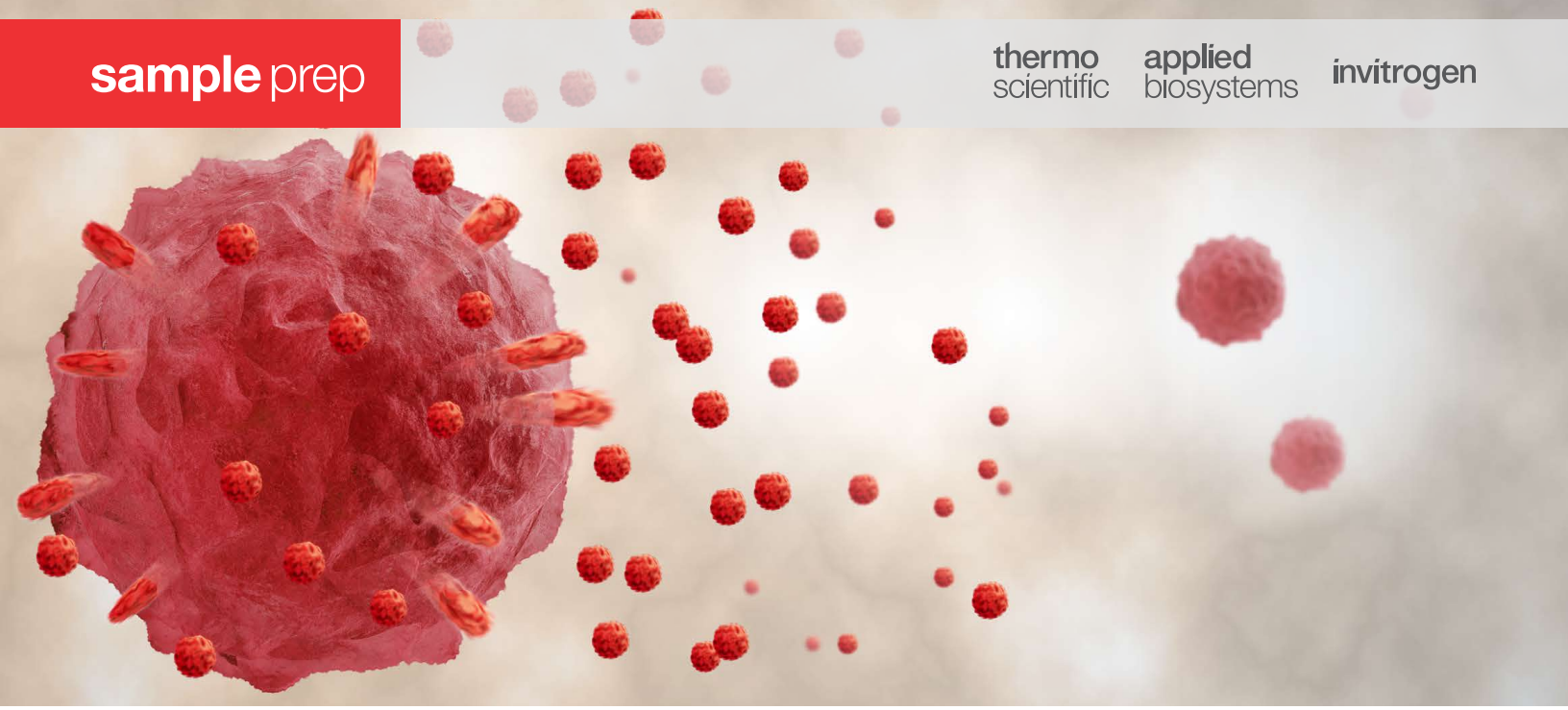

Take a closer look at exosomes

Vesicle transport and cell signaling

The 2013 Nobel Prize in Physiology or Medicine was jointly awarded to three scientists for their discovery of vesicle transport in cells, a clear indication of the importance of vesicles in physiology and their potential for applications in diagnostics and therapy.

Exosomes and other extracellular vesicles are powerful mediators and strong biomarker candidates. There's an urgent need for the development of new methods for vesicle isolation and characterization.

To address this need, we're continually expanding our product offerings. The products presented here can help simplify the overall study of exosomes, and are also applicable to translational research.

A list of published scientific papers citing the use of these products can be found at **thermofisher.com/exosomes**

Watch "Exosomes: the next small thing"

We asked ten prominent scientists to share their thoughts on the field of exosome research. Based on these fascinating interviews, a six-part video documentary was produced. This miniseries tells the story of exosome research and its impact on important research areas such as cancer, immunology, stem cell research, and potential future therapeutic and diagnostic applications.

Watch the six-part video series at **thermofisher.com/exosomesdocumentary**

Exosomes webinar

Find out about exosome isolation and monitoring as well as how leading scientists are using Invitrogen™ Dynabeads™ magnetic beads for specific isolation and analysis of exosome subpopulations.

