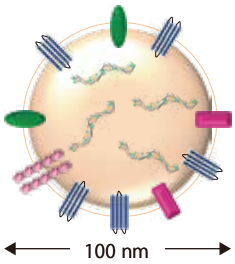
Detect extracellular vesicles such as exosomes quickly and easily

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 a biomarkers

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

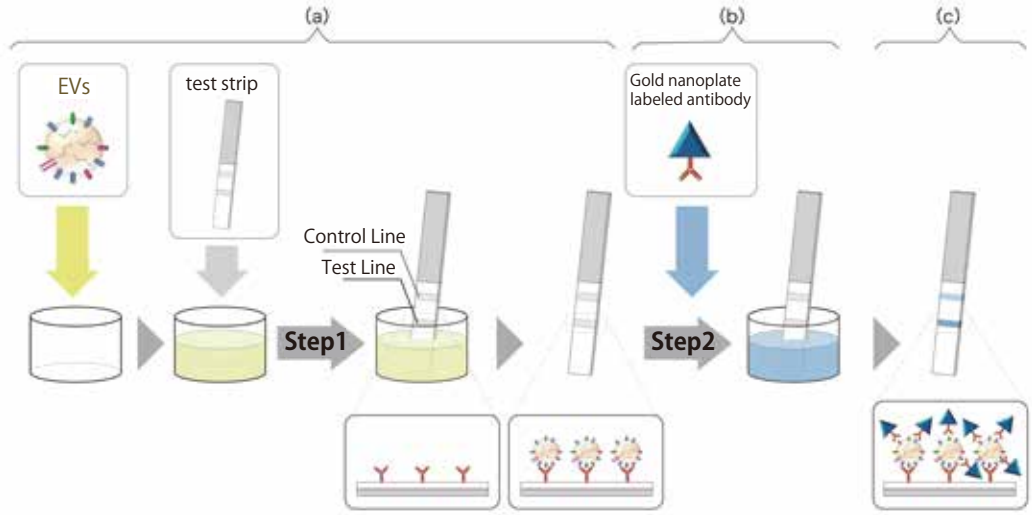
Features

- 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 lines 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)
	DNT-EXO-K01-T	1 KIT (12test)
Exorapid-qIC Immunochromatographic Kit for Extracellular Vesicles (CD63)	DNT-EXO-K02	1 KIT (40test)
	DNT-EXO-K02-T	1 KIT (12test)

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Comparison with conventional analysis methods (According to our survey)

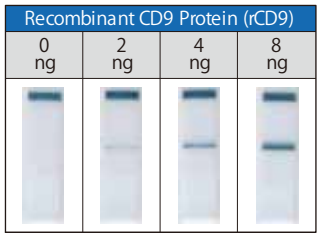
	IC (Exorapid-qIC)	ELISA	WB	NTA	FCM
Test time	Short (0.75 hours)	Short to mid. (3 hours)	Long (5-8 hours)	Short (1 hour)	Short (1 hour)
Operability	Very good	Good	Neutral	Good	Good
Throughput	High	Very high	High	Neutral	Neutral
Detection sensitivity	High	High to very high	High	Very high	Very high
Accuracy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

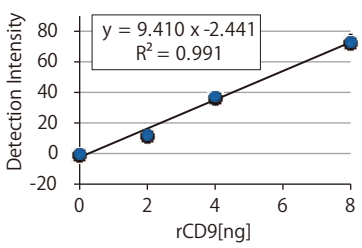
ELISA : Enzyme-linked immunosorbent assay　　WB : Western blotting
NTA : Nanoparticle Tracking Analysis　　FCM : Flow cytometry

Application Data

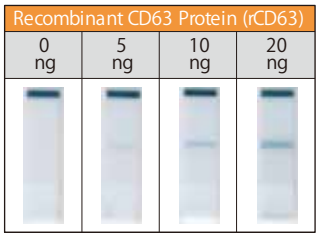
Calibration curve

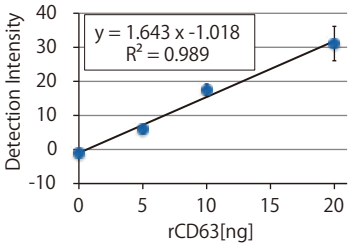
CD9 kit (DNT-EXO-KIT01)





CD63 kit (DNT-EXO-KIT02)

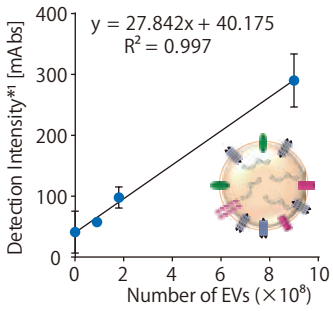




It is possible to create a calibration curve using the standard substance included with each kit.
(The intensity of the test line is analyzed and graphed using Image.)

