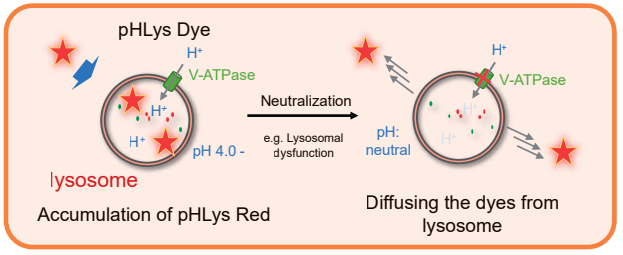
Lysosomal Analysis

LysoPrime Green / Deep Red - High Specificity and pH Resistance
pHLys Red - Lysosomal Acidic pH Detection

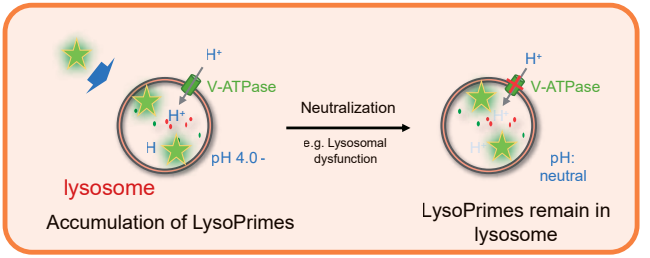
LysoPrime Green

pHLys Red

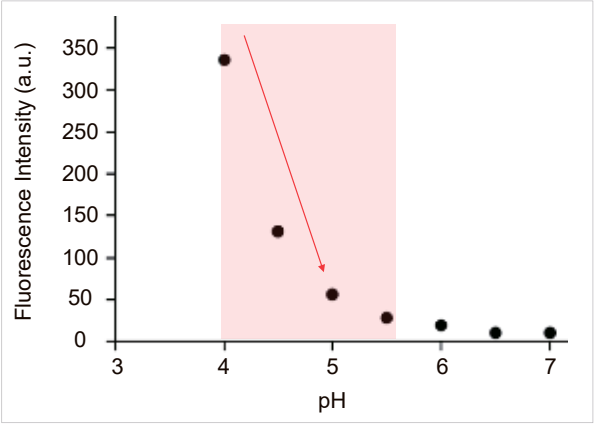
Lysosomal pH-dependent Fluorescent Probe



Lysosomal pH-independent Fluorescent Probe

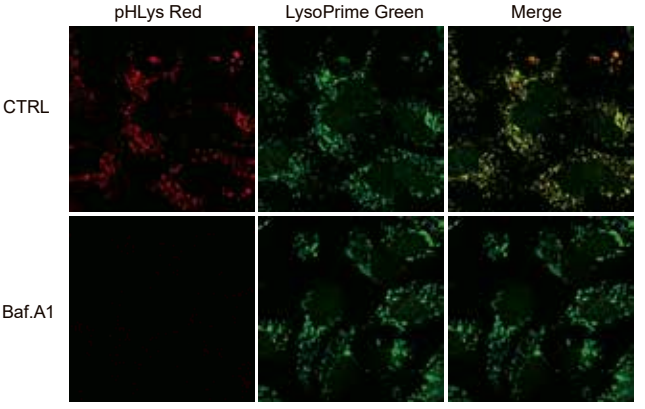


pH dependence of pHLys Red



The fluorescence intensity of pHLys Red at each pH was confirmed in vitro, and it was confirmed that the fluorescence intensity changed sensitively within the range of lysosomal pH (pH 4.0-5.5).

