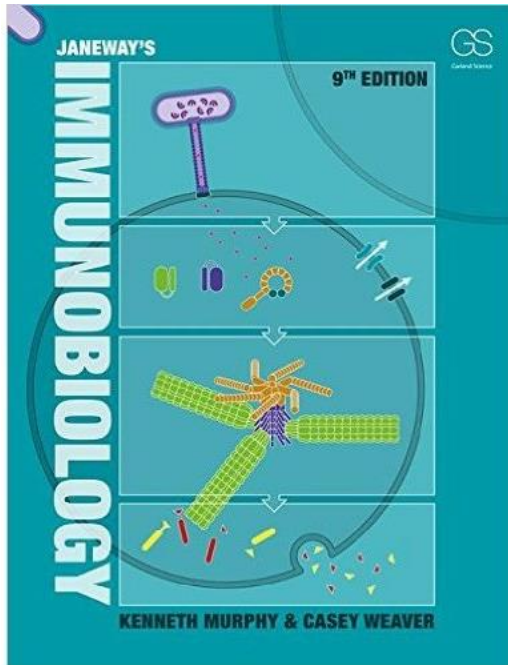


Intézeteti beszámoló^{v2}



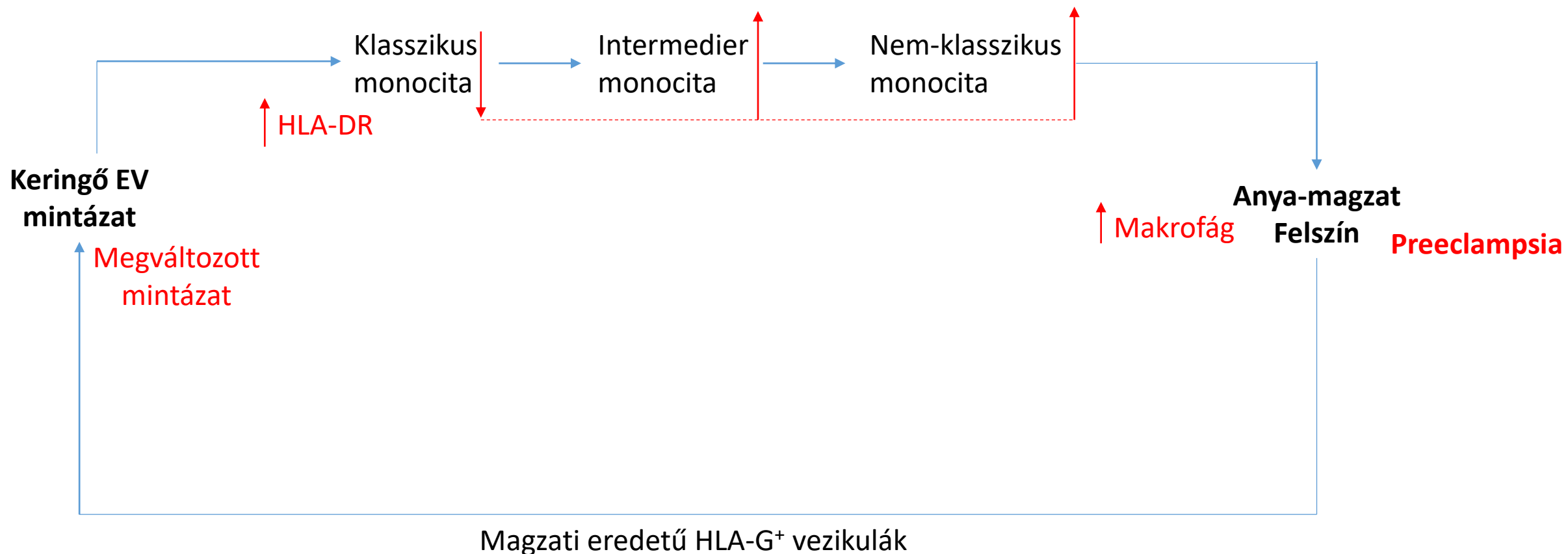
2016. 11. 03



“The fetus is an allograft that is tolerated repeatedly”

Janeway, Immunobiology 9th edition, 2017

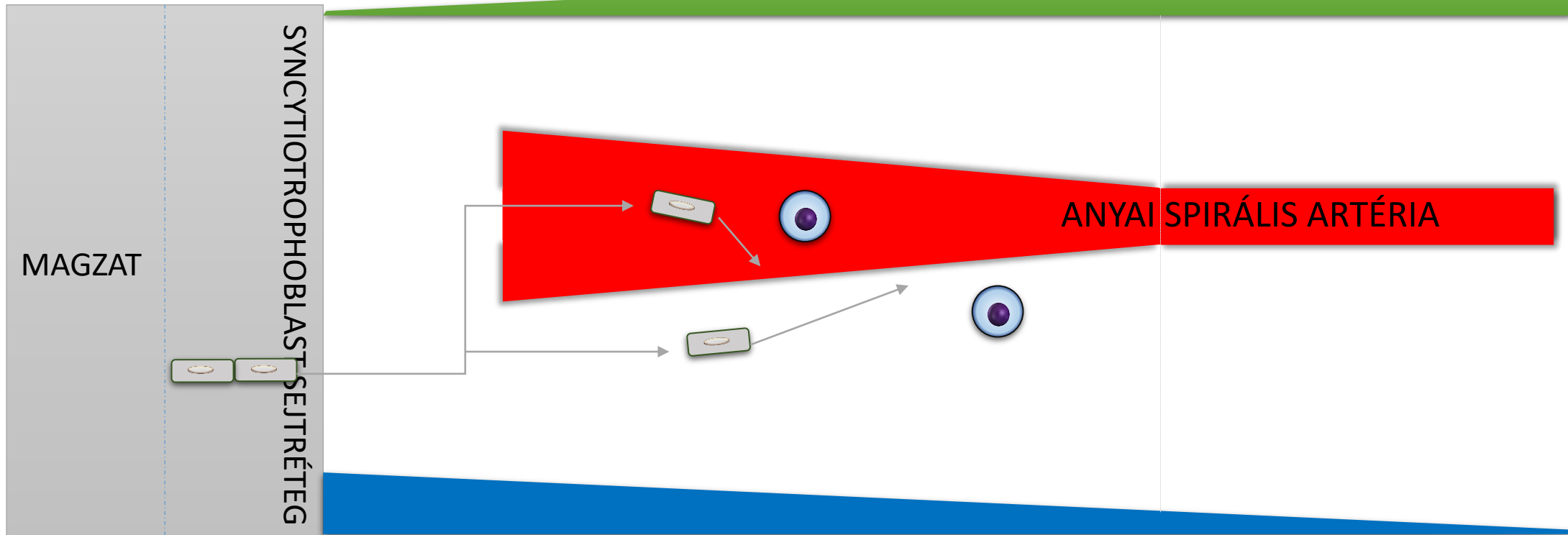
Monocyta macrophag rendszer várandósságban



Spirális artéria remodelling modellje

ANYAI MAGZATI HATÁRFELÜLET

Anyai eredetű extracelluláris vezikula koncentráció



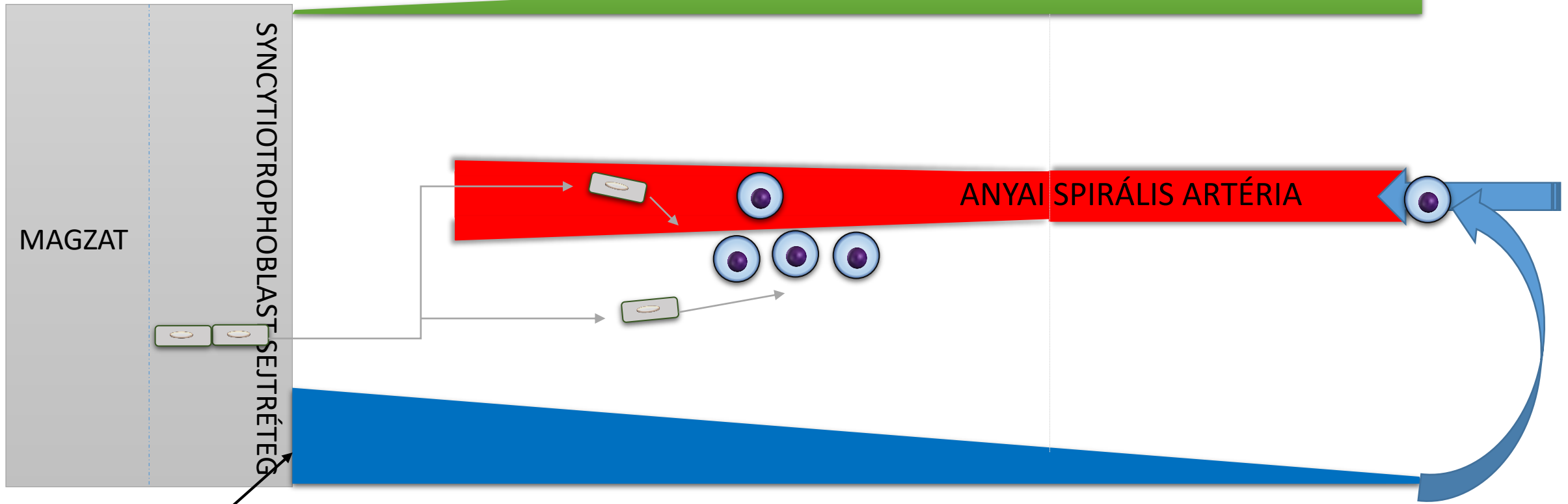
Magzati eredetű extracelluláris koncentráció

ÁTALAKULT SPIRÁLIS ARTÉRIA

Preeclampsia patomechanizmusa

ANYAI MAGZATI HATÁRFELÜLET

Anyai eredetű extracelluláris vezikula koncentráció



Magzati eredetű extracelluláris koncentráció

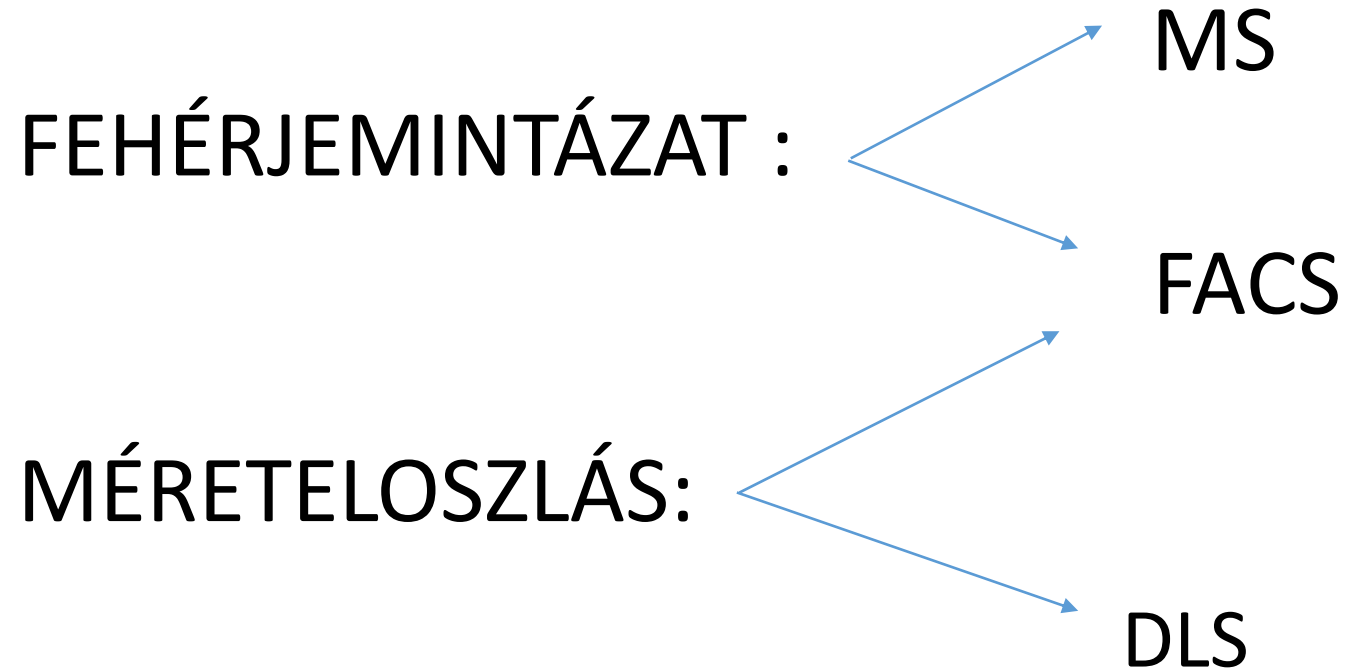
Megváltozott vezikula cargo
+ Fokozott apoptózis

