

Isolation and Analysis of Traceable and Functionalized Extracellular Vesicles from the Plasma and Solid Tissues

Yuan Cao^{*1,2}, Ji-Yu Qiu^{*1,3}, Da Chen^{1,2}, Chen-Yao Li^{1,4}, Shu-Juan Xing^{1,5}, Chen-Xi Zheng¹, Xin Liu², Yan Jin^{1,6}, Bing-Dong Sui¹

¹State Key Laboratory of Military Stomatology & National Clinical Research Center for Oral Diseases & Shaanxi International Joint Research Center for Oral Diseases, Center for Tissue Engineering, School of Stomatology, The Fourth Military Medical University ²Department of Orthodontics, School of Stomatology, The Fourth Military Medical University ³Department of VIP Dental Care, School of Stomatology, The Fourth Military Medical University

⁴School of Basic Medicine, The Fourth Military Medical University ⁵College of Life Science, Northwest University ⁶Xi'an Institute of Tissue Engineering and Regenerative Medicine

* These authors contributed equally

Corresponding Authors

Xin Liu
986922403@qq.com

Yan Jin
yanjin@fmmu.edu.cn

Bing-Dong Sui
bingdong@fmmu.edu.cn

Citation

Cao, Y., Qiu, J.Y., Chen, D., Li, C.Y., Xing, S.J., Zheng, C.X., Liu, X., Jin, Y., Sui, B.D. Isolation and Analysis of Traceable and Functionalized Extracellular Vesicles from the Plasma and Solid Tissues. *J. Vis. Exp.* (188), e63990, doi:10.3791/63990 (2022).

Date Published

October 17, 2022

DOI

10.3791/63990

URL

jove.com/video/63990

Materials

Name	Company	Catalog Number	Comments
4% paraformaldehyde	Biosharp	143174	Transmission electron microscope
Alexa fluor 488 anti-goat secondary antibody	Yeason	34306ES60	Flow cytometry
Alexa fluor 488 anti-rabbit secondary antibody	Invitrogen	A11008	Flow cytometry
Anti-CD18 antibody	Abcam	ab131044	Flow cytometry
Anti-CD81 antibody	Abcam	ab109201	Western blot
anti-CD9 antibody	Huabio	ET1601-9	Western blot
Anti-Mitofilin antibody	Abcam	ab110329	Western blot
APOA1 Rabbit pAb	Abclone	A14211	Western blot
BCA protein assay kit	TIANGEN	PA115	Western blot
BLUeye Prestained Protein Ladder	Sigma-Aldrich	94964-500UL	Western blot
Bovine serum albumin	MP Biomedical	218072801	Western blot
Caveolin-1 antibody	Santa Cruz Biotechnology	sc-53564	Western blot
CellMask Orange plasma membrane stain	Invitrogen	C10045	Flow cytometry
Chemiluminescence	Amersham Biosciences	N/A	Western blot
Curved operating scissor	JZ Surgical Instrument	J21040	EV isolation
Electronic balance	Zhi Ke	ZK-DST	EV isolation
Epoch spectrophotometer	BioTek	N/A	Western blot
Eppendorf tubes	Eppendorf	3810X	EV isolation
Flotillin-1 antibody	PTM BIO	PTM-5369	Western blot
Gel imaging system	Tanon	4600	Western blot
Golgin84	Novus	nbp1-83352	Western blot

Grids - Formvar/Carbon Coated - Copper 200 mesh	Polysciences	24915	Transmission electron microscope
Heparin Solution	StemCell	7980	EV isolation
Liberase Research Grade	Sigma-Aldrich	5401127001	EV isolation
Microscopic tweezer	JZ Surgical Instrument	JD1020	EV isolation
NovoCyte flow cytometer	ACEA	N/A	Flow cytometry
Omni-PAGE Hepes-Tris Gels Hepes 4~20%, 10 wells	Epizyme	LK206	Western blot
OSCAR(D-19) antibody	Santa Cruz Biotechnology	SC-34235	Flow cytometry
PBS (2x)	ZHHC	PW013	Western blot
Pentobarbital sodium	Sigma-Aldrich	57-33-0	Anesthetization
Peroxidase AffiniPure Goat Anti-Mouse IgG (H+L)	Jacson	115-035-003	Western blot
Peroxidase AffiniPure Goat Anti-Rabbit IgG (H+L)	Jacson	111-035-003	Western blot
Phosphotungstic acid	RHAWN	12501-23-4	Transmission electron microscope
PKM2(d78a4) xp rabbit mab	Cell Signaling	4053t	Western blot
Polyethylene (PE) film	Xiang yi	200150055	Transmission electron microscope
Polyvinylidene fluoride membranes	Roche	3010040001	Western blot
Protease inhibitors	Roche	4693132001	Western blot
Recombinant anti-PGD antibody	Abcam	ab129199	Western blot
RIPA lysis buffer	Beyotime	P0013	Western blot
SDS-PAGE loading buffer (5x)	Cwbio	CW0027S	Western blot
Size beads	Invitrogen	F13839	Flow cytometry
Tabletop High-Speed Micro Centrifuges	Hitachi	CT15E	EV isolation
Transmission electron microscope	HITACHI	H-7650	Transmission electron microscope
Tween-20	MP Biomedicals	19472	Western blot
Vortex Mixer Genie	Scientific Industries	SI0425	EV isolation
ZetaView BASIC NTA - Nanoparticle Tracking Video Microscope PMX-120	Particle Metrix	N/A	Nanoparticle tracking analysis
α -Actinin-4 Rabbit mAb	Abclone	A3379	Western blot
β -actin	Cwbio	CW0096M	Western blot