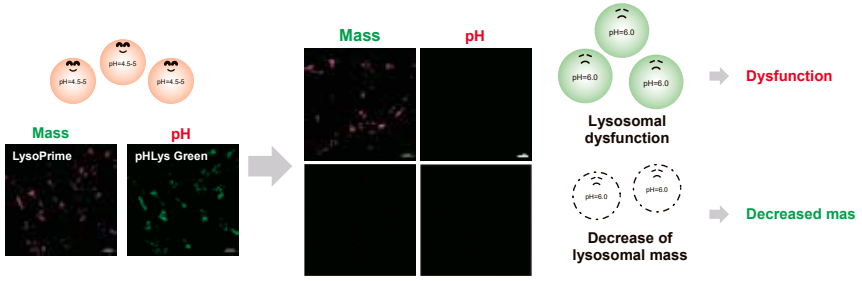
Resistance to pH changes in lysosomes



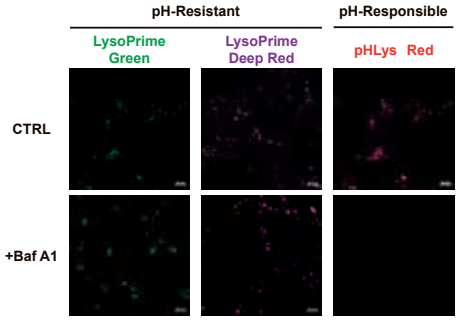
LysoPrime Green and existing dyes accumulate in acidic lysosomes, but when treated with Bafilomycin A1, a lysosomal acidity inhibitor, the existing dyes leave the lysosomes when the lysosomes are changed from acidic to neutral, resulting in a significant decrease in the fluorescence signal. On the other hand, LysoPrime Green is easily retained in the lysosome, so the decrease in the fluorescence signal is suppressed and the observation results are clearer than those of existing reagents.

Lysosomal Acidic pH Detection Kit

The kit includes lysosome staining dyes, pHLys Red/Green (pH dependent), and LysoPrime Green/Deep Red (pH-independent). The pHLys and LysoPrime dyes accumulate in the intact lysosomes. The fluorescence intensity of pHLys dyes are enhanced as the acidity increases, and weak fluorescence is observed when lysosomes are neutralized due to the lysosomal dysfunction. On the other hand, LysoPrime dyes gives stable emissions even lysosomes are neutralized. Lysosomal pH and lysosomal mass can be measured by combining these pHLys and LysoPrime dyes.

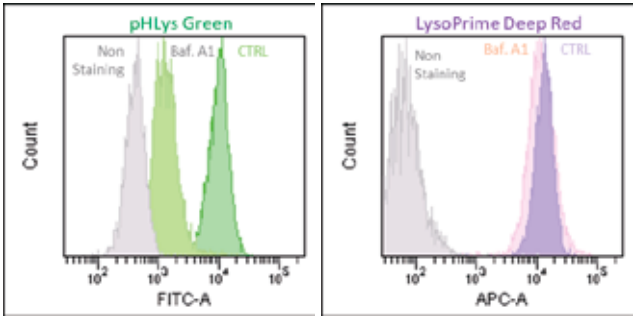


Imaging Analysis: Green/Red (#L266-10)



<Experimental Conditions>
LysoPrime Green: Ex = 488 nm, Em = 490 – 550 nm
pHLys Red: Ex = 561 nm, Em = 560 – 620 nm

FCM Analysis: Green/Deep Red (#L268-10)



<Experimental Conditions>
pHLys Green: FITC Filter (Ex = 488 nm, Em = 515 – 545 nm)
LysoPrime Deep Red: APC Filter (Ex = 640 nm, Em = 650 – 670 nm)

Description	Unit	Code
Lysosomal Acidic pH Detection Kit – Green/Red *1	1 set	L266-10
Lysosomal Acidic pH Detection Kit – Green/Deep Red *2	1 set	L268-10
LysoPrime Green – High Specificity and pH Resistance	10 µl × 1	L261-10
	10 µl × 3	L261-12
LysoPrime Deep Red - High Specificity and pH Resistance	1 tube	L264-10
	3 tube	L264-12
pHLys Red - Lysosomal Acidic pH Detection	1 tube	L265-10
	3 tube	L265-12

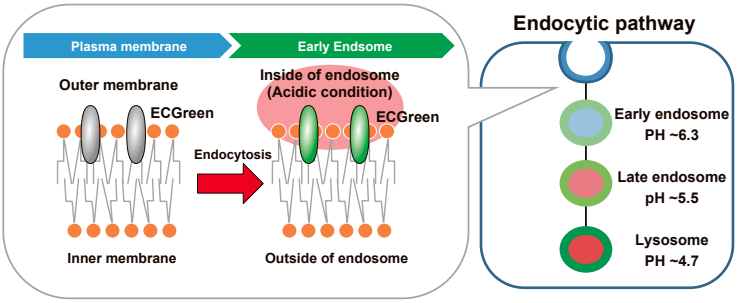
*1 Green/Red: combination of LysoPrime Green and pHLys Red, *2 Green/Deep Red: combination of pHLys Green and LysoPrime Deep Red

Endocytosis

ECGreen-Endocytosis Detection

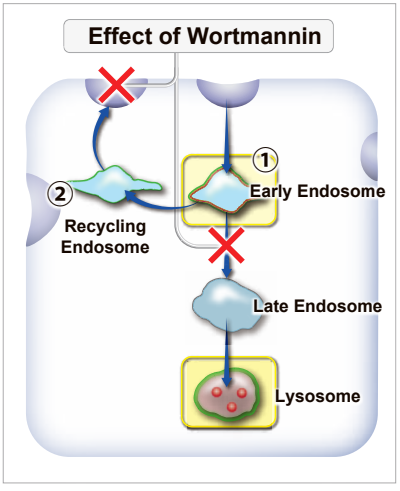
ECGreen-Endocytosis Detection is a pH dependent fluorescence dye that localizes to vesicle membrane. The visualization of endocytosis using the ECGreen is a more direct method than fluorescent analogs and allows visualization endocytosis from the stage of early endosomes.