TÖKÉLETLENÜL ÁTALAKULT SPIRÁLIS ARTÉRIA

Célkitűzések

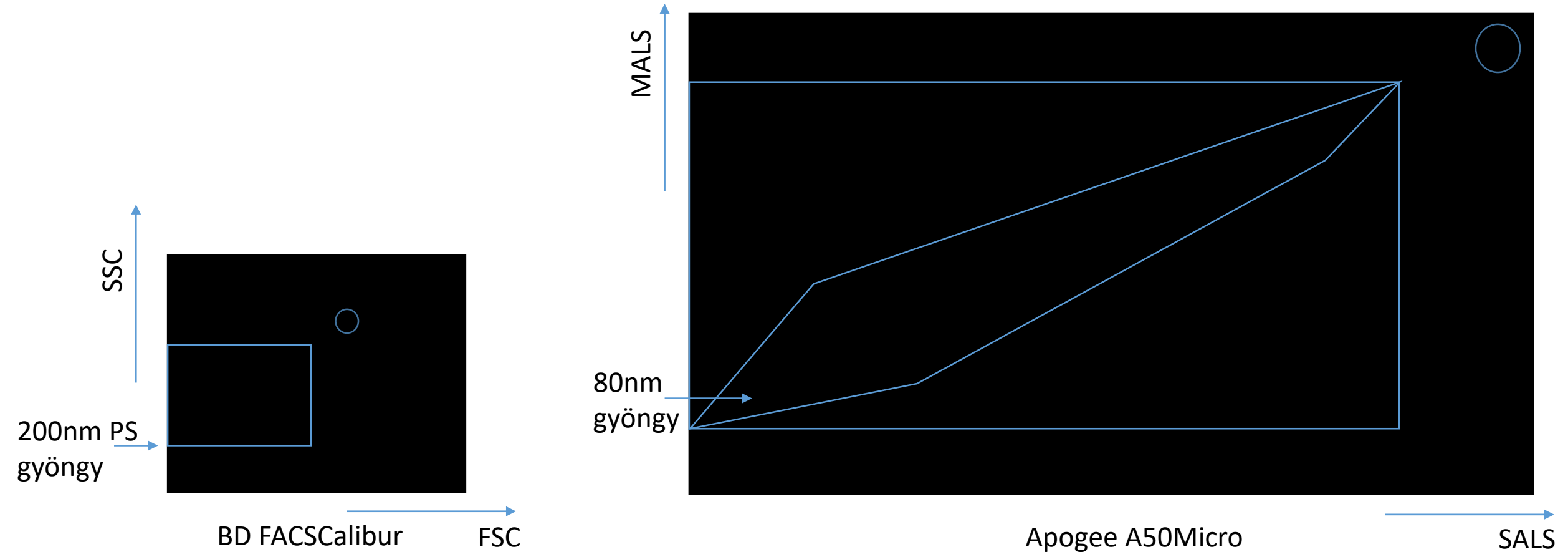
- I. Keringő extracelluláris mintázat jellemzése
- II. In vitro EV hatás vizsgálata
 1. A trophoblast sejtek migrációjára (Spiralis artéria remodelling)
 2. A monocita sejtek migrációjára és fenotípus változás (Monocita toborzás)
- III. Észlelt EV hatás molekuláris mechanizmusának magyarázata

I. Vezikula mintázat jellemzése

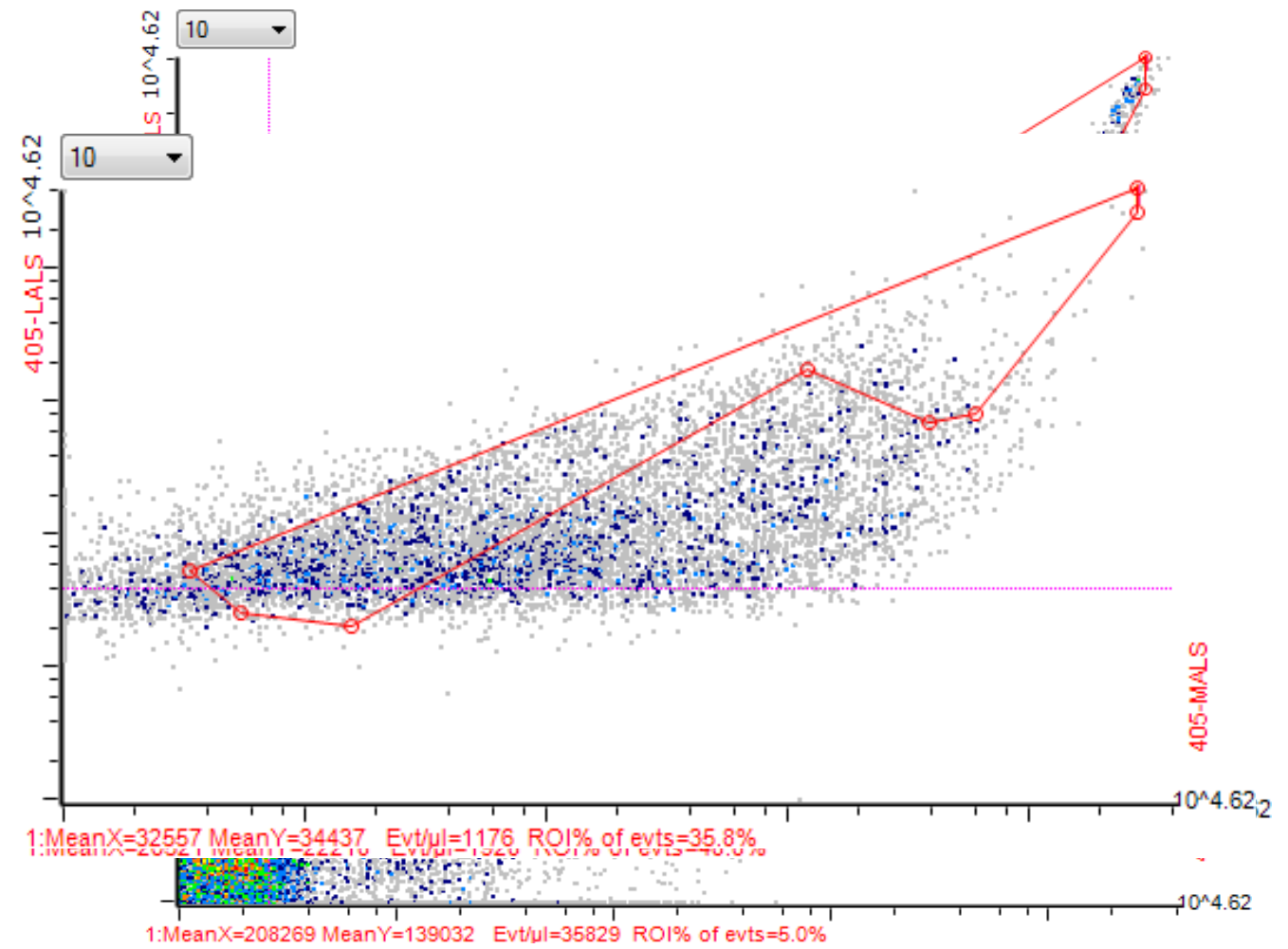
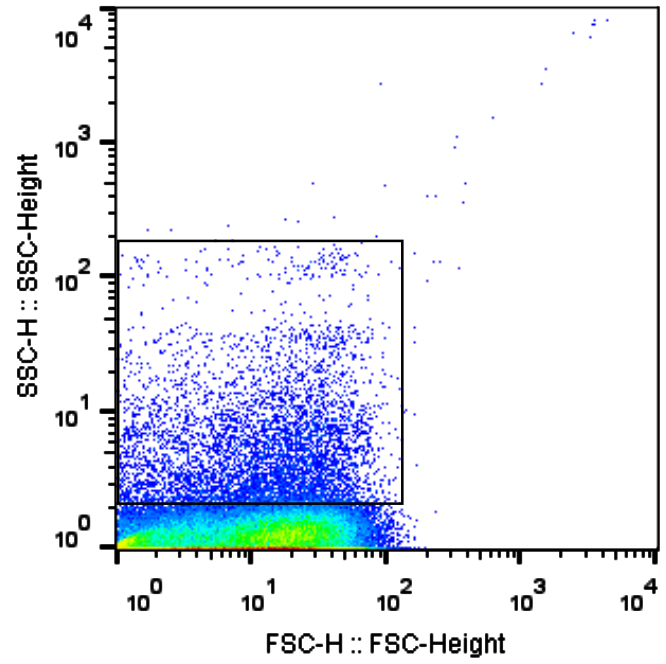


ELMI

Vezikula populáció jellemzése – Áramlási citometria

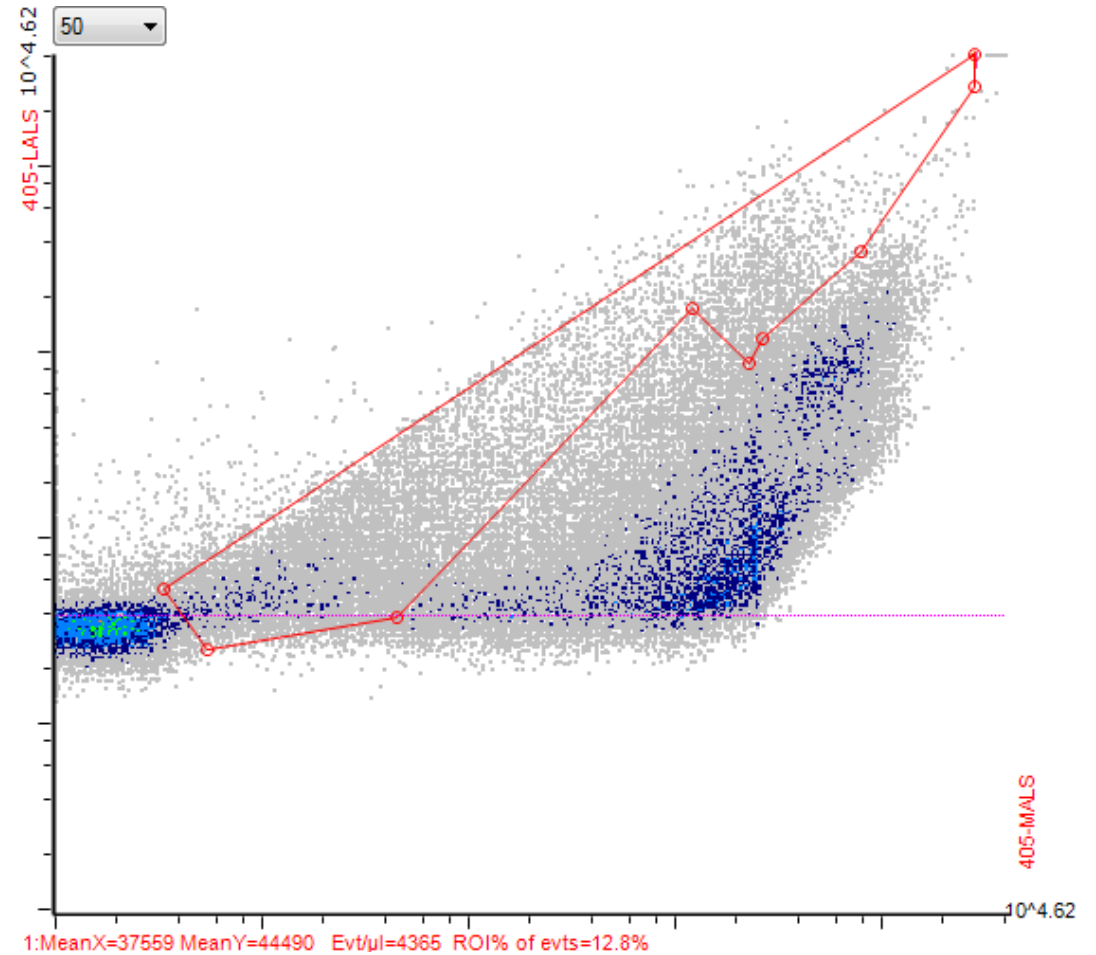
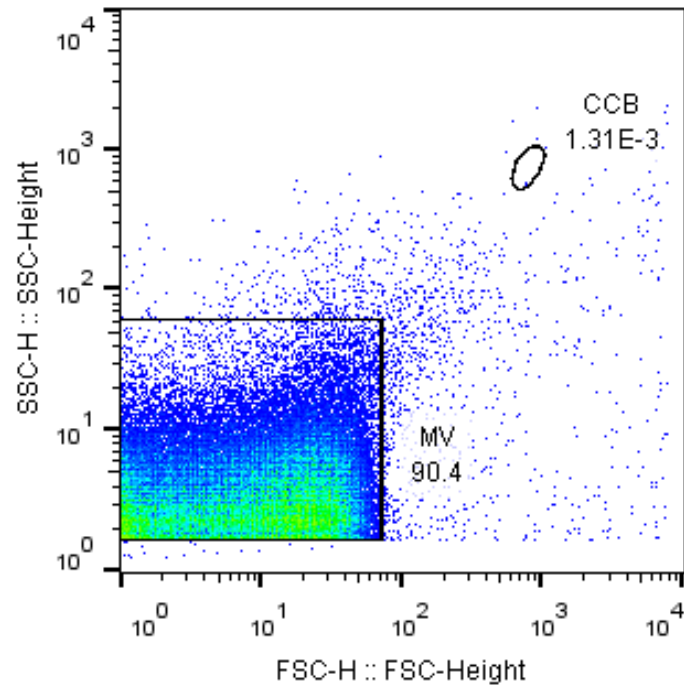


