

Efficient exosome subpopulations isolation and proteomic profiling by Sub-ExoProfile chip towards cancer diagnosis and treatment

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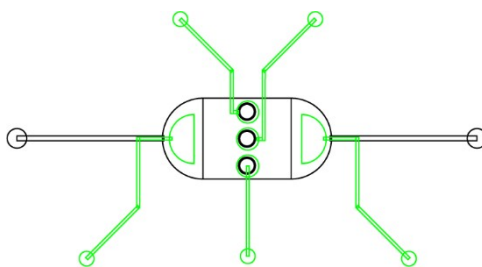


Fig S1. Design of the Sub-ExoProfile chip.

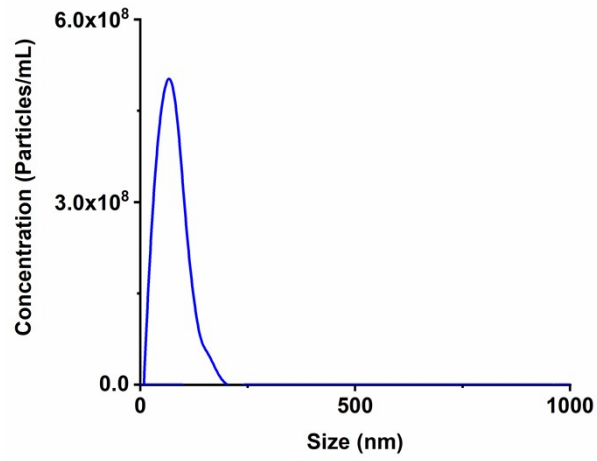


Fig. S2 NTA measurements of exosomes isolated from MCF-7 breast cancer cell culture medium by classical ultracentrifugation.

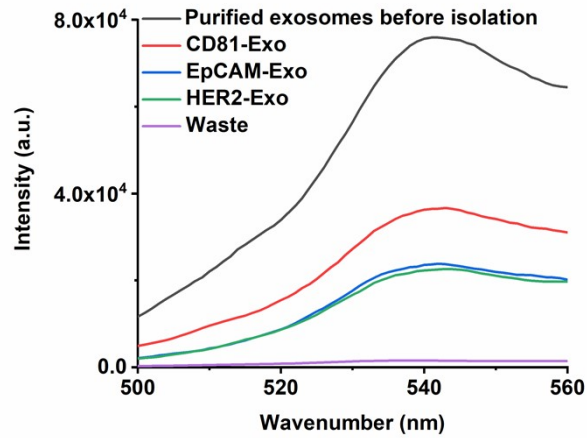


Fig. S3 Fluorescence change of the DiO-stained exosomes before and after isolation by the Exo-SubProfile chip.

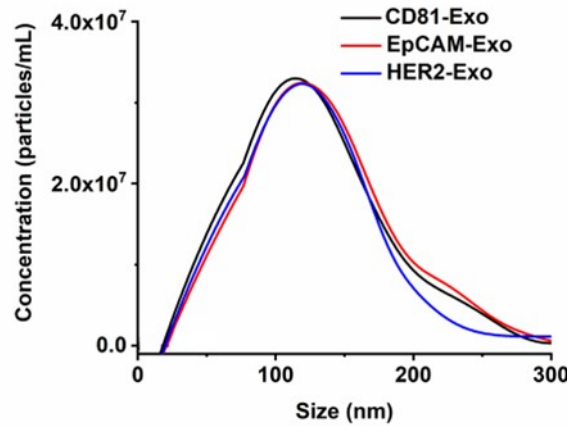


Fig. S4 Size distribution of the three CD81-Exo, EpCAM-Exo, and HER2-Exo captured by the Exo-SubProfile chip.

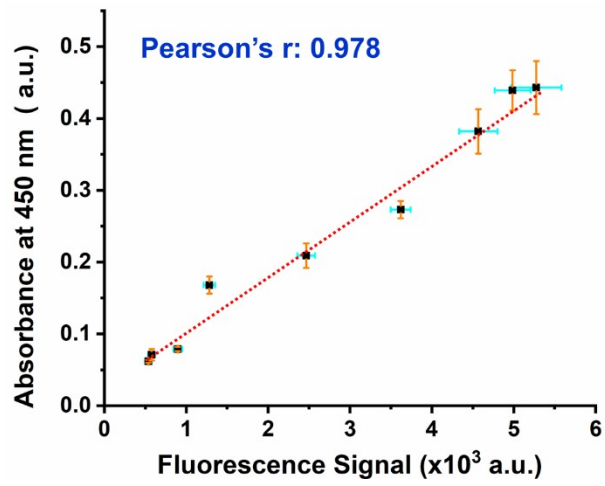


Fig. S5 Correlations between microplate ELISA and on-chip analysis of CD81, EpCAM and HER2 positive exosomes from three cell lines.