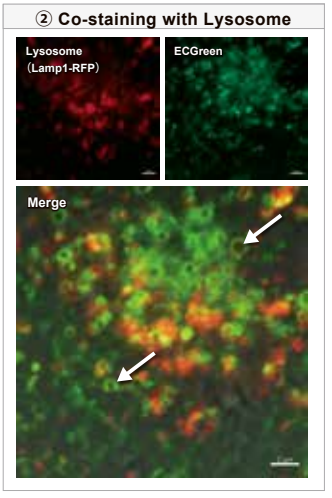
The detection mechanism of endocytosis



Clear visualization of intracellular vesicular trafficking

It has been known that Wortmannin inhibits the recycling of endosomes or transition to lysosomes and causes enlargement of endosomes. To evaluate these changes caused by Wortmannin, early endosomes were co-stained by ECGreen and Rab5-RFP (marker protein of early endosomes), and lysosomes were co-stained by ECGreen and lysosome staining reagent. In adding Wortmannin, ECGreen was colocalized with enlarged endosomes (Rab5-RFP). On the other hand, ECGreen wasn't colocalized with lysosomes.





Description	Unit	Code
ECGreen-Endocytosis Detection	40 µl	E296-10

AcidSensor Labeling Kit – Endocytic Internalization Assay

This kit is an all-in-one kit that allows visualization of the endocytosis uptake of a target substance. The NH_2-Reactive AcidSensor (fluorescent probe) included in the kit has an intramolecular active ester group that forms a stable covalent bond when mixed with an amino group-containing target substance (protein). The AcidSensor label can be excited at 633 nm, allowing for multiple staining with green or red fluorescence (Figure 1). The AcidSensor label shows little fluorescence in neutral conditions and fluoresces when acidified in the cells where it is taken up by endocytosis (Figure 2).*Notice:

- Unlike the endocytosis detection dye: ECGreen (code: E296), this kit stains target substances that enter the cell.
- This kit can label samples with molecular weights of more than 50,000 and with reactive amino groups.

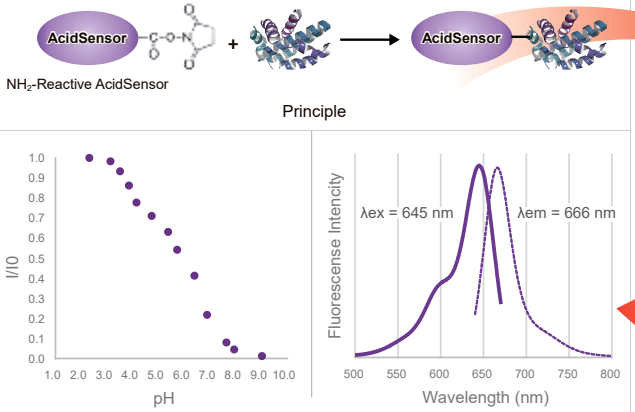


Figure 1. pH dependence and fluorescence spectrum

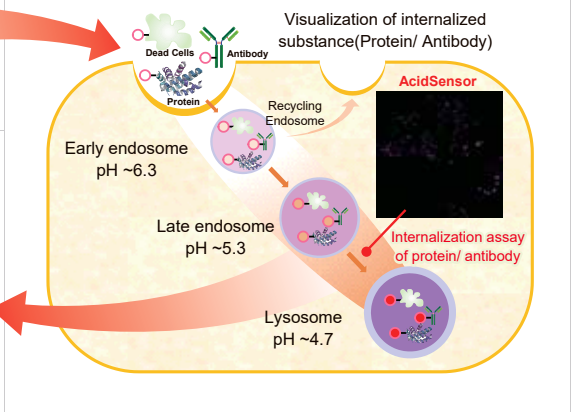
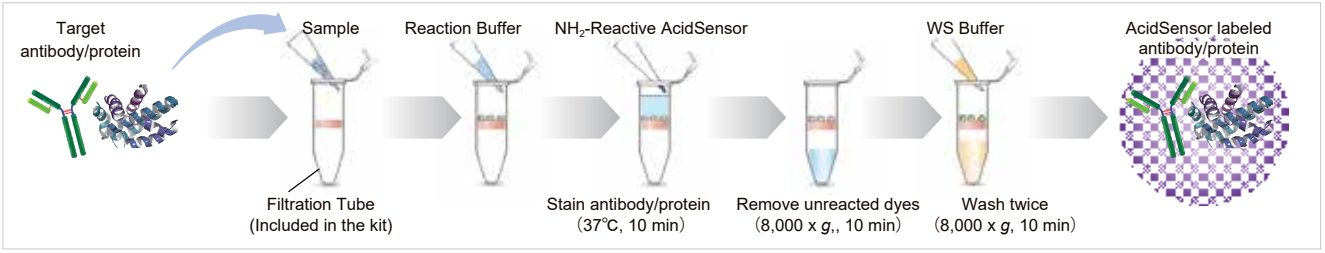