Magas felbontású áramlási citometria

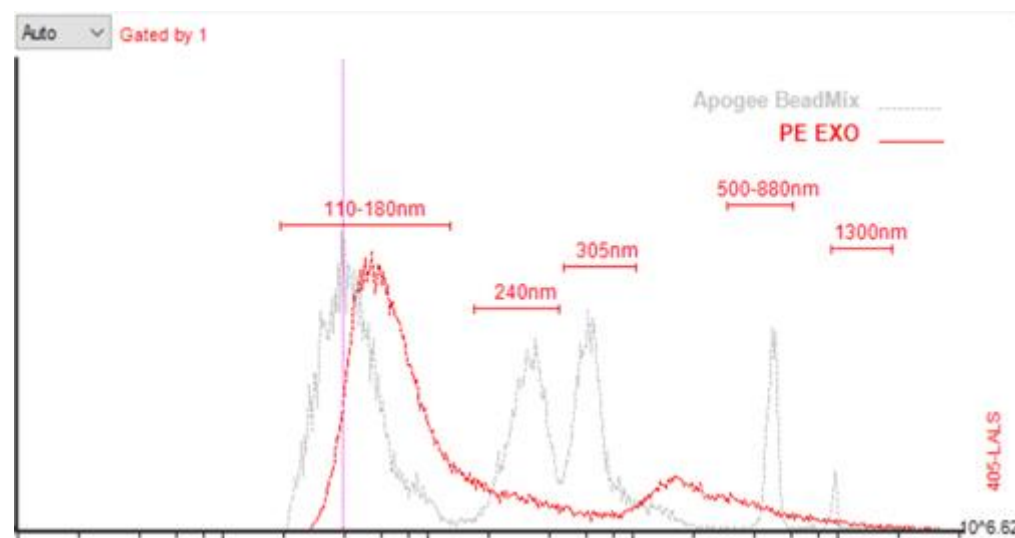
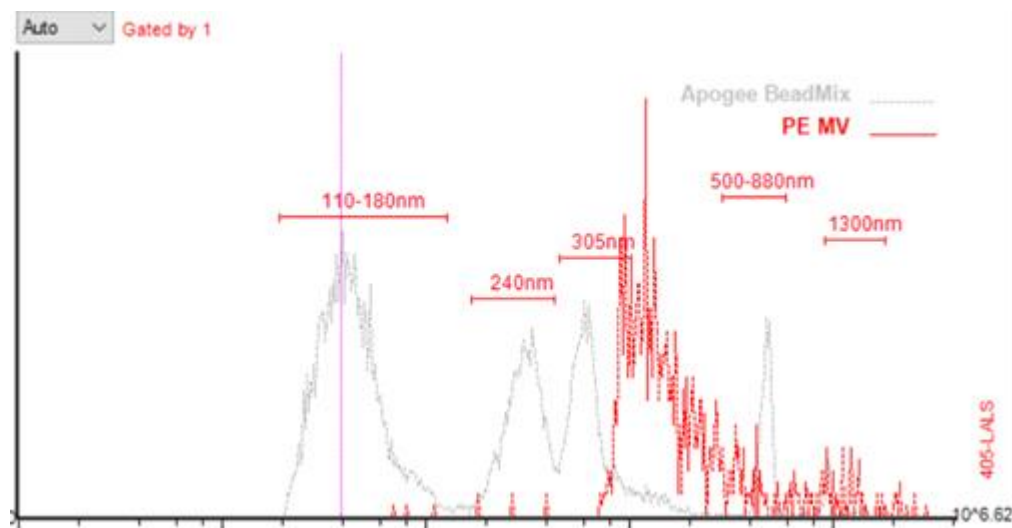
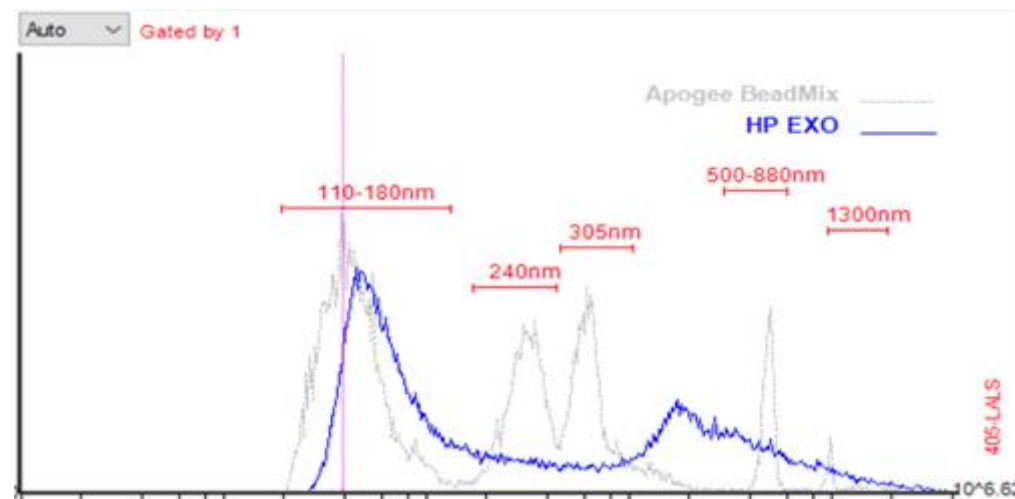
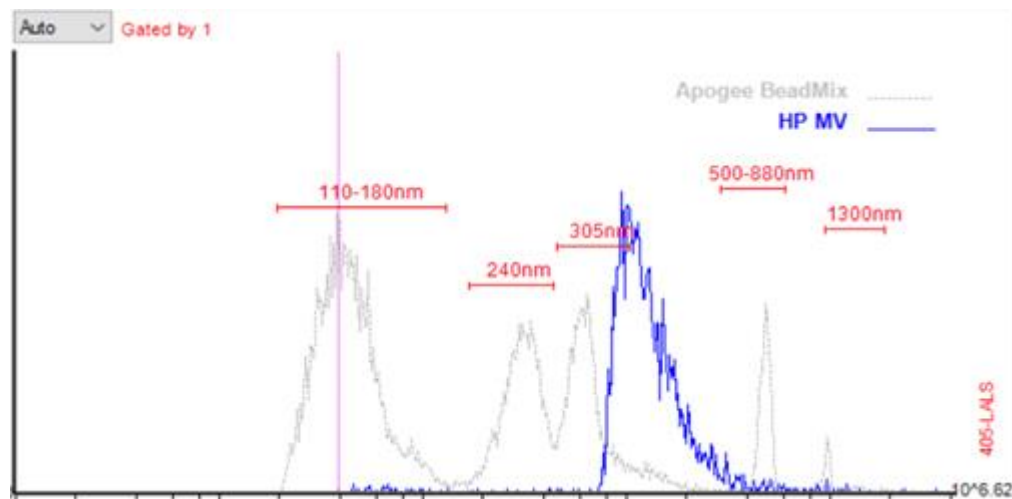


Magas felbontású áramlási citometria

PBS



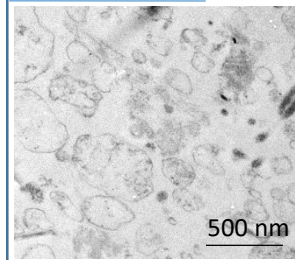
Magas felbontású áramlási citometria



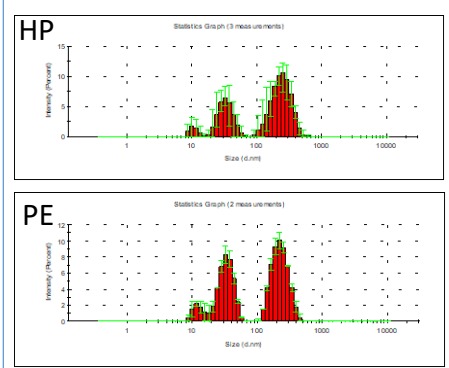
CIRCULATING EV PATTERN

12.5K pellet MIKROVESICLE

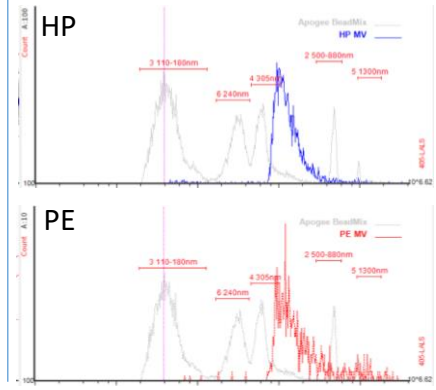
ELMI



DLS

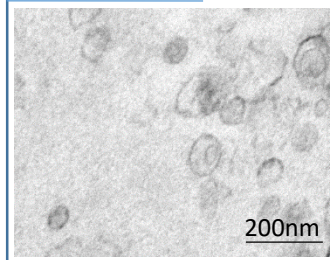


APOGEE

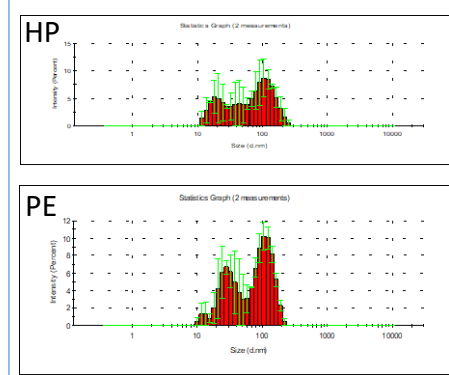


100K pellet EXOSOME

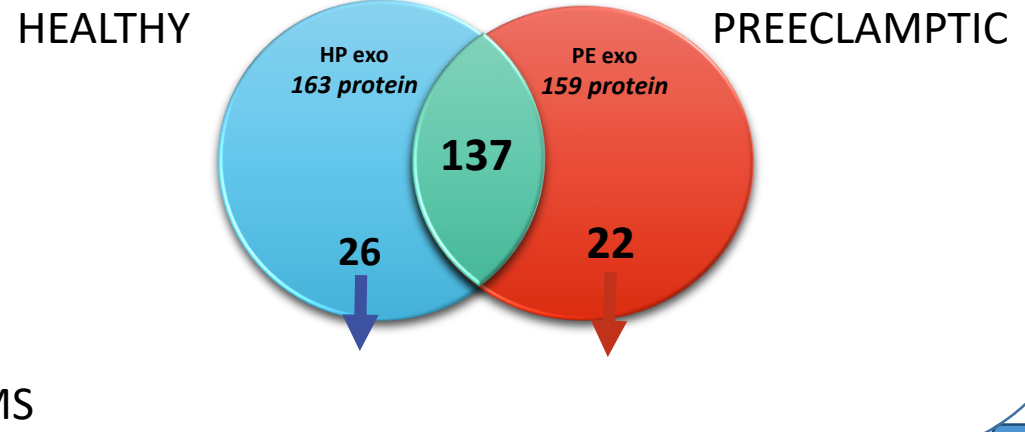
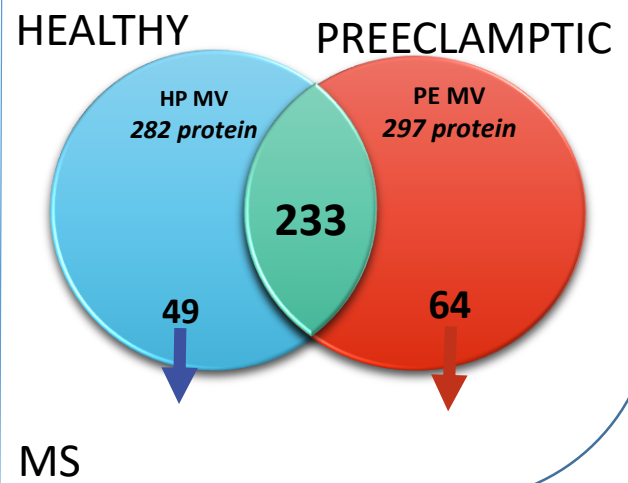
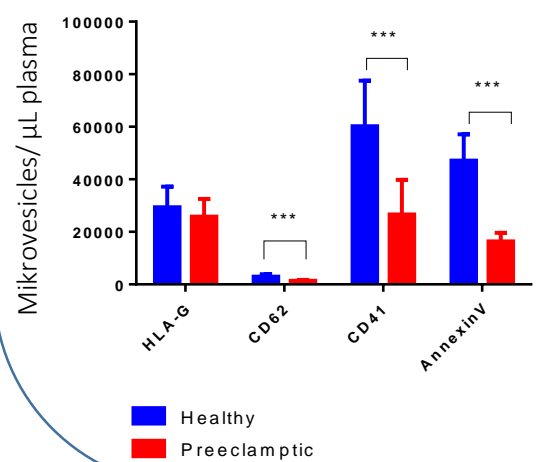
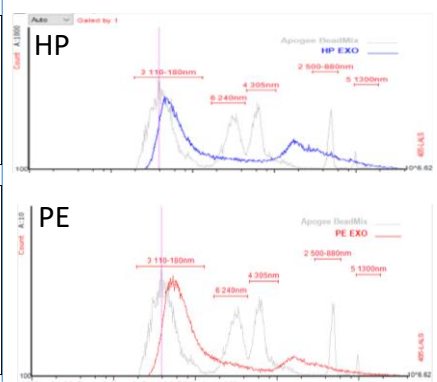
ELMI