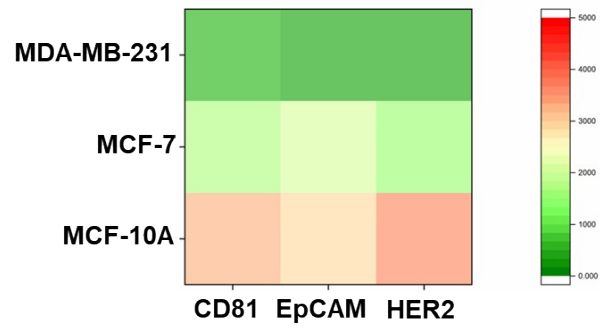


Fig. S6. Profiling of CD81, EpCAM, HER2 specific exosome subpopulations from MDA-MB-231, MCF-7, MCF-10A cell lines by flat glass chip.

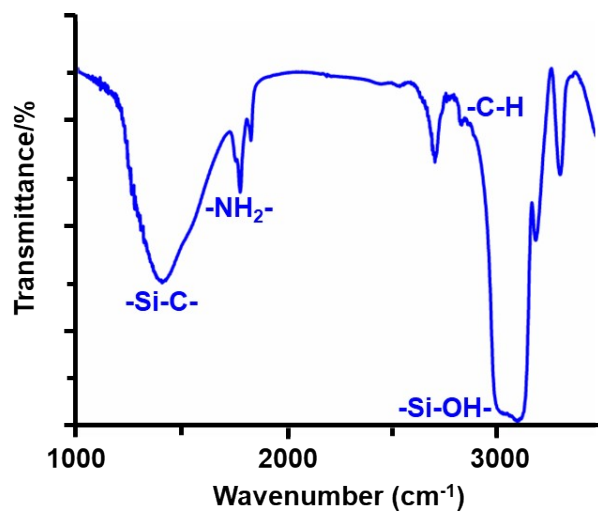


Fig. S7 FT-IR of the self-assembled amphiphilic mesoporous SiNPs on the nanopillars of Exo-SubProfile chip.

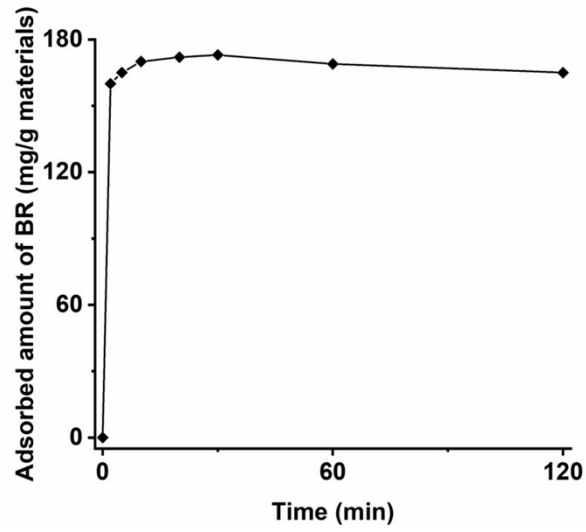


Fig. S8 Adsorption of model membrane protein bacteriorhodopsin into the self-assembled amphiphilic mesoporous SiNPs on the nanopillars of Exo-SubProfile chip as a function of incubation time.

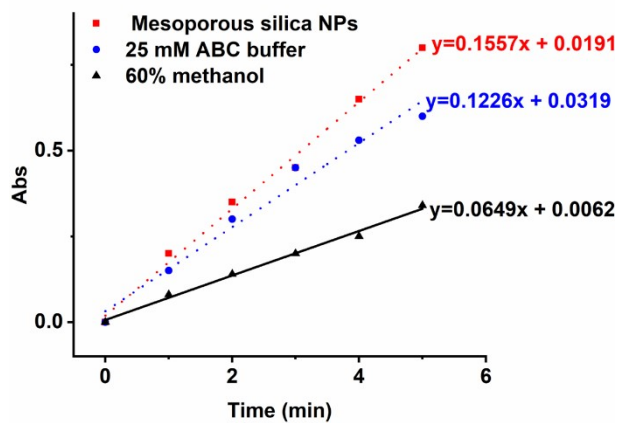


Fig. S9 Trypsin activity in different system. The substrate involved in the system was BAEE with an initial concentration of 0.8 mg/mL. The absorbance (Abs) corresponds to BA at 253 nm, which was generated from BAEE by trypsin-catalyzed hydrolysis. The concentration of enzyme was 0.5 mg/mL.

