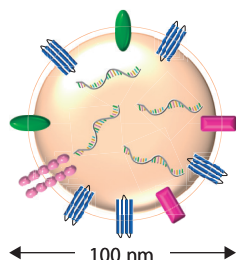


Detect extracellular vesicles such as exosomes quickly and easily

Exorapid-qIC® Immunochromatographic kit for extracellular vesicles

"Exorapid-qIC® Immunochromatographic kit for extracellular vesicles" can detect extracellular vesicles (EVs) such as exosomes, microvesicles, apoptotic bodies, and oncosomes for research. This kit was jointly developed with Shimadzu Corporation and uses "gold nanoplates" developed by Dai Nippon Toryo Co., Ltd.

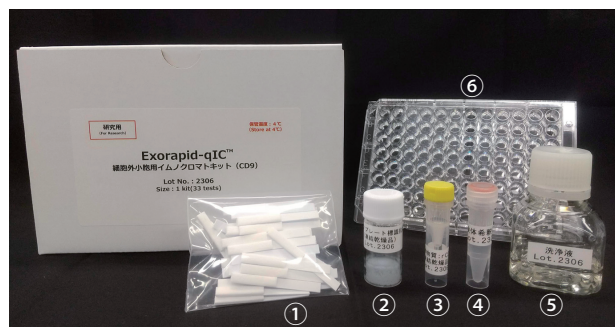


- Vesicles secreted from cells, etc.
- Contains characteristic surface proteins (Tetraspanins: CD9, CD63, CD81)

Features of EVs

1. Intercellular communication
2. Regulation of immune responses
3. Tumor progression and metastasis
4. Regenerative medicine
5. Use as biomarkers

EVs have such diverse functions. Thus, they are being applied in various research fields.



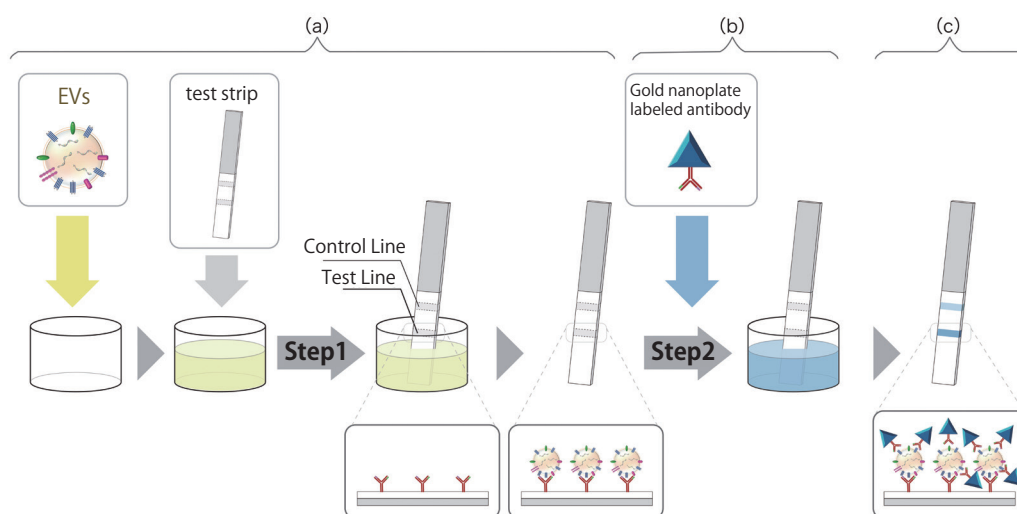
Features

- This immunochromatographic can detect extracellular vesicles (EVs).
- Blood (serum, plasma) and cell culture supernatant can be used.
- The test time is approximately 45 minutes (two-stage detection), for rapid evaluation.