Find out more at **thermofisher.com/exosomeswebinar**

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Products for exosome research

Exosome isolation

Invitrogen™ Total Exosome Isolation Reagents enable efficient precipitation and high recovery of intact exosomes (Figure 1).

- Typically 15–20 minutes of hands-on time
- No need for time-consuming ultracentrifugation

Ordering information

Isolation of exosomes	Quantity	Cat. No.
Total Exosome Isolation Reagent (from cell culture media)	50 mL	4478359
Total Exosome Isolation Reagent (from serum)	6 mL	4478360
Total Exosome Isolation Reagent (from plasma)	6 mL	4484450
Total Exosome Isolation Reagent (from urine)	50 mL	4484452
Total Exosome Isolation Reagent (from other body fluids)	6 mL	4484453

By targeting classical exosome surface proteins, Invitrogen™ Dynabeads™ magnetic separation technology allows you to easily pull out specific exosome subpopulations (preenrichment from cell culture) (Figure 2).

- Scalable protocol with minimal hands-on time
- Enables you to obtain a highly pure exosome subset

Ordering information

Subpopulation isolation	Quantity	Cat. No.
Human CD9 isolation	50 preps	10614D
Human CD81 isolation	50 preps	10616D
Human EpCam isolation	50 preps	10618D
Human CD63 (isolation/detection)	30 preps	10606D
Flexible streptavidin-based system (use your own biotinylated antibody)	30 preps	10608D

Exosome characterization

Preenriched exosomes can be easily visualized by flow cytometry while bound to the surface of Dynabeads magnetic beads, enabling the detection of specific exosomal markers.

- Clear and defined FSC/SSC for easier gating
- Typically less than 1 hour of hands-on time

Ordering information

Subpopulation analysis	Quantity	Cat. No.
Human CD9 detection	100 rxns	10620D
Human CD81 detection	100 rxns	10622D
Human EpCam detection	100 rxns	10624D
Human CD63 (isolation/detection)	150 rxns	10606D
Flexible streptavidin-based system (use your own biotinylated antibody)	150 rxns	10608D

Light microscopy enables the study of the release, distribution, and uptake of the labeled vesicles. Exosomal RNA and membrane components can be tagged using fluorescent dyes; unincorporated dye can be removed using specific spin columns (Figure 3).

- Fast and simple labeling protocol
- Remove all low molecular weight contaminants from exosome preparations

Ordering information

Supporting products	Quantity	Cat. No.
BODIPY™ TR Ceramide	250 µg	D7540
SYTO™ RNASelect™ Green Fluorescent Cell Stain	100 µL	S32703
Exosome Spin Columns (MW 3000)	30 columns	4484449

Isolation of exosomal cargo

The Invitrogen™ Total Exosome RNA & Protein Isolation Kit enables the isolation of:

- Highly pure total RNA (including small-RNA fraction)
- Protein and RNA from the same sample

Ordering information

Isolation of total RNA & proteins	Quantity	Cat. No.
Total Exosome RNA & Protein Isolation Kit	40 preps	4478545

Ordering information

Immunoprecipitation products	Quantity	Cat. No.
Exosome Immunoprecipitation Reagent (Protein A)	1 mL	10610D
Exosome Immunoprecipitation Reagent (Protein G)	1 mL	10612D

Using fast and gentle Dynabeads magnetic separation technology, you can isolate proteins and protein complexes from preenriched exosomes—typically in only 30 minutes.

- 10–50x concentration of exosomal proteins
- Helps to significantly minimize background

Analysis of exosomal cargo

Specific monoclonal antibodies allow for the detection of cellular and exosomal antigens (Figure 2).

- Antibodies verified for western analysis

Ordering information

Antibodies for western analysis	Quantity	Cat. No.
Anti–Human CD9	0.2 mL	10626D
Anti–Human CD63	0.2 mL	10628D
Anti–Human CD81	0.2 mL	10630D

Ordering information

Supporting products	Quantity	Cat. No.
Fetal Bovine Serum, exosome-depleted	500 mL	A272801
Fetal Bovine Serum, exosome-depleted, One Shot™ format	50 mL	A272803

Products for exosome analysis and quantitation
Continue your exosome analysis with quantitation instruments such as Thermo Scientific™ NanoDrop™ spectrophotometers and Invitrogen™ Qubit™ fluorometers, as well as products for downstream applications such as next-generation sequencing and RT-qPCR.