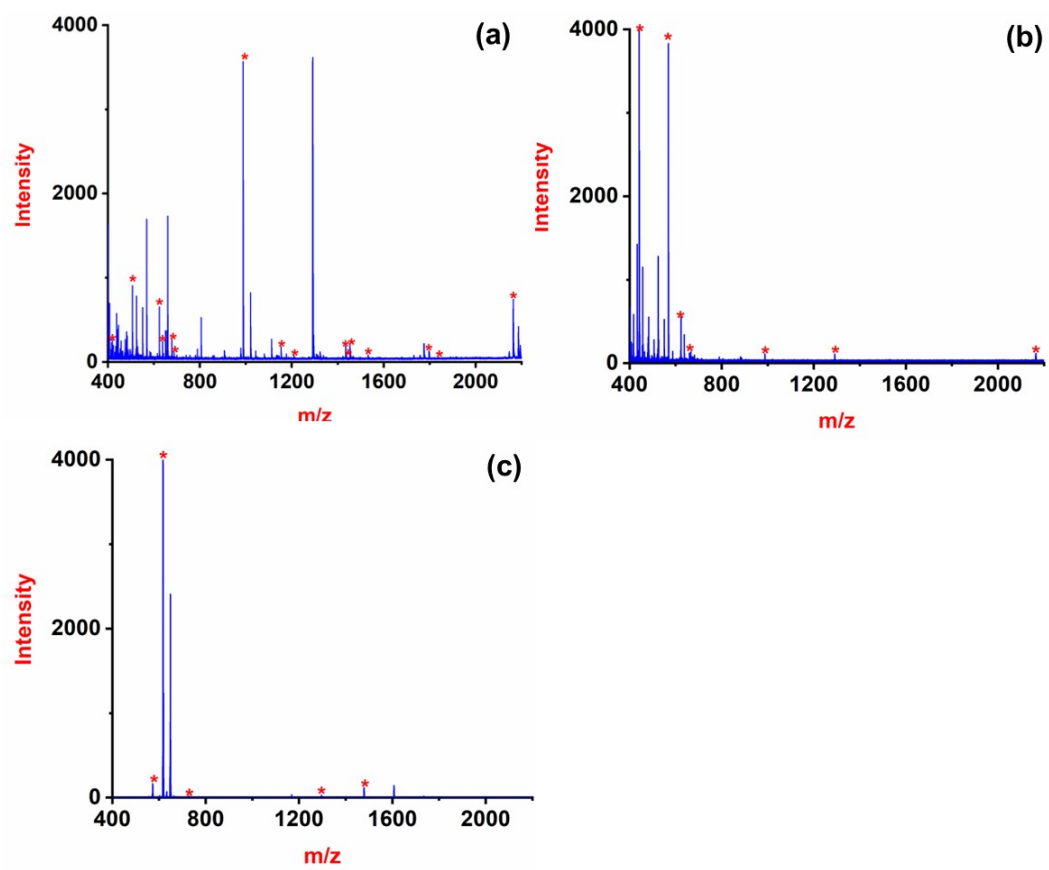


Fig. S10 Mass spectra of proteolysis products from (a) 10 min self-assembled mesoporous SiNPs on the Exo-SubProfile chip-assisted BR (100 ng/μL) digestion, (b) 10 min nonporous SiNPs-assisted BR (100 ng/μL) digestion, and (c) 10 min traditional in-solution BR (100 ng/μL) digestion