Kit components

- ① Immunochromatographic test strip
- ② Gold nanoplate labeled antibody [lyophilized product]
- ③ Standard substance [lyophilized product]
- ④ Dilution solution
- ⑤ Washing solution
- ⑥ Assay microplate 96 wells

Detection mechanism



- (a) Antibodies immobilized on the test paper capture the EVs in sample.
 (b) Gold nanoplate labeled antibody binds to the EVs immobilized on the test strip in step (a).
 (c) A blue line is visually confirmed by gold nanoplate labeled antibodies bound to EVs.

Lineup

Product name	Product number	Size
Exorapid-qIC® Immunochromatographic Kit for Extracellular Vesicles (CD9)	DNT-EXO-K01	1 KIT (40test)
Exorapid-qIC® Immunochromatographic Kit for Extracellular Vesicles (CD63)	DNT-EXO-K02	1 KIT (40test)
Exorapid-qIC® Immunochromatographic Kit for Extracellular Vesicles (CD81)	DNT-EXO-K03	1 KIT (40test)
Exorapid-qIC® Immunochromatographic Kit for Extracellular Vesicles (CD9, CD63,CD81 set)	DNT-EXO-K123	1 KIT (6test×3)



COSMO BIO USA

Comparison with conventional analysis methods (According to our survey)

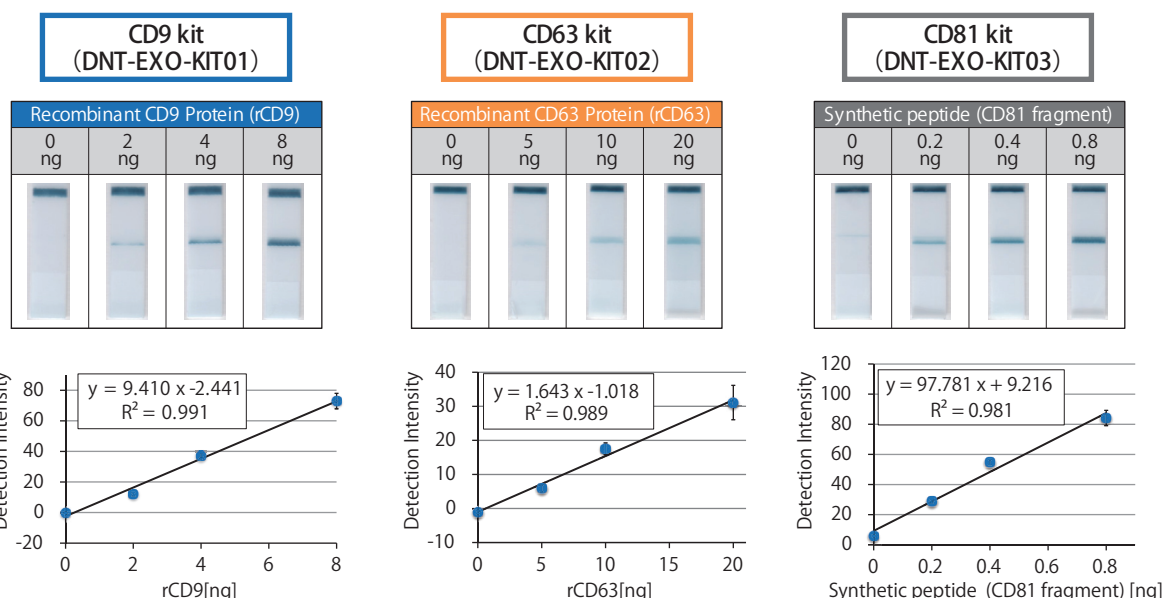
	IC (Exorapid-qIC [®])	ELISA	WB	NTA	FCM
Test time	Short (0.75 hours)	Short to mid. (3 hours)	Long (5-8 hours)	Short (1 hour)	Short (1 hour)
Operability	Very good	Good	Neutral	Good	Good
Throughput	High	Very high	High	Neutral	Neutral
Detection sensitivity	High	High to very high	High	Very high	Very high
Accuracy	Neutral	High	High	Neutral	Very high
Initial cost	Very low	Low	Low	High	High
Analyzer	Unnecessary	Necessary	Necessary	Necessary	Necessary