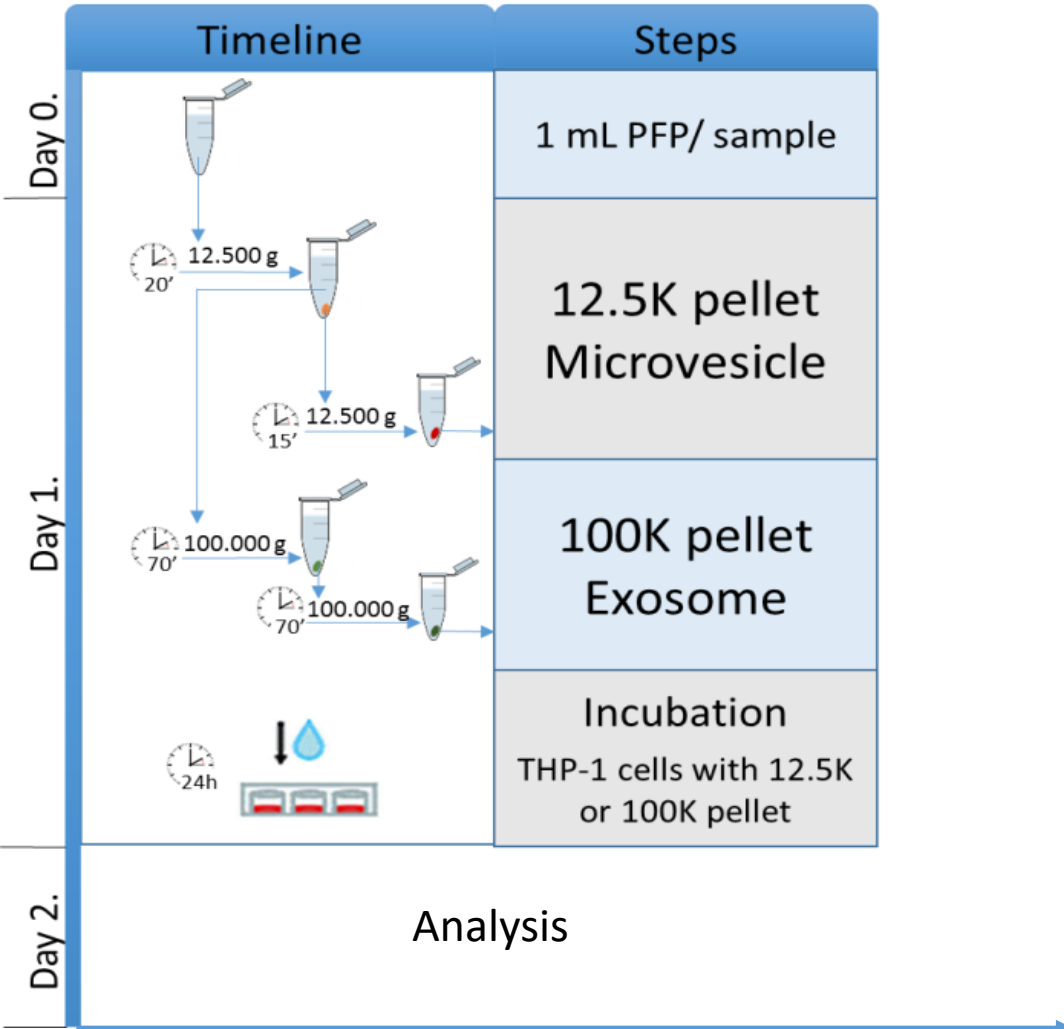
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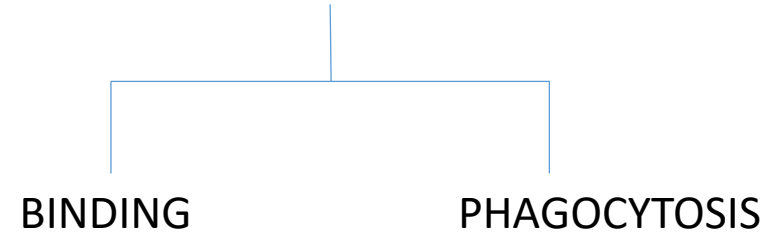
APOGEE



Workflow



THP-1 CELL – EV INTERACTION



Workflow

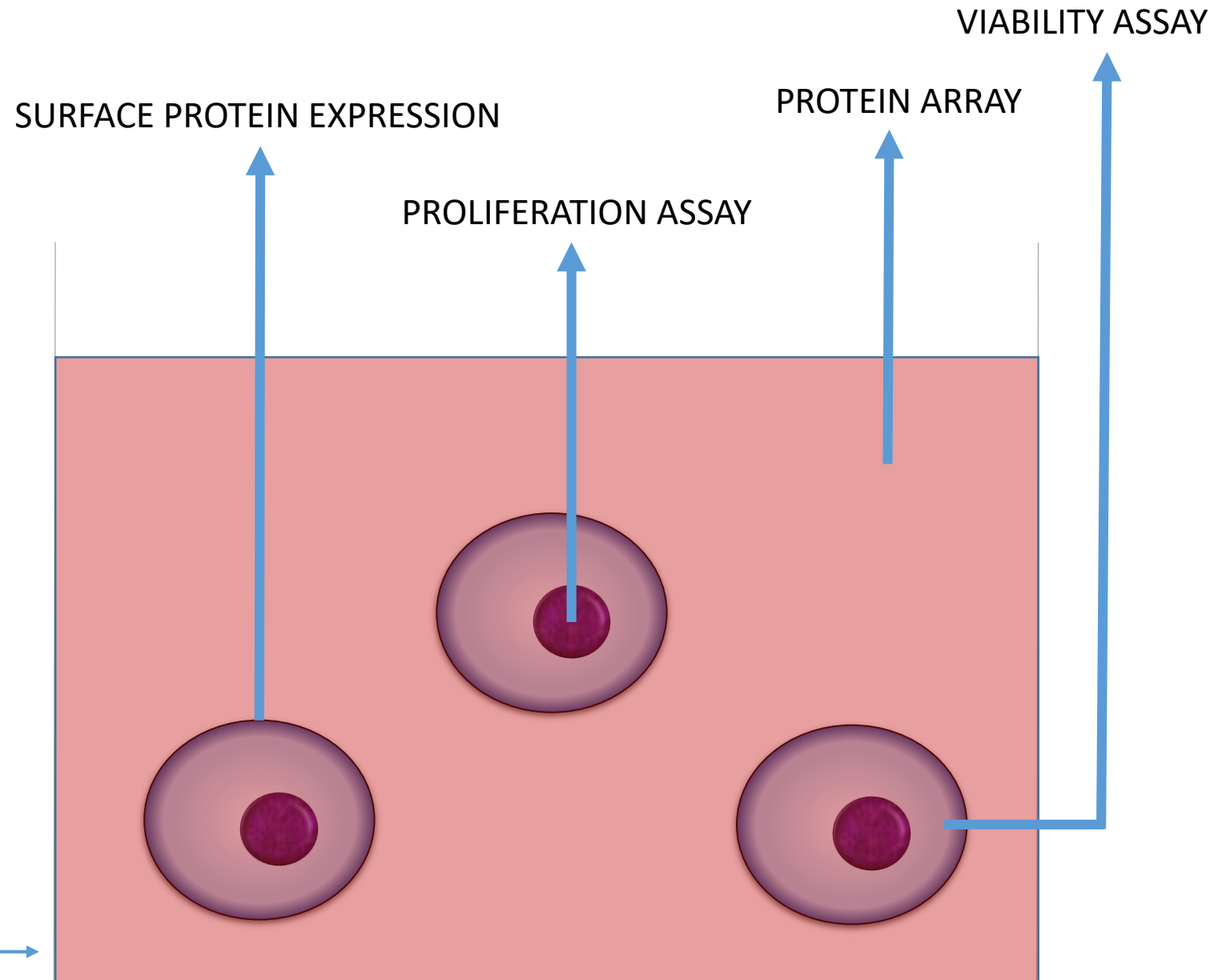
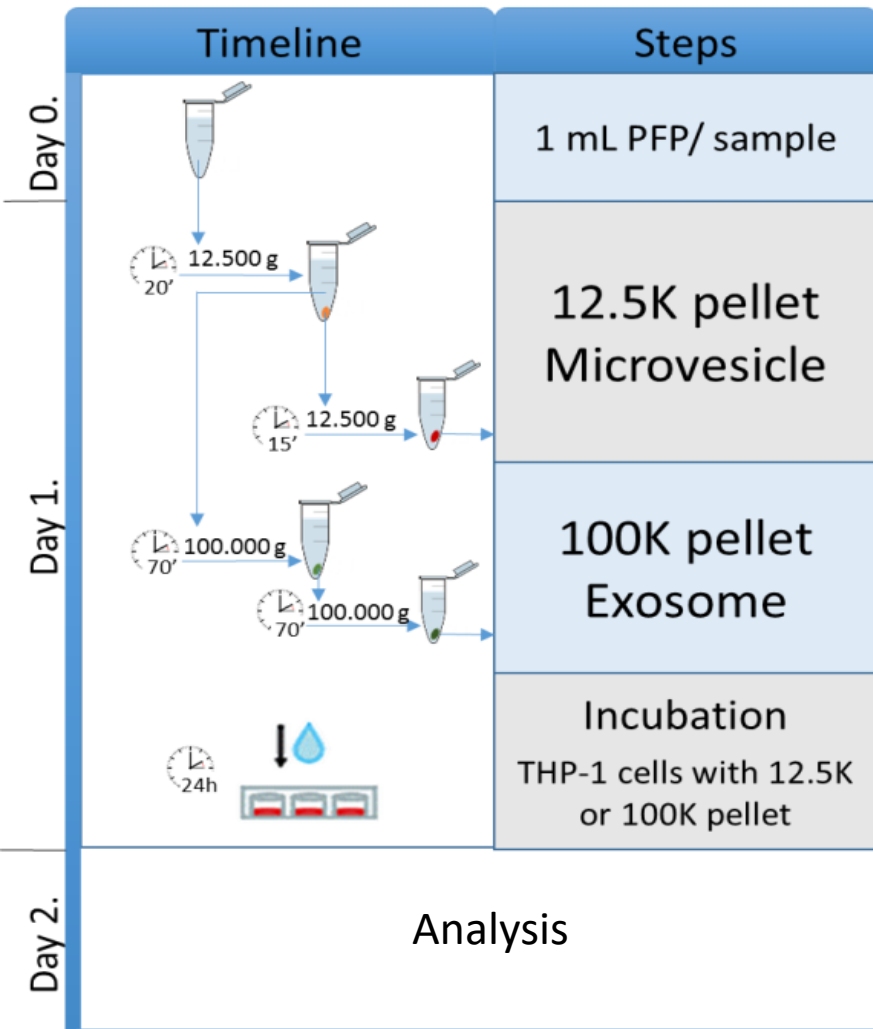
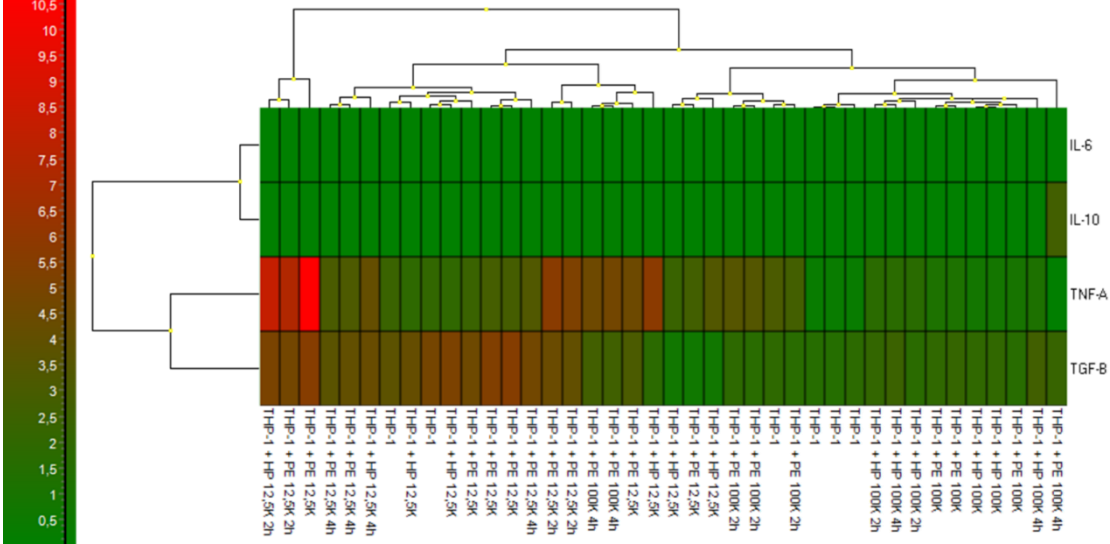
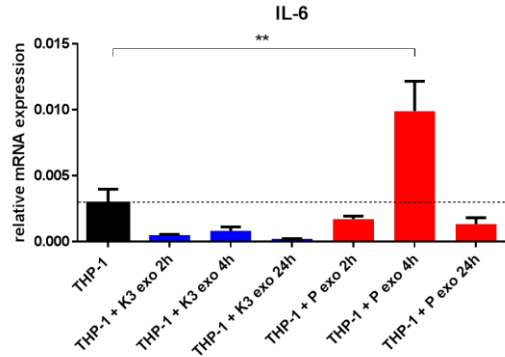
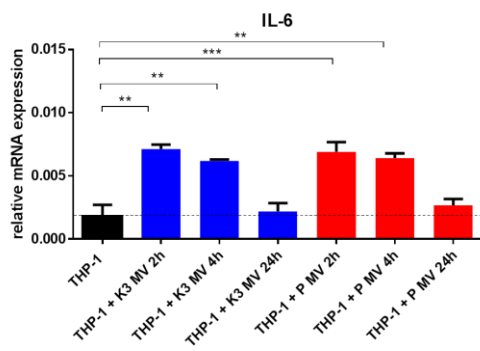


Figure 3.