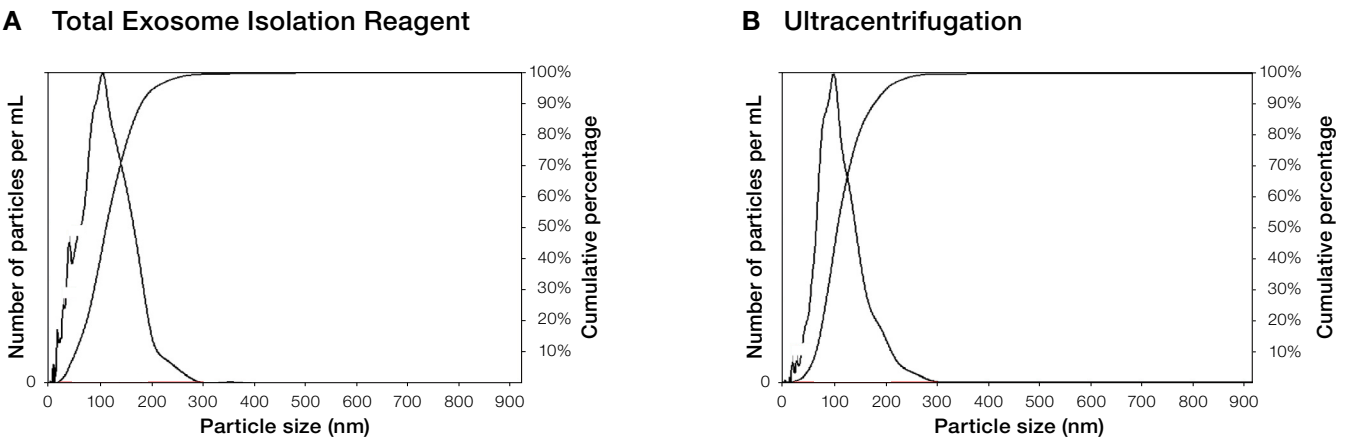

Figure 1. Analysis of exosomes recovered from HeLa culture medium. (A) Exosomes recovered with the Total Exosome Isolation Reagent (from cell culture media) have yield and size distribution comparable to **(B)** exosomes isolated by a traditional ultracentrifugation protocol with a sucrose gradient. Profiles as analyzed on a NanoSight™ LM10 instrument show all particles to be smaller than 300 nm; most are about 50–150 nm in size.

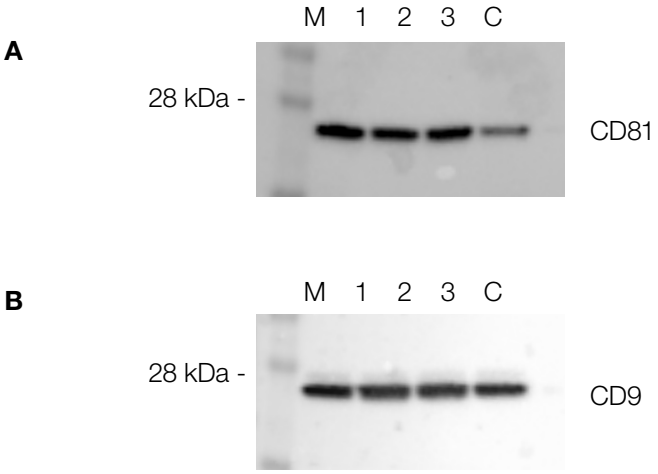

Figure 2. Analysis of exosomal preparations for CD81 and CD9 markers. Exosomes were preenriched from SW480 cell culture by ultracentrifugation. Immunoaffinity-based Dynabeads magnetic separation was used to further purify **(A)** CD81-positive or **(B)** CD9-positive exosomes from 15 µL preenriched samples (lanes 1–3). The isolated subpopulations were analyzed by western blot with antibodies against CD81 and CD9. M: molecular weight marker. C: Control with 7.5 µL preenriched exosomes without further antibody-based purification.

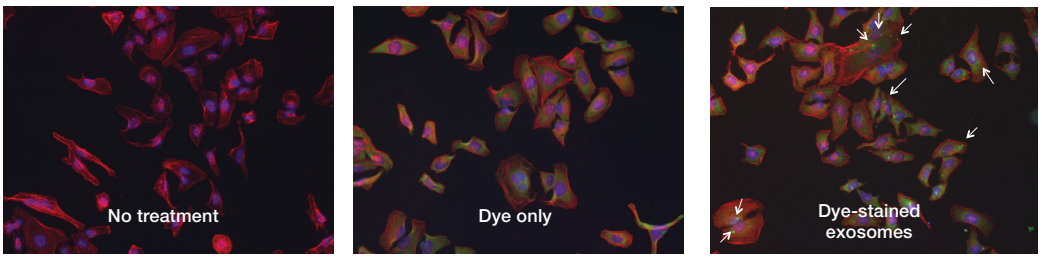

Figure 3. Uptake of labeled exosomes by HeLa cells. An Invitrogen™ FLoid™ Cell Imaging Station was used. Red: Invitrogen™ Alexa Fluor™ 594 phalloidin; blue: DAPI; green: Invitrogen™ SYTO™ RNASelect™ stain.

Find out more at **thermofisher.com/exosomes**

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