Quantitative analysis of EVs

EVs calibration curve (CD9 kit)

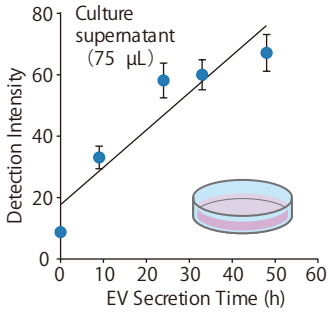


High linearity in EV count and detection intensity was obtained

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

Monitoring of EVs amount

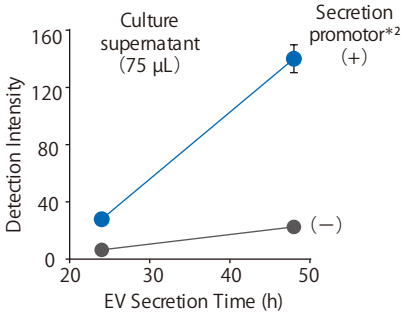
(CD9 kit)



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

Confirmation of Drug-induced promotion of EVs secretio

(CD9 kit)

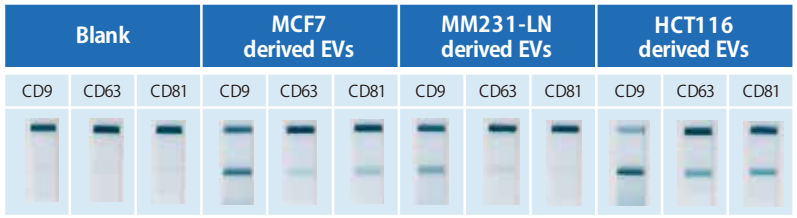


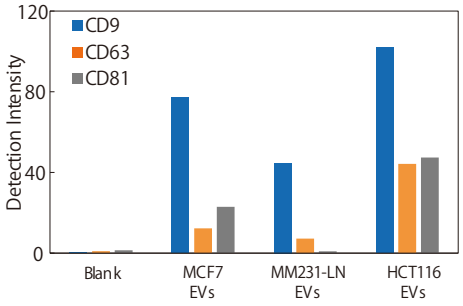
Simple monitoring of EVs secretion from cells is possible (cell line: HCT116)

*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.



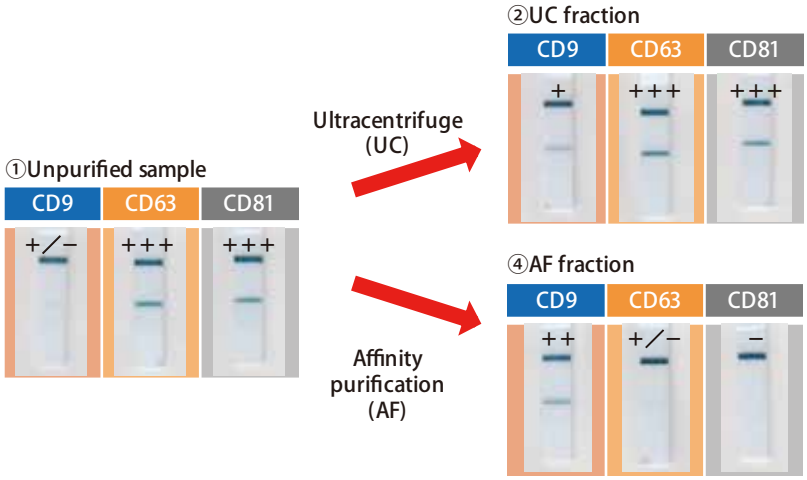


The intensity of detection of CD9, CD63, and CD81 differed among cell types, revealing their characteristics

Test strips and gold nanoplate-labeled antibodies of each kits can be combined to suit the test system.

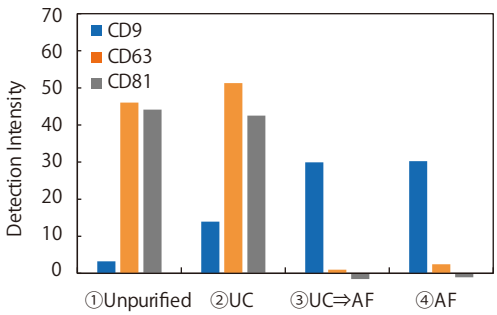
Evaluation with serum

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

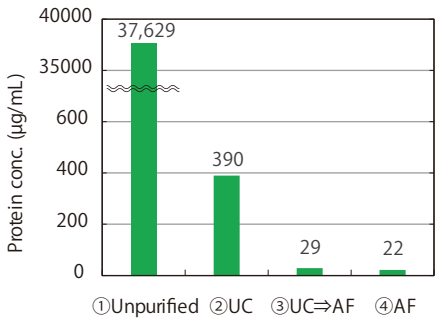


[Sample preparation]
Unpurified: After removing debris, dilute 3 times
Others: Dilute to 1.0 x 10^9 particles per test.