Figure 2. pH dependent fluorescence intensity of AcidSensor

This kit includes a filtration tube necessary to remove the unreacted dye, and allows you to perform everything from labeling to purification operations.* In addition, even first-time users can easily label AcidSensor by conducting experiments according to the instruction manual. * Protein/Antibody is not included.



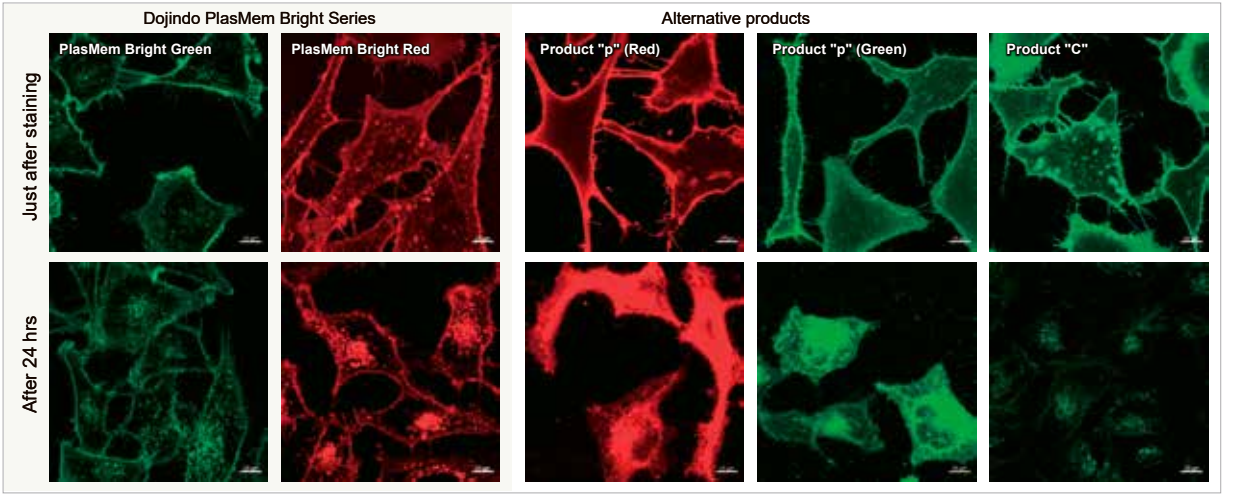
Description	Unit	Code
AcidSensor Labeling Kit – Endocytic Internalization Assay	3 samples	A558-10

Cell Membrane Staining

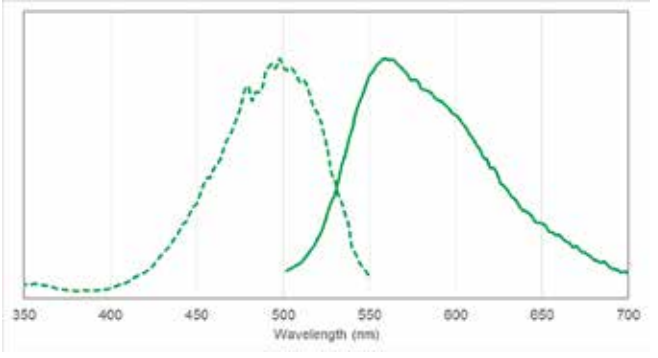
PlasMem Bright Green / Red

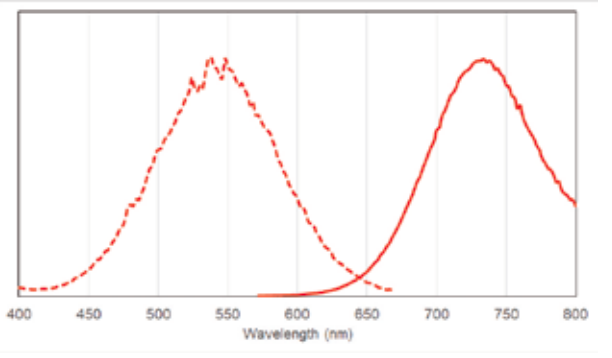
PlasMem Bright dyes overcome these limitations. PlasMem Bright dyes are designed to stain PMs for over a day. Furthermore, the PlasMem Bright dyes are more water-soluble compared with other commercially available dyes and can be diluted with culture medium. The PlasMem Bright dyes offer two different color options (green and red) and are provided as ready-to-use DMSO solutions. A working solution can be prepared easily via a single dilution step using growth medium or HBSS.