ELISA : Enzyme-linked immunosorbent assay
NTA : Nanoparticle Tracking Analysis

WB : Western blotting
FCM : Flow cytometry

Application Data

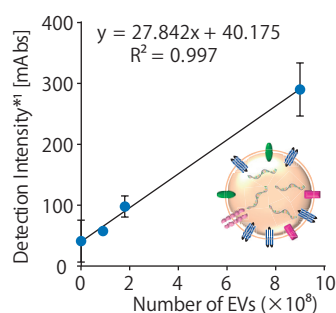
Calibration curve



It is possible to create a calibration curve using the standard substance included with each kit.
(The intensity of the test line is analyzed and graphed using Image.)

Quantitative analysis of EVs

EVs calibration curve (CD9 kit)

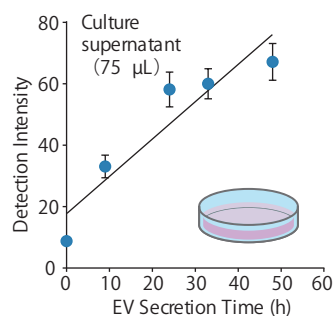


High linearity in EV count and detection intensity was obtained

*1: Measured with lateral flow reader
(C10066-10, HAMAMATSU PHOTONICS K.K.)

Monitoring of EVs amount

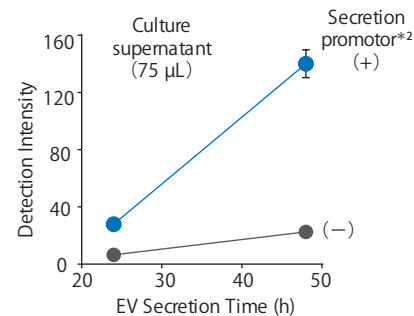
(CD9 kit)



The amount of EVs can be monitored by direct testing of the culture supernatant (cell line: MCF7)

Confirmation of Drug-induced promotion of EVs secretion

(CD9 kit)

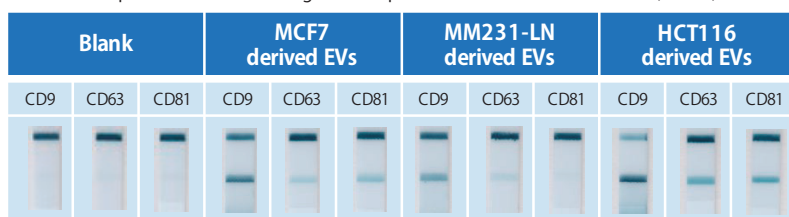


Simple monitoring of EVs secretion from cells is possible (cell line: HCT116)

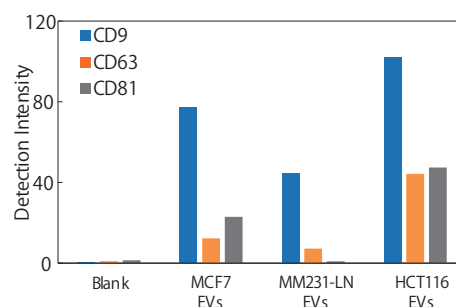
*2: Cucurbitacin B

Evaluation with cancer cell-derived exosomes

*Uses test strips for the CD9 kit and gold nanoplate-labeled antibodies for CD9, CD63, and CD81.