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

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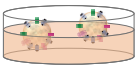
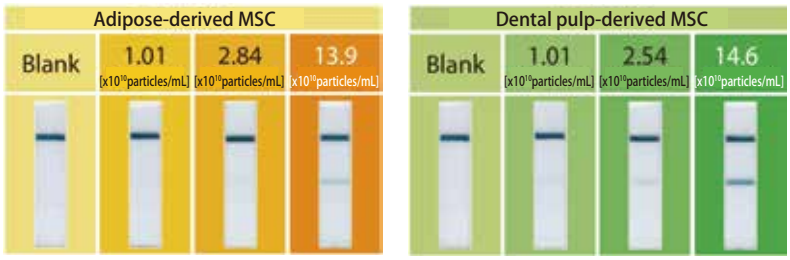
Evaluation with mesenchymal stem cell (MSC)-derived exosomes

①Relationship 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.

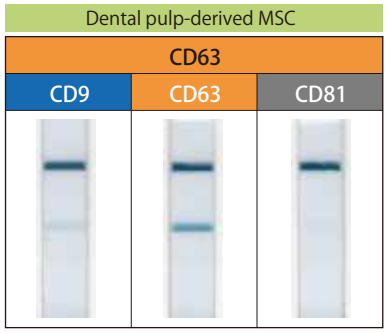


Use Exorapid-qIC (CD63)

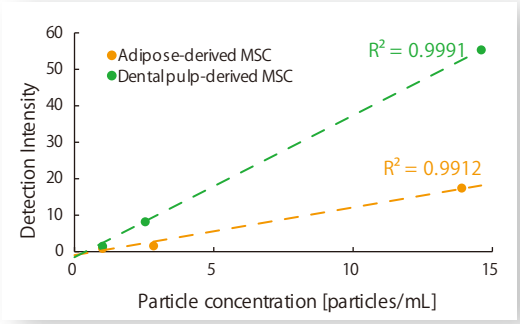


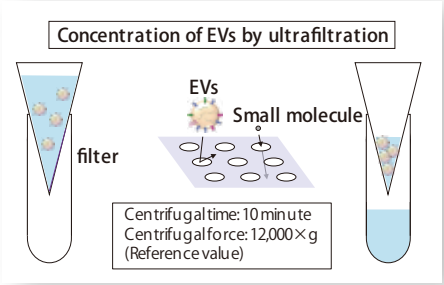
*Culture supernatant was concentrated by ultrafiltration and 40 µL was used for evaluation

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



*Particle concentration: 14.6×10¹⁰particles/mL



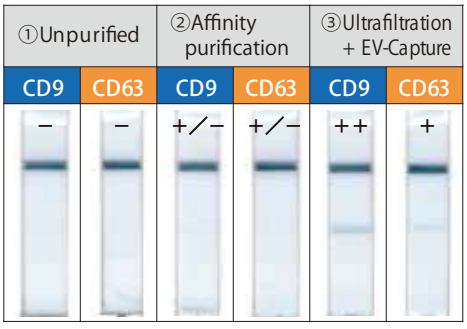


- Using a CD63 kit, EVs in the culture supernatant of adipose-derived and dental pulp-derived MSCs were detected in a concentration-dependent manner.
- EVs containing both markers (CD9 and CD63) were detected in the culture supernatant of dental pulp-derived MSCs.

②Combination 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	Requierd time [hour]	Cost	Particle concentration [x10¹⁰particles/mL]	Recovered volume (Per 1mL of stock sample) [µL]	Recovered particle (Per 1mL of stock sample) [x10¹⁰particles]	Protein concentration (µg/mL)
①Unpurified	—	—	2.5	1,000	2.5	727
②Affinity purification	2～3	High	6.9	50	0.35	ND
③Ultrafiltration + EV-Capture	1.5	Low	8.7	60	0.52	39



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 !

About Sales information

COSMO BIO USA

2792 Loker Ave West, Suite 101 Carlsbad, CA 9200, USA
URL: www.cosmobiousa.com
Phone/FAX:(+1)760-431-4600
Email: support@cosmobiousa.com

About technical information

DNT DAI NIPPON TORYO CO.,LTD.

Minami-Senba 1CHOME, Chuo-ku, Osakashi,
Osaka, 542-0081, Japan
URL: https://www.dnt.co.jp
Email: dnt10023120@star.dnt.co.jp

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