High retentivity on plasma membrane



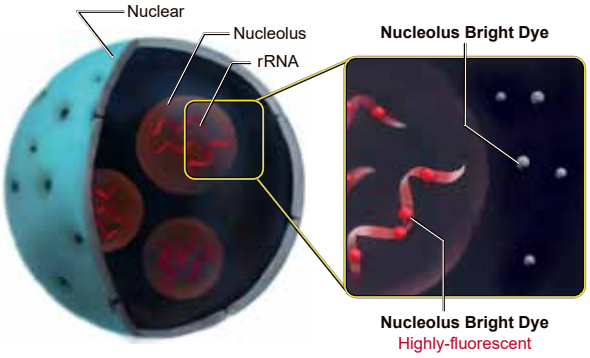
Excitation and emission spectra of PlasMem Bright dyes





Description	Unit	Code
PlasMem Bright Green	100 µl	P504-10
PlasMem Bright Red	100 µl	P505-10

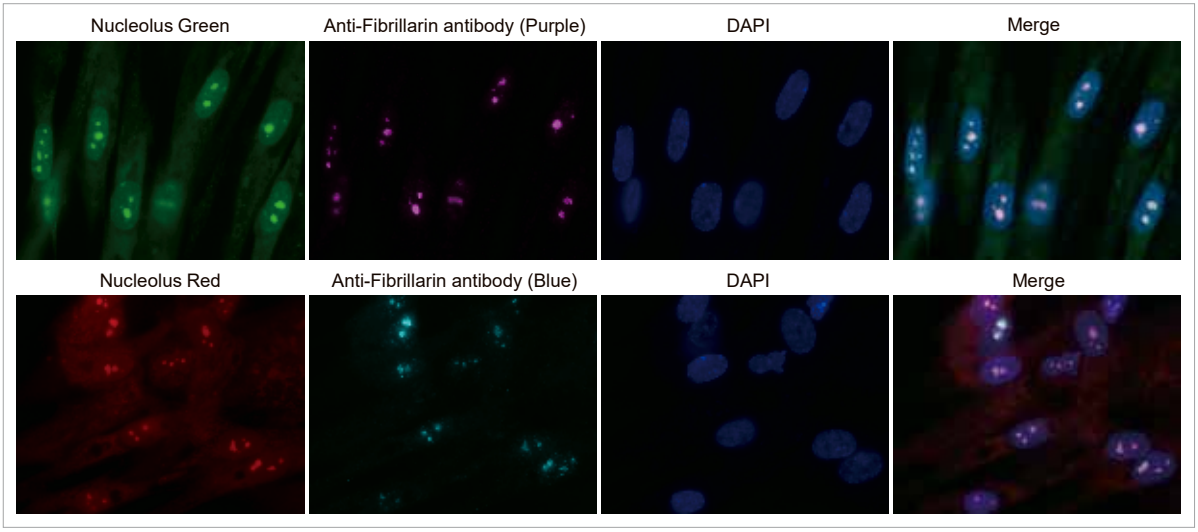
Nucleolus Bright Green / Red



Nucleolus Bright reacts to RNAs present besides nucleolus, but it shows strong fluorescence in nucleolus, which is the site of rRNA production. We recommend to co-stain with DAPI in order to image nucleolus clearly. For co-staining protocol, please refer to the Q&A tab.

	Maximum Excitation Wavelength	Maximum Emission Wavelength	Fluorescence of MeOH fixed cells	Fluorescence of PFA fixed cells
Nucleolus Bright Green	513 nm	538 nm	○	○
Nucleolus Bright Red	537 nm	605 nm	○	○

