
Supplementary information

**Isolation and characterization of
extracellular vesicle subpopulations from
tissues**

In the format provided by the
authors and unedited

Supplementary table 1 - Antibodies, EV-binding beads, cell line, instrument and software used for evaluation of enzymatic effect by flow cytometry

Antibody	vendor	cat. no.	RRID	clone	fluorochrome	dilution (cells)	dilution (EVs)
anti-CD9	BD Biosciences	341647	AB_400240	M-L13	PE		
anti-CD63	BD Biosciences	557305	AB_396631	H5C6	PE	20 µl/	15 µg/10
anti-CD81	BD Biosciences	561957	AB_10892807	JS-81	PE	500,000	µg EVs
Mouse IgG1 k Isotype control	BD Biosciences	555749	AB_366091	MOPC-21	PE	cells	

EV-binding beads	vendor	cat. no.	dilution
Exosomes - Human			10,000
CD63 Isolation/ Detection Reagent	Invitrogen	10606D	beads/10 µg EVs

Cell line	vendor	cat. no.	RRID
HMC-1	DSMZ	ACC-283	CVCL_0003

Instrument	vendor	used for
BD FACSVerse™	BD Biosciences	data acquisition

Software	vendor	used for
BD FACS Suite™	BD Biosciences	data collection
Flow Jo 7.6.5	BD Biosciences	data analysis

Supplementary table 2 - Information about the number of cells and bead-EV complexes in the gates shown in Extended data Figure 2

	% beads in the gate out of the total events (10 000 events collected in total)			
	Large EVs - Non-treated	Large EVs - Treated	Small EVs - Non-treated	Small EVs - Treated
Isotype Control - Exp 1	88,2	88,1	82,9	88,5
Anti-CD63 - Exp 1	92	95,2	95,6	94,8
Anti-CD9 - Exp 1	87,7	84,3	81,7	70,4
Anti-CD81 - Exp 1	88,6	85,8	87,9	79,6
Isotype Control - Exp 2	79,3	69,8	77	60,8
Anti-CD63 - Exp 2	86,5	90,2	90,4	96,2
Anti-CD9 - Exp 2	78	72	64,9	76,5
Anti-CD81 - Exp 2	79	73,4	68,8	69,2
Mean	84,9	82,4	81,2	79,5
SD	5,3	9,4	10,5	12,8

T-test large EVs non-treated vs treated 0,51
T-test small EVs non-treated vs treated 0,78
T-test large EVs non-treated vs small EVs non- treated 0,38
T-test large EVs treated vs small EVs treated 0,62

	% cells in the gate out of the total events (10 000 events collected in total)	
	Non-treated	Treated
Unstained - Exp 1	92,1	94,1
Isotype Control - Exp 1	92	91,2
Anti-CD63 - Exp 1	94,1	94,5
Anti-CD9 - Exp 1	83,9	81,9
Anti-CD81 - Exp 1	93,2	94,2
7AAD - Exp 1	90,9	94,6
Unstained - Exp 2	86,6	88,8
Isotype Control - Exp 2	88	89,2
Anti-CD63 - Exp 2	88,2	88,5
Anti-CD9 - Exp 2	70,3	64,2
Anti-CD81 - Exp 2	86,8	87,6
7AAD - Exp 2	88,3	88,2
Mean	87,9	88,1
SD	6,3	8,4

T-test non-treated vs treated 0,94