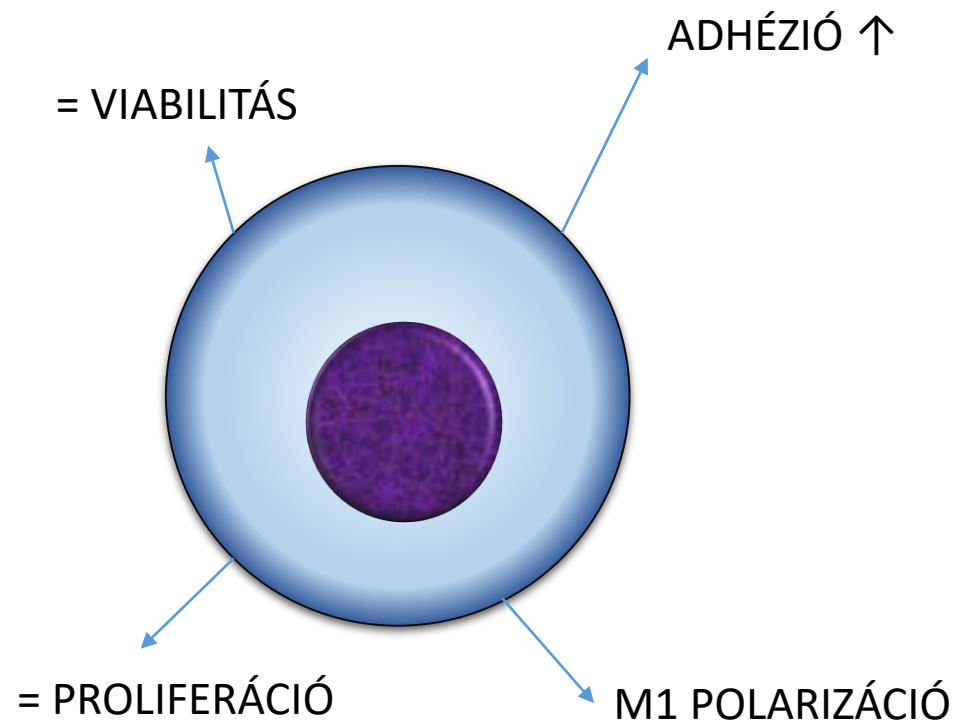
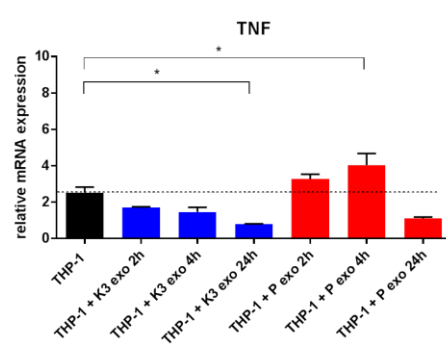
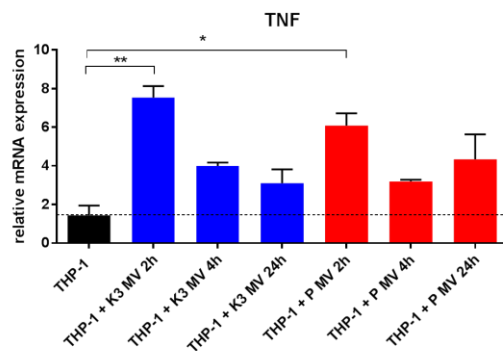
Microvesicle 12.5K pellet treatment

Exosome 100K pellet treatment



Microvesicle 12.5K pellet treatment

Exosome 100K pellet treatment



Következő lépés: November 2016

FACS - TNF THP-1 sejtekben

Workflow

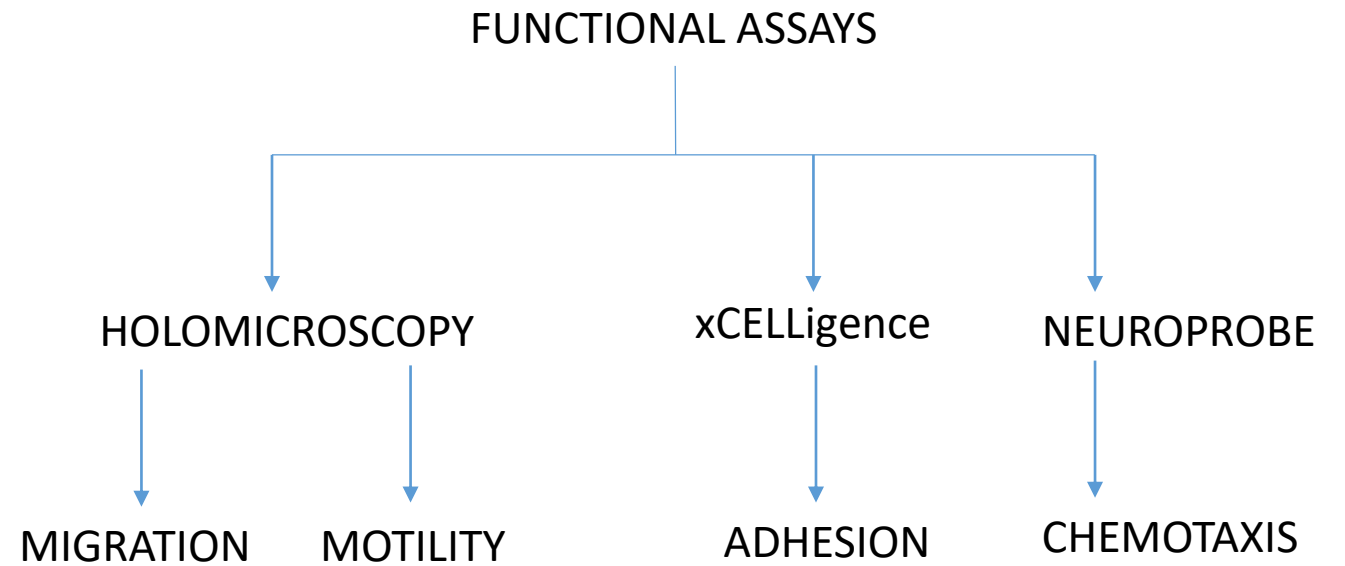
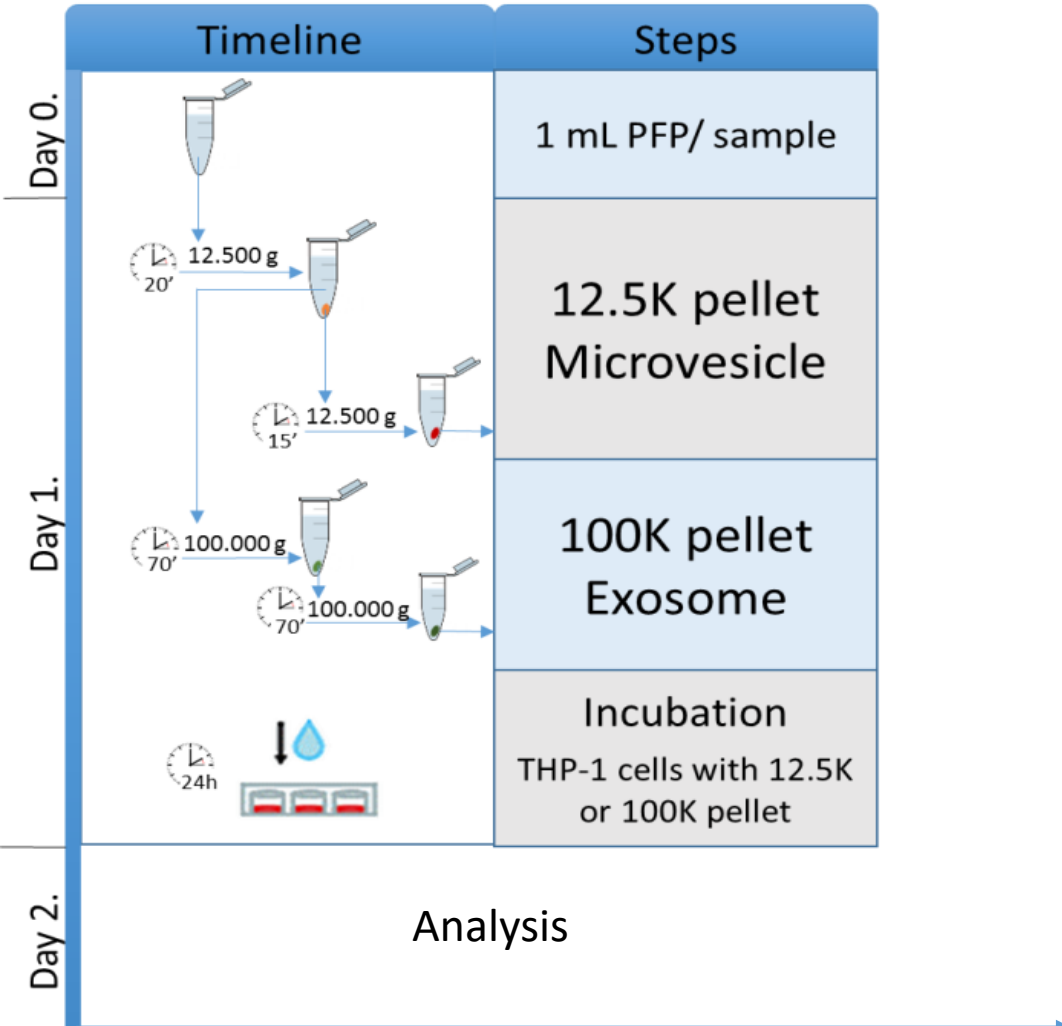
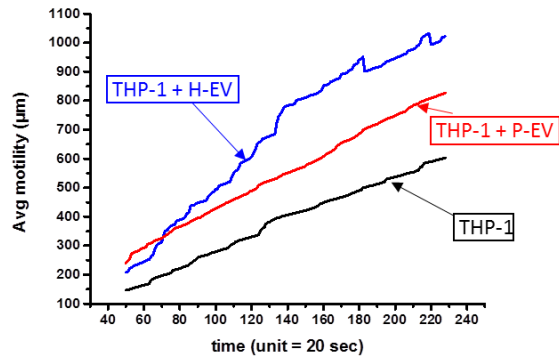
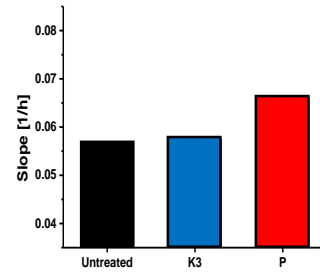
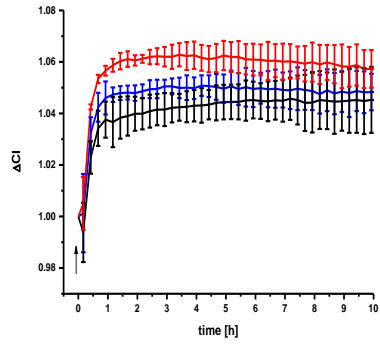
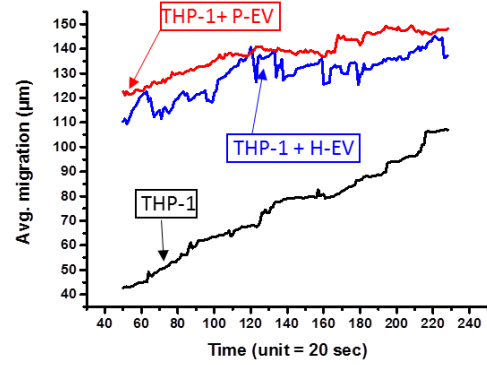


Figure 1.