Nucleolus Localization

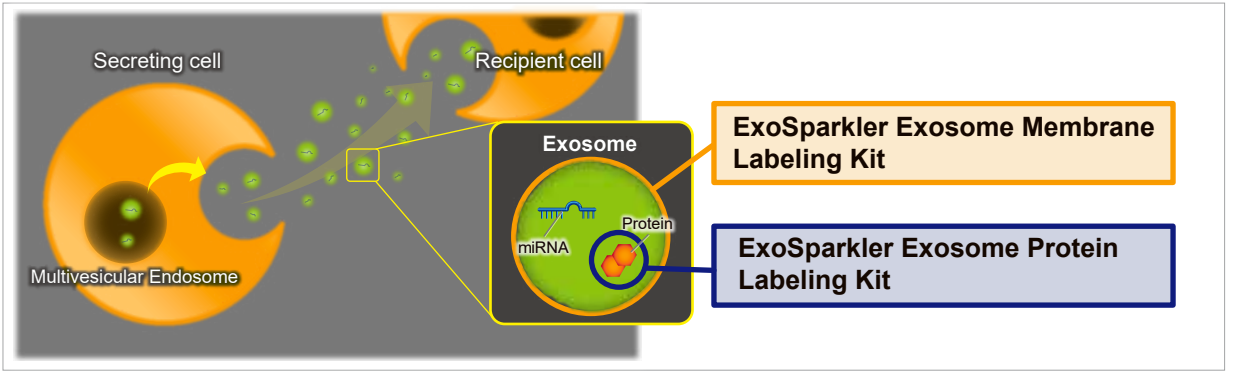


Description	Unit	Code
Nucleolus Bright Green	60 nmol	N511-10
Nucleolus Bright Red	60 nmol	N512-10

Exosome Staining

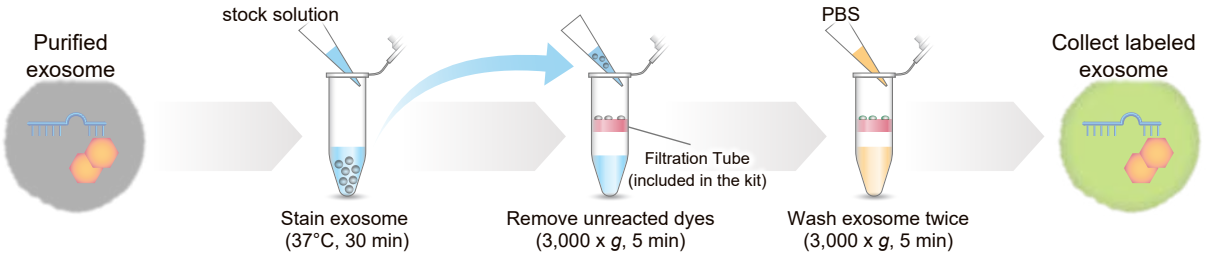
Exosome Labeling Kits

The ExoSparkler series can be used to stain purified exosomal membrane or protein and allows imaging of labeled exosomes taken up by cells.



Labelling Procedure

Staining procedure using ExoSparkler series



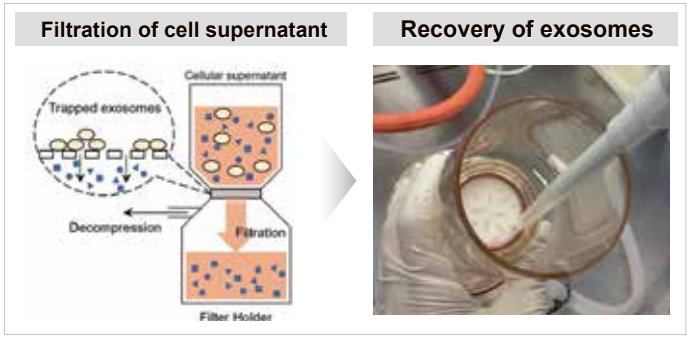
ExoSparkler series contains filtration tubes available for the removal of dyes unreacted after fluorescence labeling, as well as an optimized protocol for labeling exosomes. Our ExoSparkler series makes it possible to prepare fluorescence labeling of exosomes using the simple procedure.

Description	Unit	Code
ExoSparkler Exosome Membrane Labeling Kit-Green	5 samples	EX01-10
ExoSparkler Exosome Membrane Labeling Kit-Red	5 samples	EX02-10
ExoSparkler Exosome Membrane Labeling Kit-Deep Red	5 samples	EX03-10
Exosparkler Exosome Protein Labeling Dye-Green	5 samples	EX04-10
Exosparkler Exosome Protein Labeling Dye-Red	5 samples	EX05-10
Exosparkler Exosome Protein Labeling Dye-Deep Red	5 samples	EX06-10

Exo*Isolator* Exosome Isolation Kit

Exo*Isolator* Exosome Isolation Kit can collect exosomes from cell supernatants with a recovery rate equivalent to the ultracentrifugation(UC) method. Science Exo*Isolator* Exosome Isolation Kit requires only the filtration procedure, unlike the UC, exosomes are obtained quickly without any complicated operations.