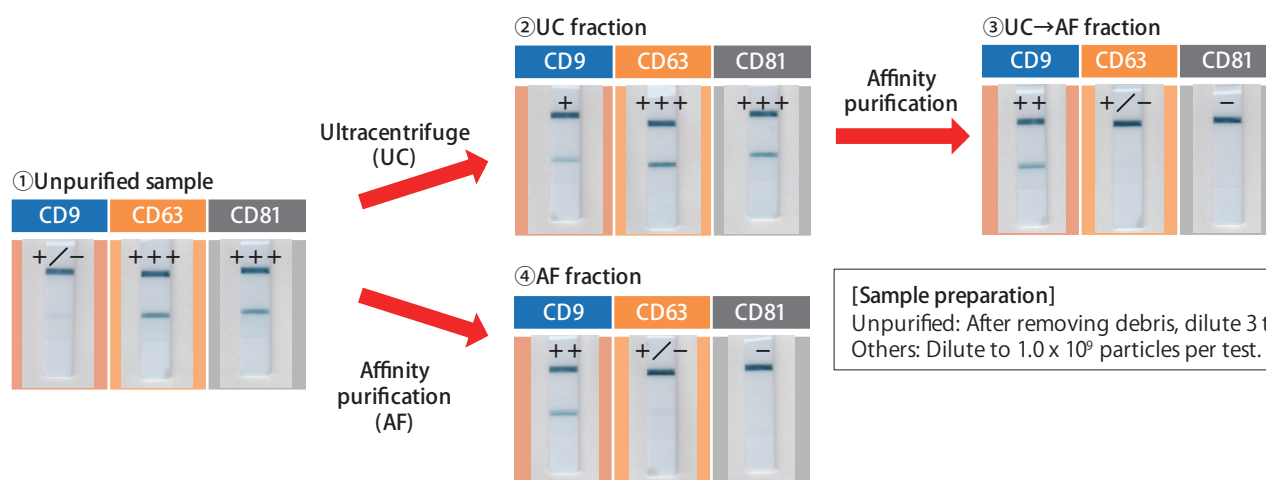
The intensity of detection of CD9, CD63, and CD81 differed among cell types, revealing their characteristics



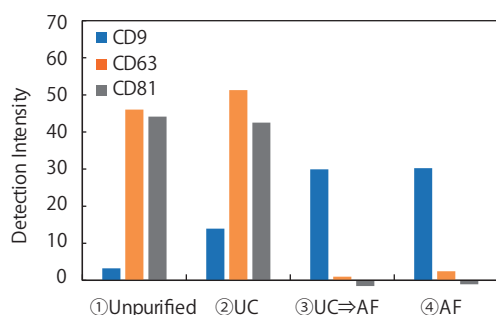
Our kits can be used in combination with CD9, CD63, and CD81 kits depending on the test system.

Evaluation with serum

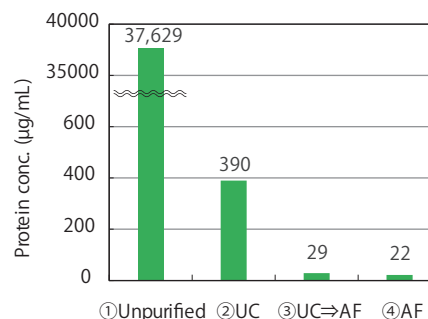
①Unpurified sample, ②Ultracentrifuged fraction, ③Affinity-purified fraction after ultracentrifugation, and ④Direct affinity-purified fraction of serum (pooled product) were evaluated using each kits.



Immunochromatographic analysis



Protein quantification (BCA assay)



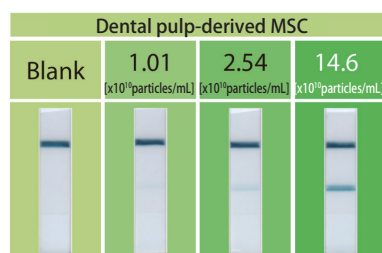
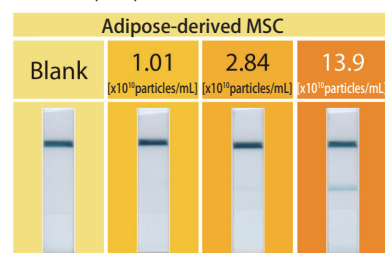
AF reduced the concentration of contaminating proteins, improving the detection intensity of CD9 and decreasing the detection intensity of CD63 and CD81. Although there will be differences in EVs recovered with each purification method, nonspecific detection is expected to be reduced with purification methods that reduce protein concentration