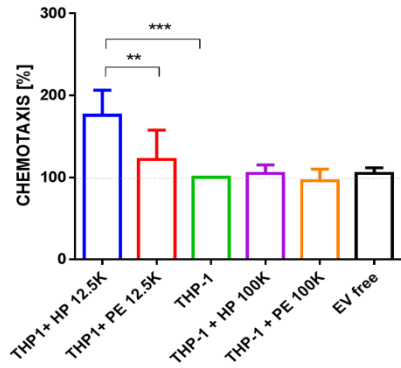
H-EV VS P-EV EFFECT ON CELL MOTILITY



H-EV VS P-EV EFFECT ON CELL MIGRATION



	Healthy EV treated THP-1	Untreated THP-1	Preeclamptic EV treated THP-1
CD44	29.7±1.9	124.2	46.3±3.4
INTEGRIN ALFA 2	17.5±4.8	19.93	9±0.9
INTEGRIN BETA 1	251.5±11.5	251.4±19.8	273.4±10.8



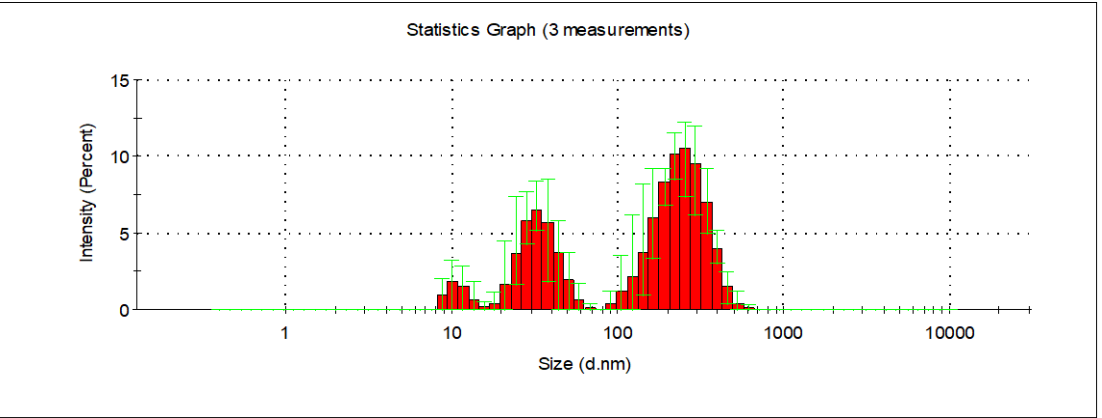
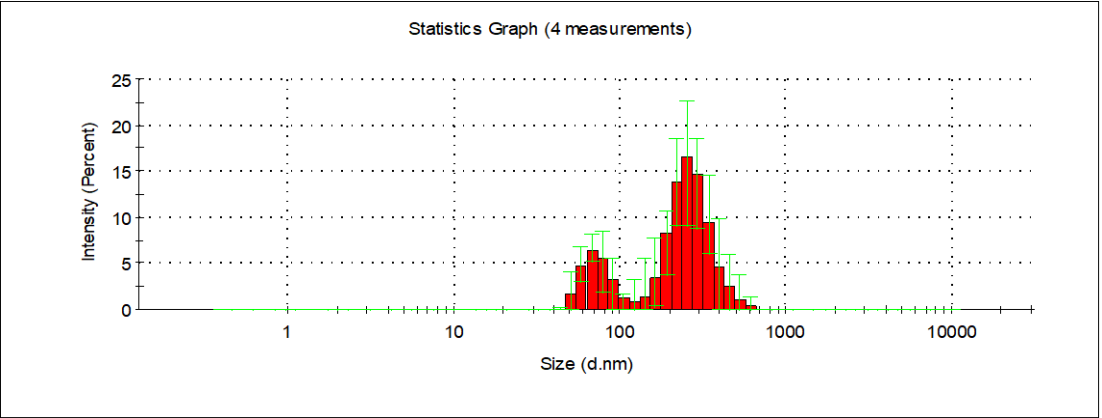
ANOVA, p<0.05, Dunnett's test

MASS SPECTROMETRY

HP-EV only	PE-EV only	HP and PE common
ITGA6	ICAM3	ITGM
CDC42	RAP2A	MYH9
	BCAM	THBS1
	BSG	ITGB3
	THBS4	ITGB2b
		ITGB2
		ITGA6
		MMRN1
		CD36
		TLN1
		MMP9
		CD99
		RHOA

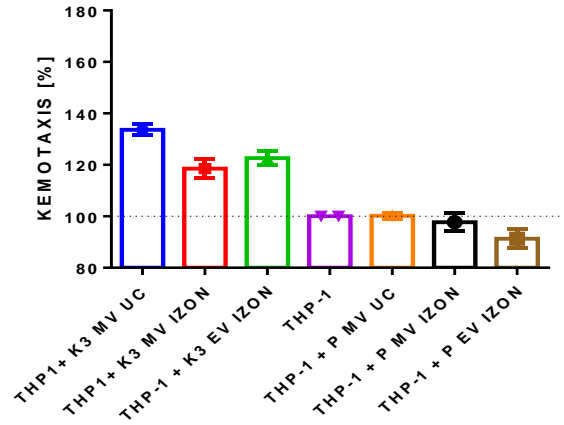
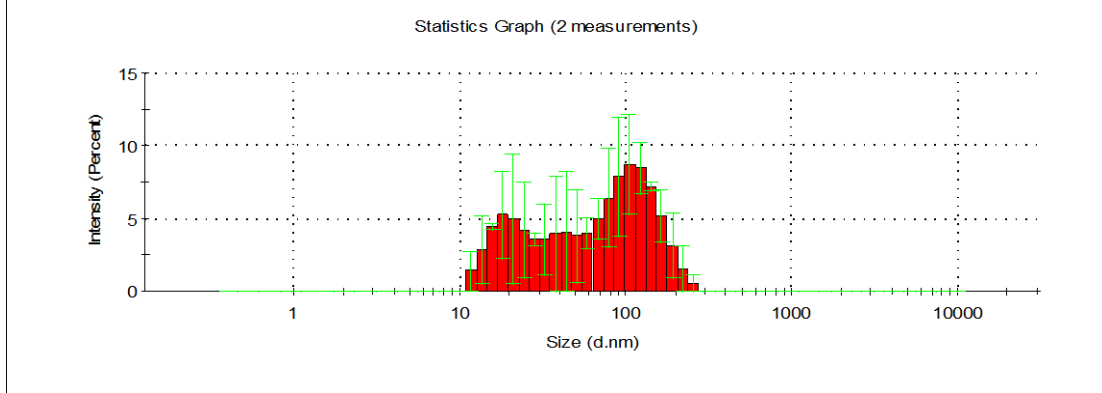
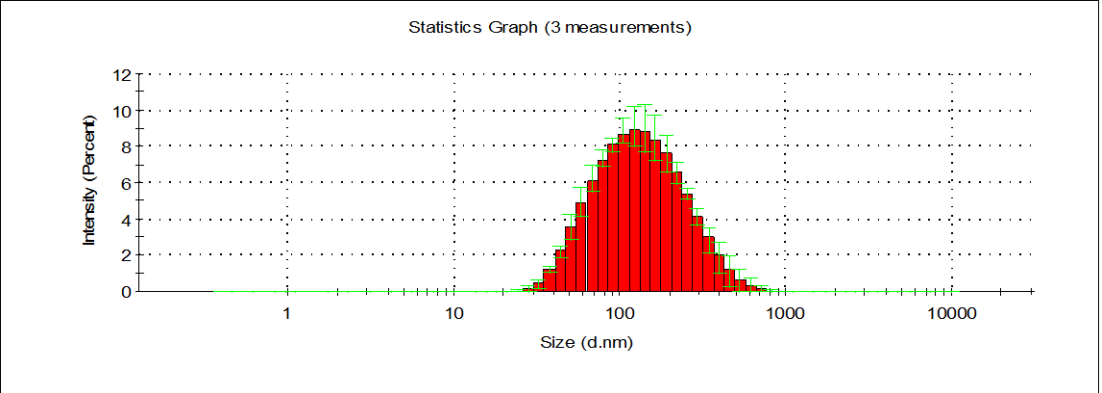
Figure S1.

MV
Izon qEV



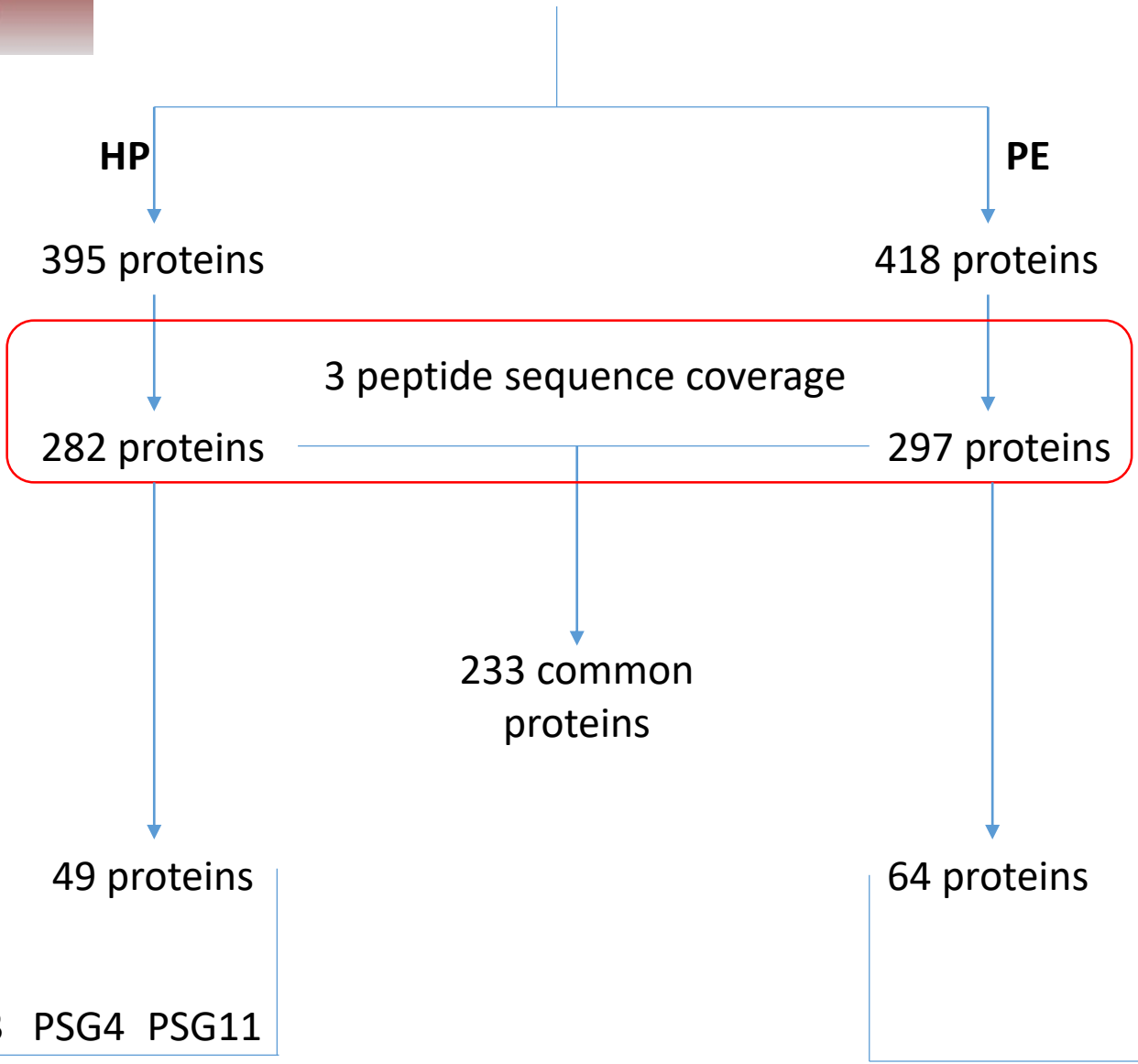
Centrifuged

EXO





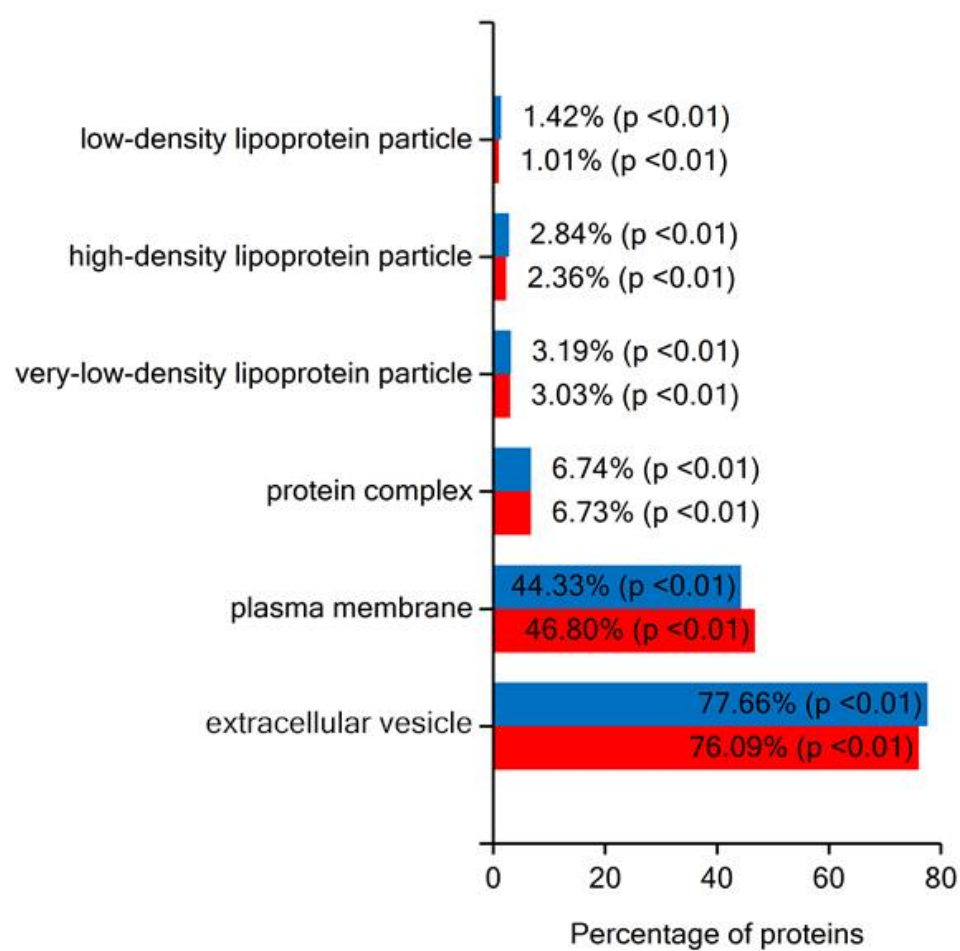
12.5K pellet (n=7 per group)