Easy to Use no Technique Required



Exo*Isolator* Exosome Isolation Kit includes Filter Holder and Isolation Filter can collect exosomes from cell supernatant by adding PBS to the filter surface after filtration. The exosomes recovery rate is high and easy to use, no technique is required during the whole process. [Patent applied]

Recovery Rate Equivalent to Ultracentrifugation

Fig.1

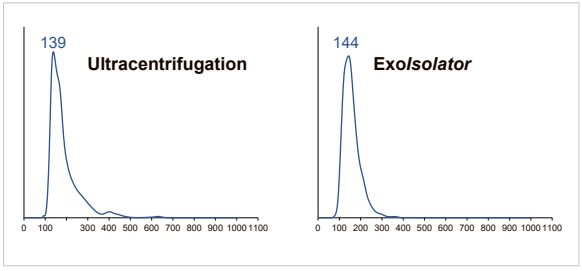


Fig.2a

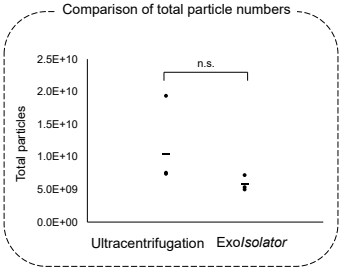
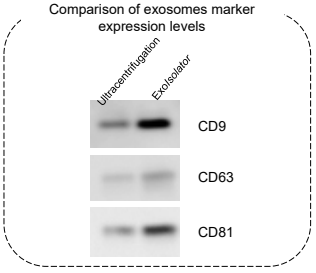


Fig.2b

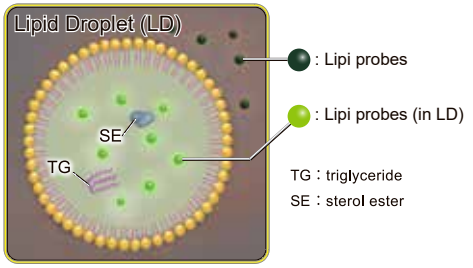


Ultracentrifugation is the most commonly used method to isolate exosomes. We isolated the exosomes from the supernatant of HEK293S using both of ultracentrifugation method and the Exo*Isolator* method. The particle size distribution (Fig. 1), the number of particles (Fig. 2(a)) and the expression level of exosome markers (Fig. 2(b)) of the isolated exosomes were tested and compared. The results showed that the Exo*Isolator* recovered exosomes with the same particle size distribution and the number of particles as the ultracentrifugation method, and the amount of exosome marker expression per protein was higher, indicating that Exo*Isolator* recovered exosomes with higher purity than the ultracentrifugation method.

Description	Unit	Code
Exo*Isolator* Exosome Isolation Kit	3 tests	EX10-10
Exo*Isolator* Isolation Filter	10 pieces	EX11-10

Lipid Droplet Staining

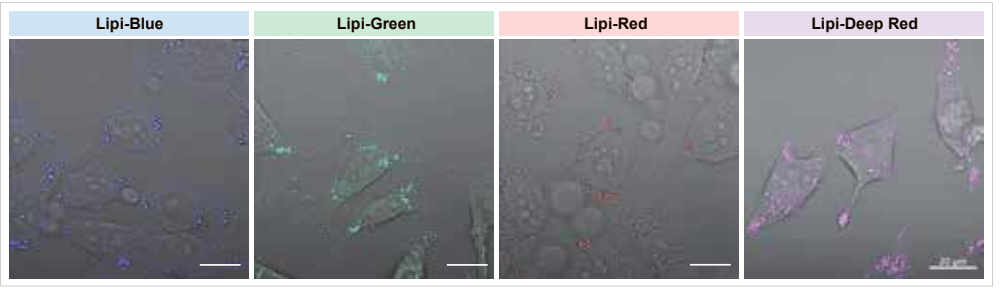
Lipi-Blue / Green / Red / Deep Red



Lipi probes are small molecules that emit strong fluorescence in a hydrophobic environment such as LDs, which can be observed without any washing steps after staining with Lipi probes.

A medium that contained oleic acid (200 µmol/l) was added and incubated overnight. Then, the supernatant was removed and the cells were washed with PBS. Each Lipi product series (1 µmol/l) was added and the cells were incubated for 15 minutes.