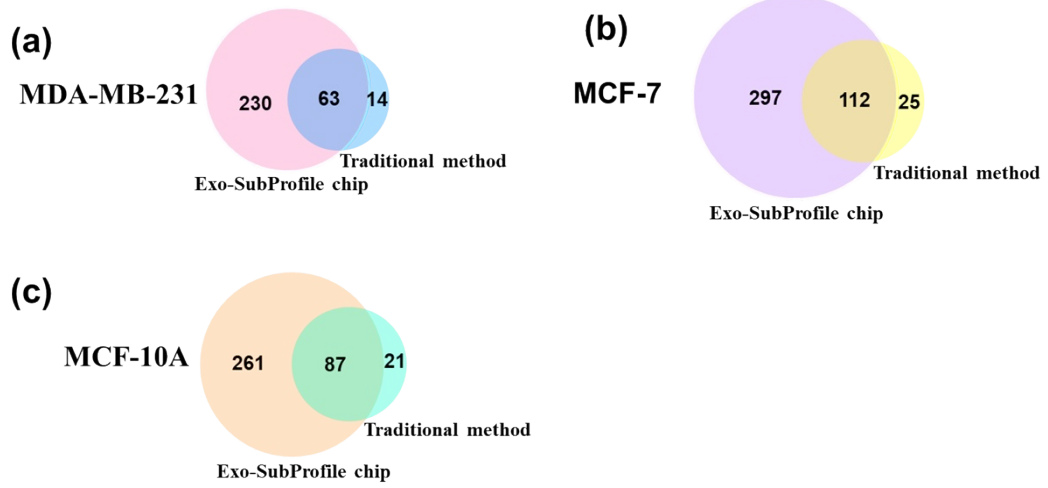


Fig. S11 Venn diagram of identified total proteins from the three CD81-Exo, EpCAM-Exo, and HER2-Exo of (a) MDA-MB-231, (b) MCF-7, and (c) MCF-10A by both the Exo-SubProfile chip and traditional method.

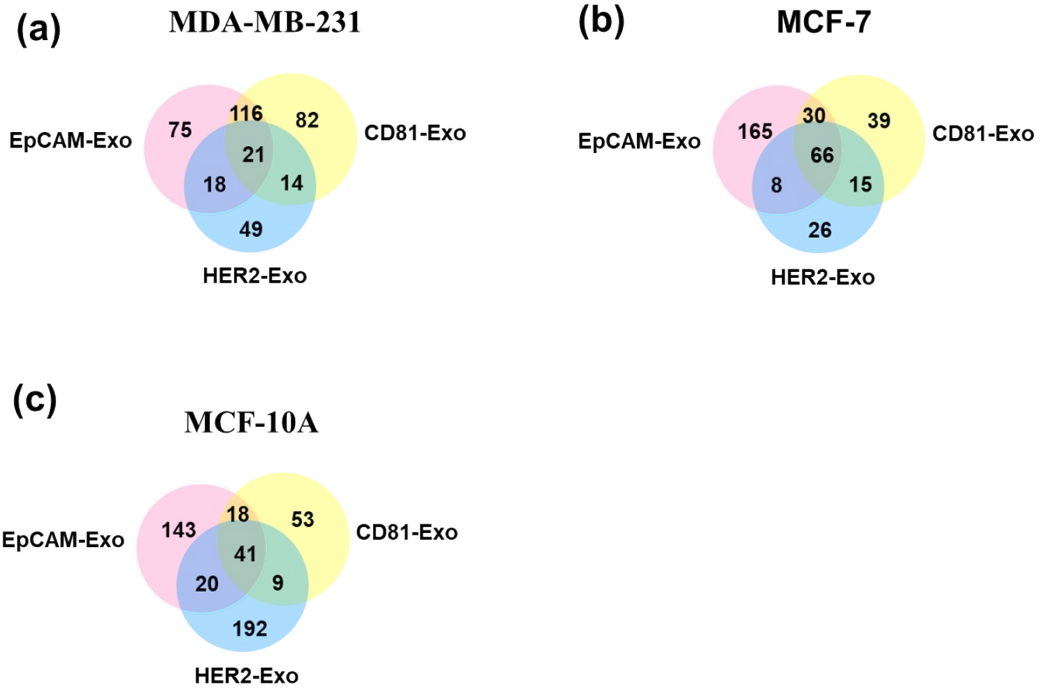


Fig. S12 Venn diagram of identified proteins from the three CD81-Exo EpCAM-Exo, and HER2-Exo of (a) MDA-MB-231, (b) MCF-7, and (c) MCF-10A by the Exo-SubProfile chip.