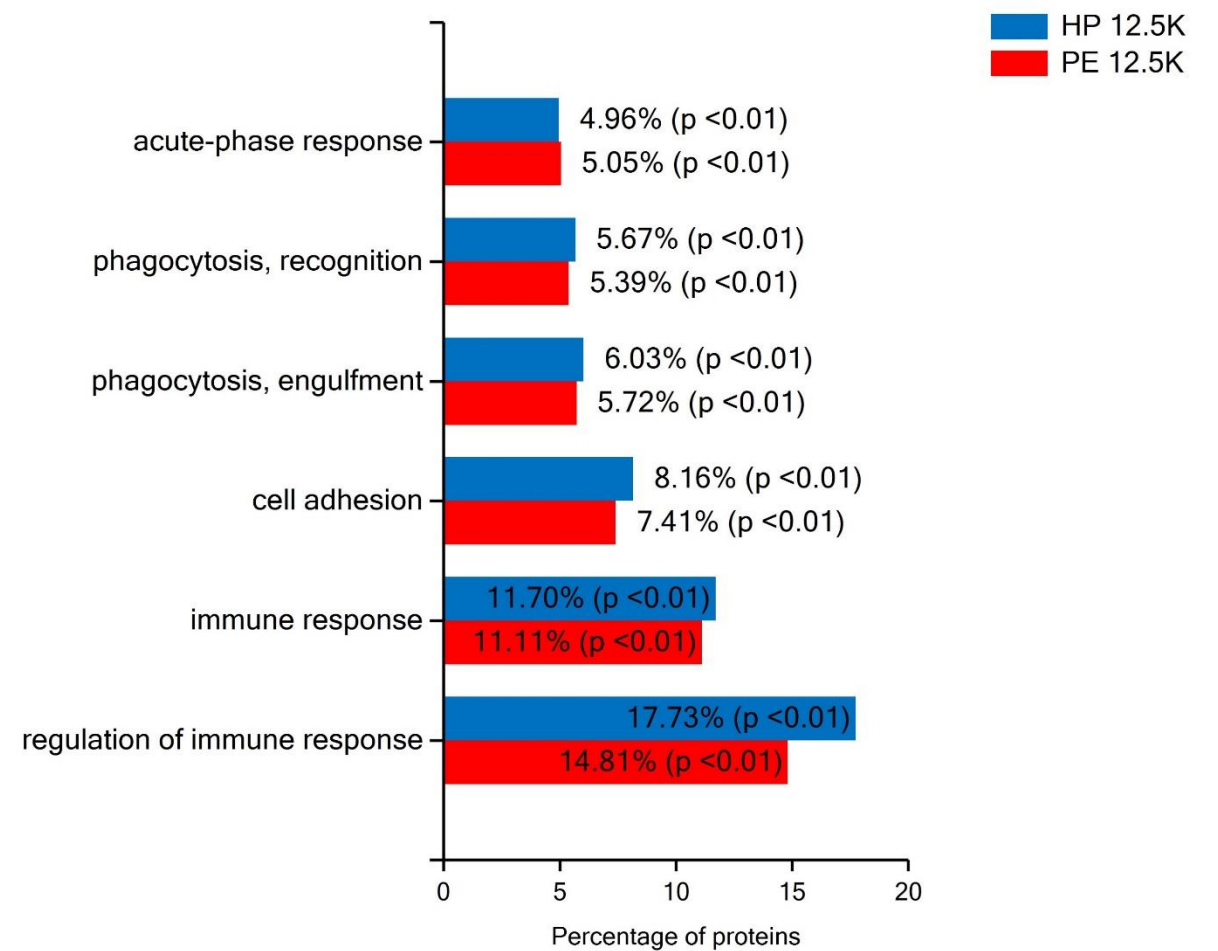
ITGA6 PSG3 PSG4 PSG11

Figure S2.

Cellular component for HP 12.5K and PE 12.5K



Biological process for HP 12.5K and PE 12.5K



HP 12.5K MV pellet only

Proteins involved in vesicular trafficking: actinin, alpha 4 (ACTN4) – associates with the CART complex;

Potential inhibitor of angiogenesis: serpin peptidase inhibitor, clade F 1 (SERPINF1) –

Cell migration: cell division cycle 42 (CDC42) - Mediates CDC42-dependent cell migration; Integrin alpha 6 (ITGA6)

Pregnancy specific proteins: pregnancy specific beta-1-glycoproteins 11, 3, (PSG11, PSG3,)

Regulation of phagocytosis: ras-related C3 botulinum toxin substrate 1 (RAC1)

PE 12.5K MV pellet only

Cell adhesion proteins: Intercellular adhesion molecule 3 (ICAM3), RAP2A, basal cell adhesion molecule (BCAM), basigin (BSG), thrombospondin 4 (THBS4)

Phagocytosis: ras-related C3 botulinum toxin substrate 2 (RAC2)

Pregnancy specific proteins: pregnancy specific beta-1-glycoproteins 9, (PSG9), PAPP-A, alkaline phosphatase, placental (ALPP)

Vesicular transport: RAB1B, Flotillin1,

TGF-beta induced

12.5K MV pellet common

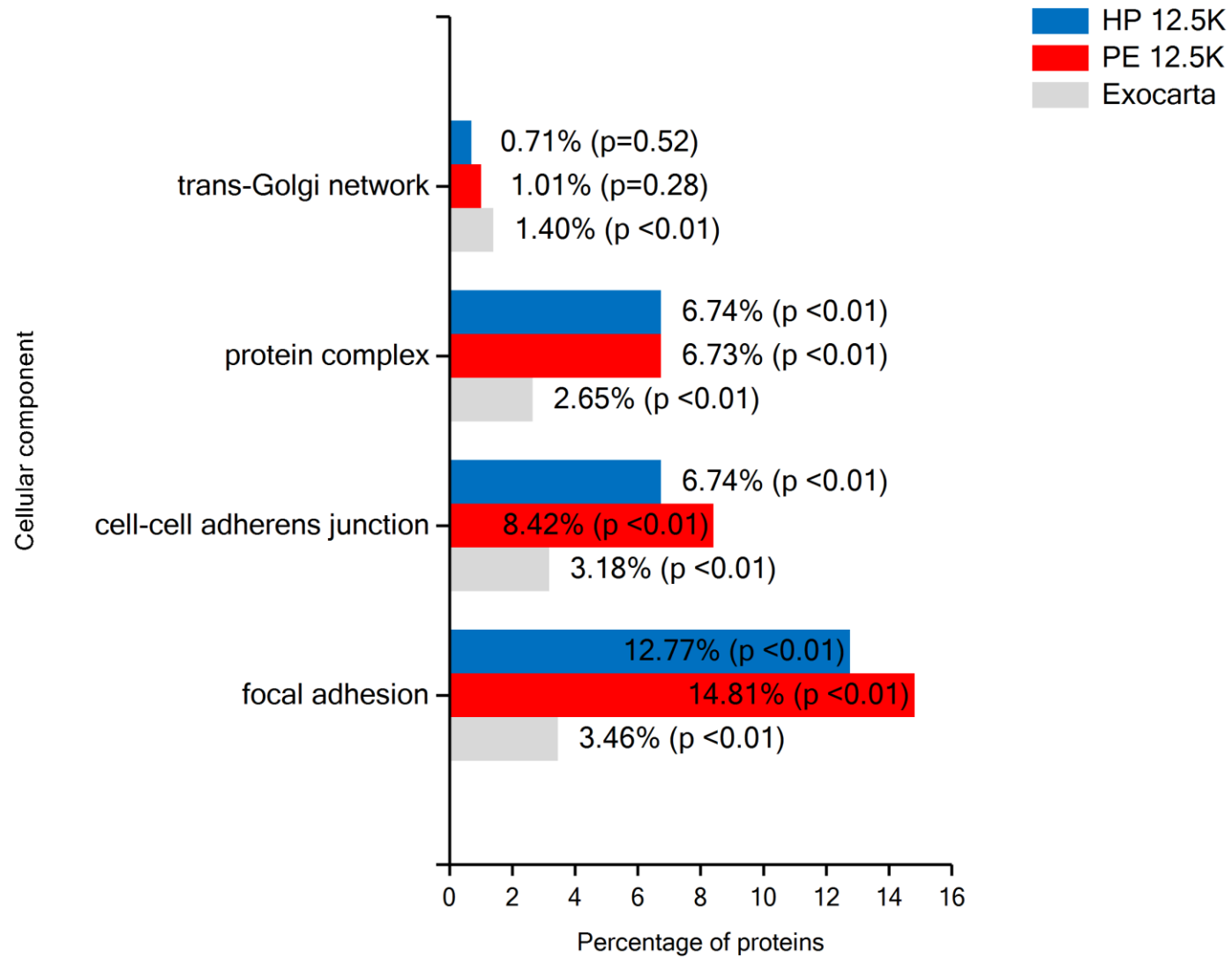
Migration related: integrin, alpha M (ITGM) – adhesion of monocytes, Myosin (MYH9), thrombospondin 1 (THBS1), integrin beta3 (ITGB3), integrin alfa2b (ITGB2b), multimerin 1(MMRN1), integrin beta 2(ITGB2), CD36 (thrombospondin receptor), talin1 (TLN1), MMP9, CD99 (T-cell adhesion), Integrin alpha6 (ITGA6), CD44, ras homolog family member A (RHOA) – adhesion,

Vesicular transport: Clathrin (CLTC), RAB1B, adenylate cyclase-associated protein 1 (CAP1) – mRNA localization, Flotillin 2 (FLOT2), RAB1A,

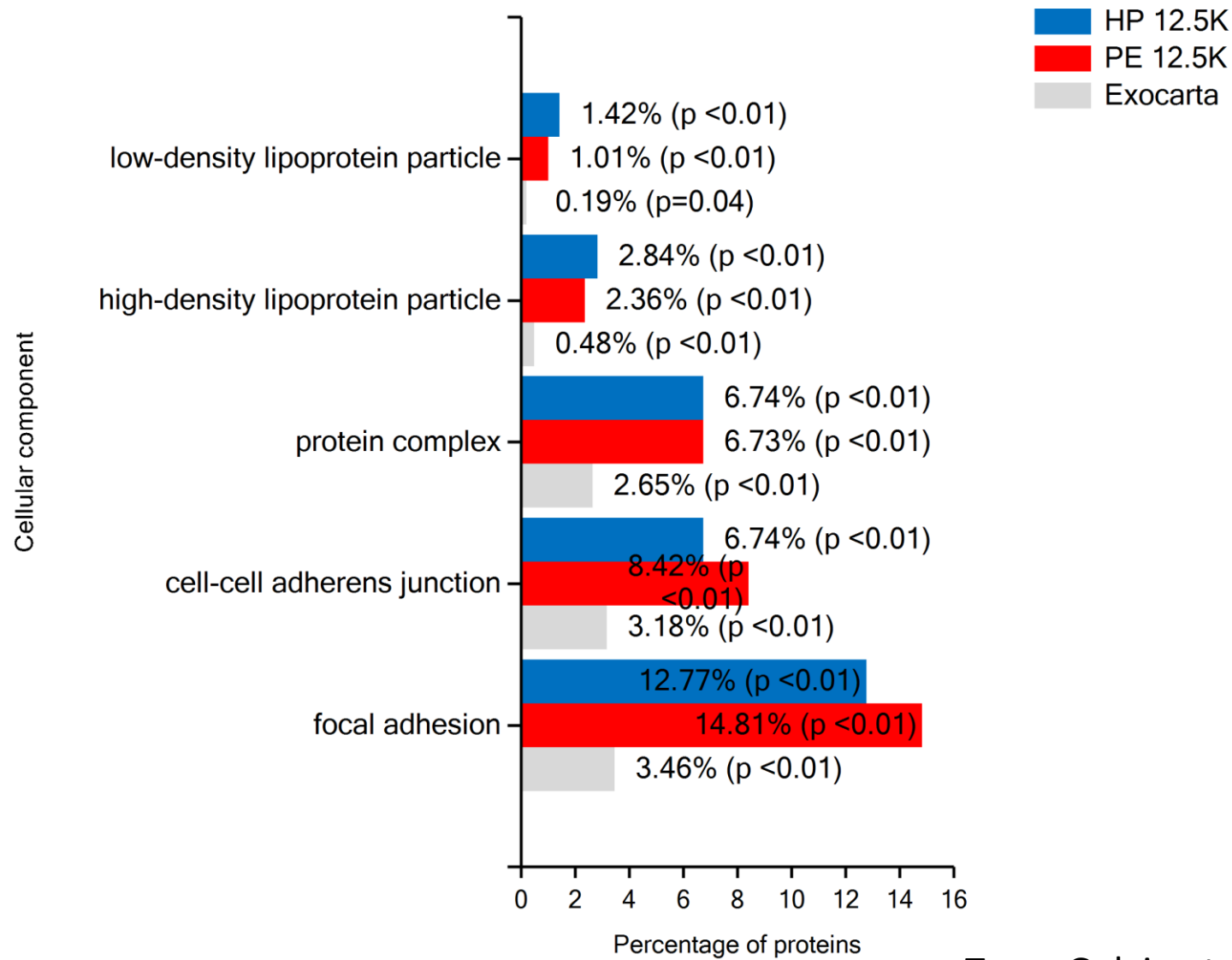
Pregnancy related: HLA-C, pregnancy zone protein, PZP, PSG6, PSG11, PSG4,