Lipi-Blue: Ex. 405 nm / Em. 450 – 500 nm, Lipi-Green: Ex. 488 nm / Em. 500 – 550 nm,
Lipi-Red: Ex. 561 nm / Em. 565 – 650 nm, Lipi-Deep Red: Ex.640 nm / Em.650-700 nm



Comparison of Reagents

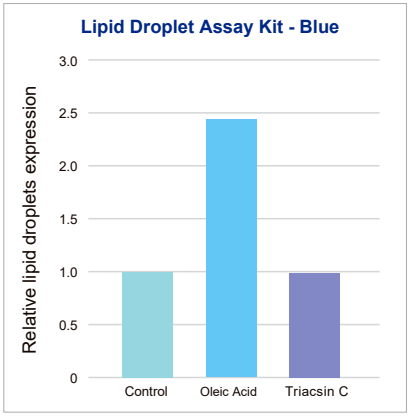
	Dojindo			Other Products		
	Lipi-Blue	Lipi-Green	Lipi-Red	Oil Red O	Nile Red	Reagent B
Live Cells	✓	✓	✓		✓	✓
Fixed Cells	✓	✓	✓	✓	✓	✓
Selectivity towards Lipid Droplet (Level of Background)	✓	✓	✓			
General Filter Accommondation*1	✓	✓	✓*2	n.d.	*3	✓
Retention in Live Cells	✓	✓		n.d.		

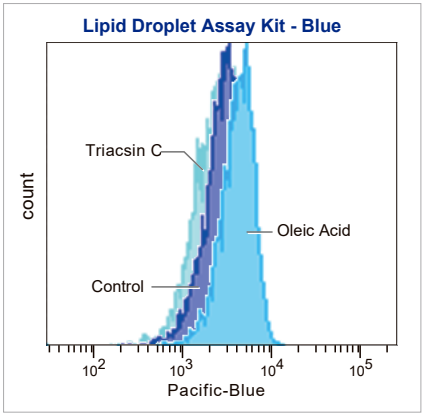
*1 Please refer to our website for the co-staining filter.
*2 When co-staining with a green fluorescent dye, a green fluorescent emission filter less than 550 nm is recommended.
*3 Leaks in GFP filter (500 ～ 540 nm)

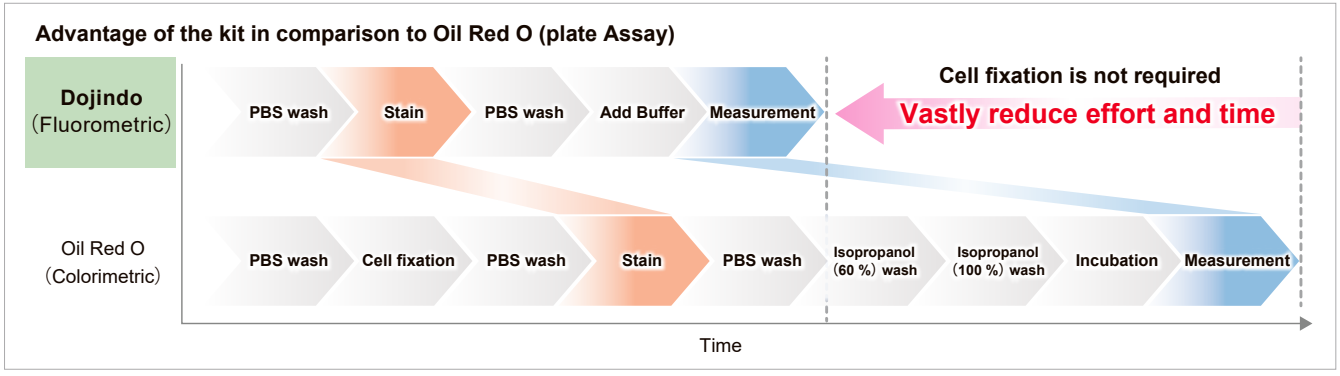
Description	Unit	Code
Lipi-Blue	10 nmol × 1	LD01-10
Lipi-Green	10 nmol × 1	LD02-10
Lipi-Red	100 nmol × 1	LD03-10
Lipi-Deep Red	10 nmol × 1	LD04-10

Lipid Droplet Assay Kit - Blue / Deep Red

The Lipid Droplet Assay Kit simplifies the quantification of fat droplets with provided protocols and buffers. It works for live cells, and its fluorescent dye is suitable for both live and fixed cells. Compared to colorimetric reagents, it reduces measuring time and increases experiment repeatability by avoiding dye deposition in the plate.







Description	Unit	Code
Lipid Droplet Assay Kit-Blue	1 set	LD05-10
Lipid Droplet Assay Kit-Deep Red	1 set	LD06-10

European Headquarters

Dojindo EU GmbH

Leopoldstr. 254, 80807, Munich, Germany

Phone: +49-89-3540-4805

email: info@dojindo.eu.com

Web: www.dojindo.com/EUROPE/