Evaluation with mesenchymal stem cell (MSC)-derived exosomes

① Relationship between particle concentration and detection intensity

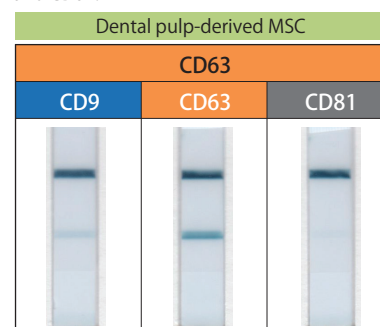
We evaluated the cell culture supernatants of adipose-derived and dental pulp-derived MSCs. Hence the EVs concentration was low, they were concentrated by ultrafiltration.

Use Exorapid-qIC[®] (CD63)

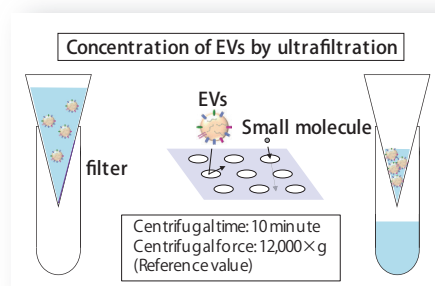
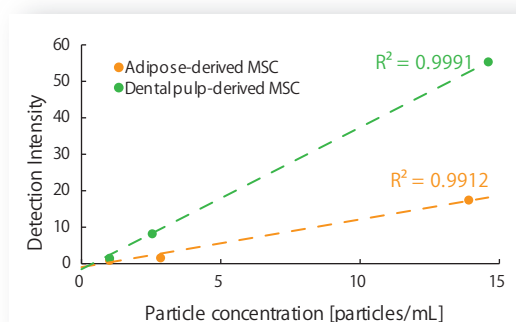


*Culture supernatant was concentrated by ultrafiltration and 40 μ L was used for evaluation

Uses test strips for the CD63 kit and gold nanoplate-labeled antibodies for CD9, CD63, and CD81.



*Particle concentration: 14.6×10^{10} particles/mL









- Using a CD63 kit, EVs in the culture supernatant of adipose-derived and dental pulp-derived MSCs were detected in a **concentration-dependent manner**.
- EVs containing both **markers (CD9 and CD63)** were detected in the culture supernatant of dental pulp-derived MSCs.

② Combination with purification kits

EVs in the culture supernatant of adipose-derived MSCs were concentrated and purified using a commercially available kit and detected by Exorapid-qIC[®].

Purification method	Required time [hour]	Cost	Particle concentration [$\times 10^{10}$ particles/mL]	Recovered volume (Per 1mL of stock sample) [μ L]	Recovered particle (Per 1mL of stock sample) [$\times 10^{10}$ particles]	Protein concentration (μ g/mL)
① Unpurified	—	—	2.5	1,000	2.5	727
② Affinity purification	2~3	High	6.9	50	0.35	ND
③ Ultrafiltration + EV-Capture	1.5	Low	8.7	60	0.52	39

①Unpurified		②Affinity purification		③Ultrafiltration + EV-Capture	
CD9	CD63	CD9	CD63	CD9	CD63
—	—	+/-	+/-	++	+
					

EV-Capture[™] is a product that utilizes the negative charge of EVs to separate and collect them using a spin column

- Even samples with low concentrations of EVs can be detected in a short time in **combination with concentration and purification methods**.
- Combination with ultrafiltration and EV-Capture[™] enables **"quick," "easy," and "inexpensive"** purification and concentration.



EV-Capture[™]

Product name	No.	Size
EV-Capture [™] EV Purification Spin Column Kit	EVP01-010	1 kit (10 prep)

Contact address If you have any questions, please feel free to contact us at the address below !

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