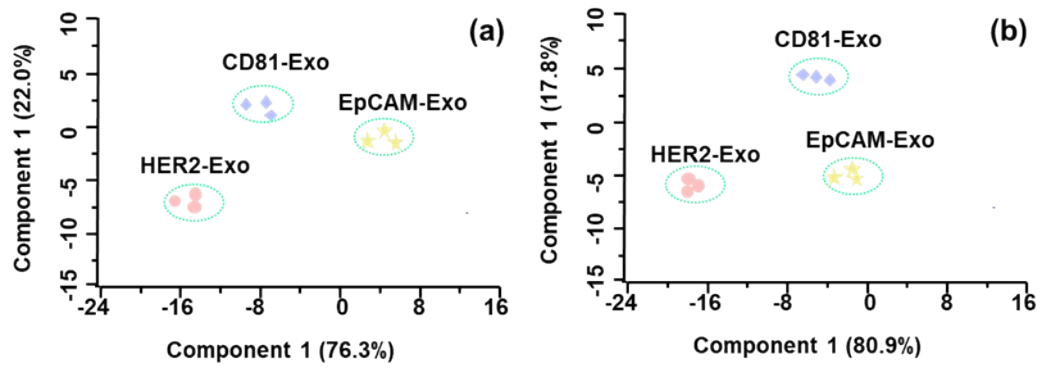


Fig. S13 Quantitative principal analysis (PCA) shows that the Exo-SubProfile chip enabled the three CD81-Exo, EpCAM-Exo, and HER2-Exo from MCF-7 and MCF-10A are well separated.

Table S1. The detailed information about the four categories of identified proteins from different exosomes subpopulations in different type of cancer patients.

Category	Uniprot	Gene	HER2 positive BC			TNBC			Description
			CD81-Exo	EpCAM-Exo	HER2-Exo	CD81-Exo	EpCAM-Exo	HER2-Exo	
Tyrosine kinases	P06213	INSR	1	0	0	5	1	0	Isoform Short of Insulin receptor
	P04626	ERBB2	0	18	0	0	0	0	Receptor tyrosine-protein kinase erbB-2
	P00533	EGFR	2	0	0	3	0	0	Epidermal growth factor receptor
	P30530	AXL	0	0	5	0	0	0	Tyrosine-protein kinase receptor UFO
Major histocompatibility class I proteins	P30508	HLA-C	0	0	1	0	1	0	HLA class I histocompatibility antigen, Cw-12 alpha chain
	P01892	HLA-A	0	0	0	3	0	0	HLA class I histocompatibility antigen, A-2 alpha chain
	P61769	B2M	1	0	0	1	0	0	Beta-2-microglobulin
Cell adhesion molecules	P14923	JUP	0	2	0	2	0		
	O60716	CTNND1	2	0	1	1	2	0	Junction plakoglobin
	P05556	ITGB1	0	0	0	20	3	1	Catenin delta-1
	P26006	ITGA3	1	0	0	0	19	0	Integrin alpha-3
	P1	ITGA2	0	0	1	20	3	0	Integrin alpha-2

	73 01	A2							
	P3 52 21	CT NN A1	2	0	0	4	0	0	Catenin alpha-1
	P4 31 21	MC AM	0	0	0	0	18	0	Cell surface glycoprotein MUC18
	Q0 24 87	DS C2	0	0	1	0	0	0	Desmocollin-2
	P1 61 44	ITG B4	0	0	1	0	0	1	Integrin beta, Isoform Beta- 4A
	P1 60 70	CD 44	0	0	0	1	0	0	CD44 antigen
	P2 19 26	CD 9	0	0	0	3	0	0	CD9 antigen
	P4 85 0	CD 151	0	0	0	0	1	0	CD151 antigen
G protein- coupled receptors	P6 28 73	GN B1	15	2	3	3	0	0	Guanine nucleotide-binding protein G(I)/G(S)/G(T) subunit beta-1
	P6 28 79	GN B2	12	0	1	7	1	0	Guanine nucleotide-binding protein G(I)/G(S)/G(T) subunit beta-2
	P0 48 99	GN AI2	11	4	0	2	0	0	Guanine nucleotide-binding protein G(i) subunit alpha-2
	P4 89 60	CD 97	2	0	8	0	1	1	CD97 antigen
	P6 39 02	GN AS	5	2	1	0	1	0	Guanine nucleotide-binding protein G(i) subunit alpha-1
	Q1 43 44	GN A13	0	3	0	0	3	0	Guanine nucleotide-binding protein subunit alpha-13