Lipid metabolism: APOB, APOC, APOA1, APOE,

Cellular component for HP 12.5K, PE 12.5K and Exocarta

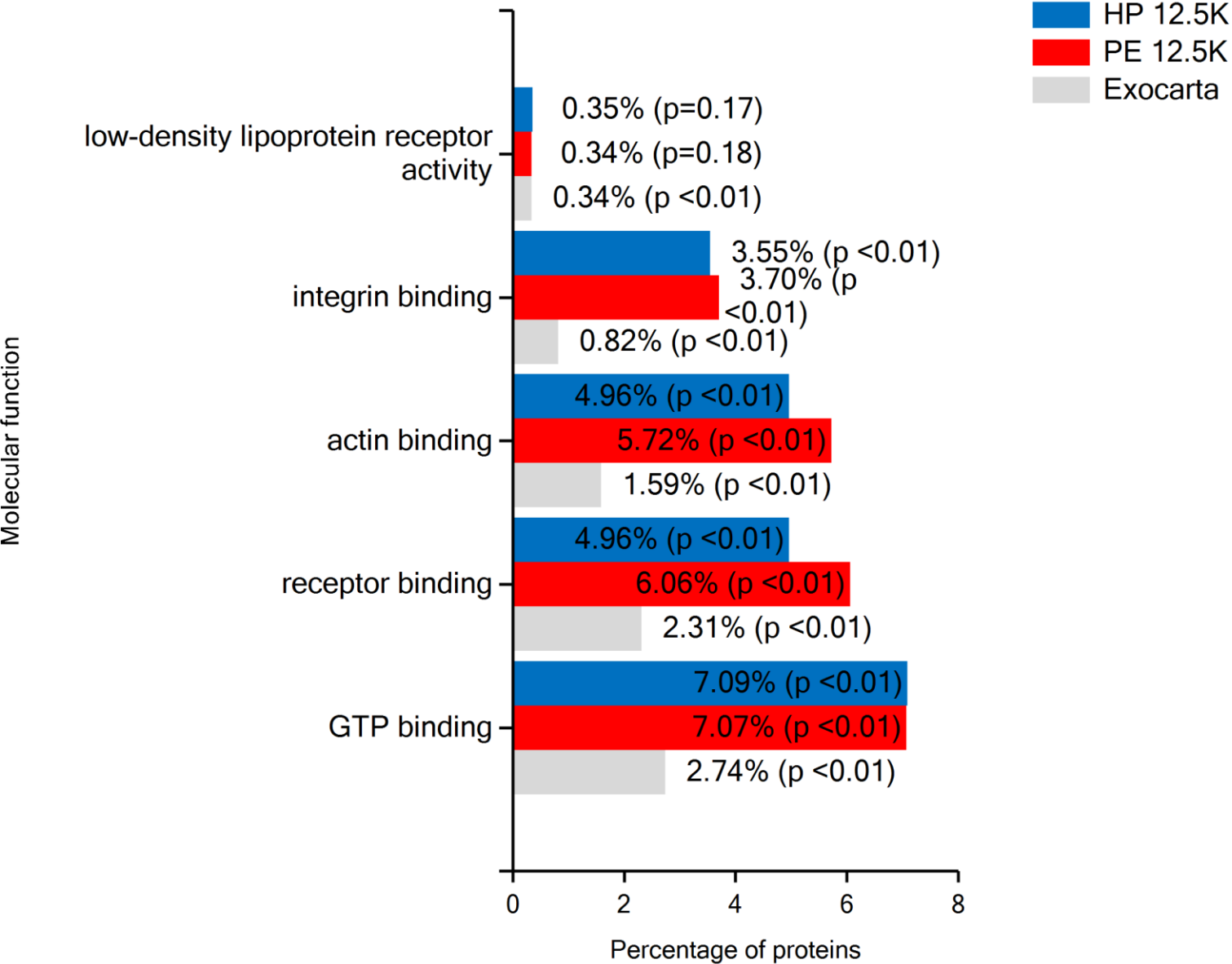


Cellular component for HP 12.5K, PE 12.5K and Exocarta

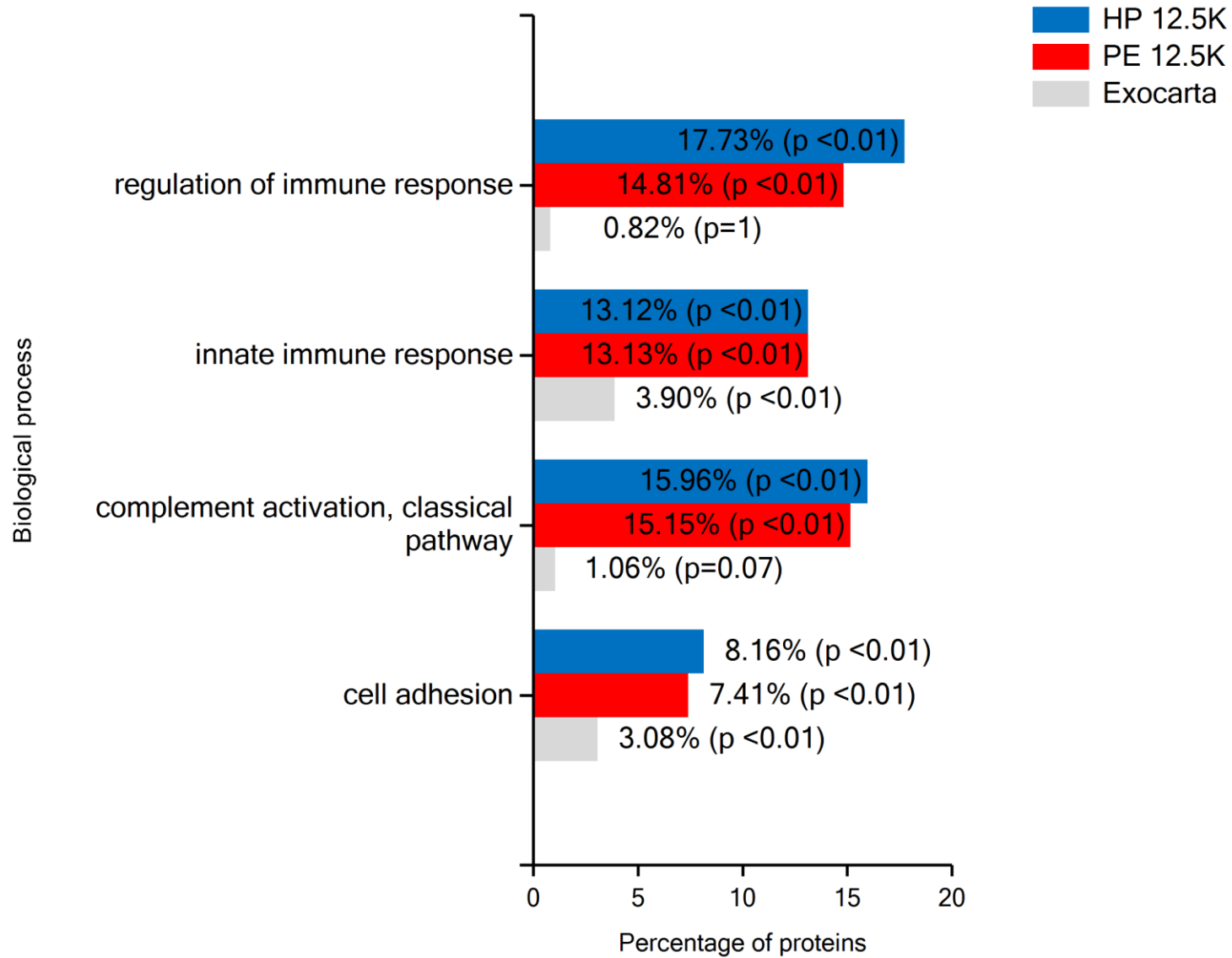


Trans-Golgi network: 0.71% HP, 1.01% PE, 1.4% Exocarta

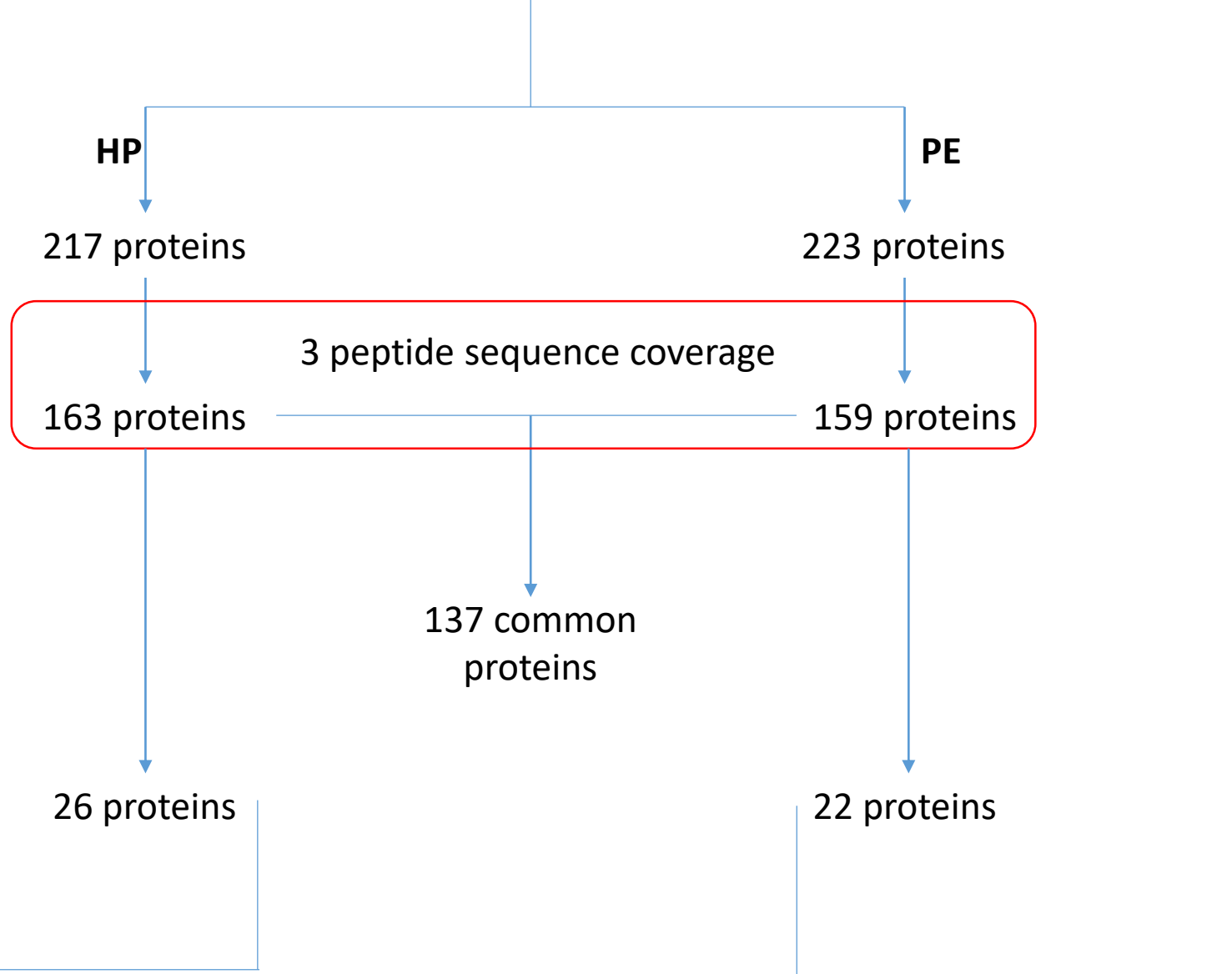
Molecular function for HP 12.5K, PE 12.5K and Exocarta



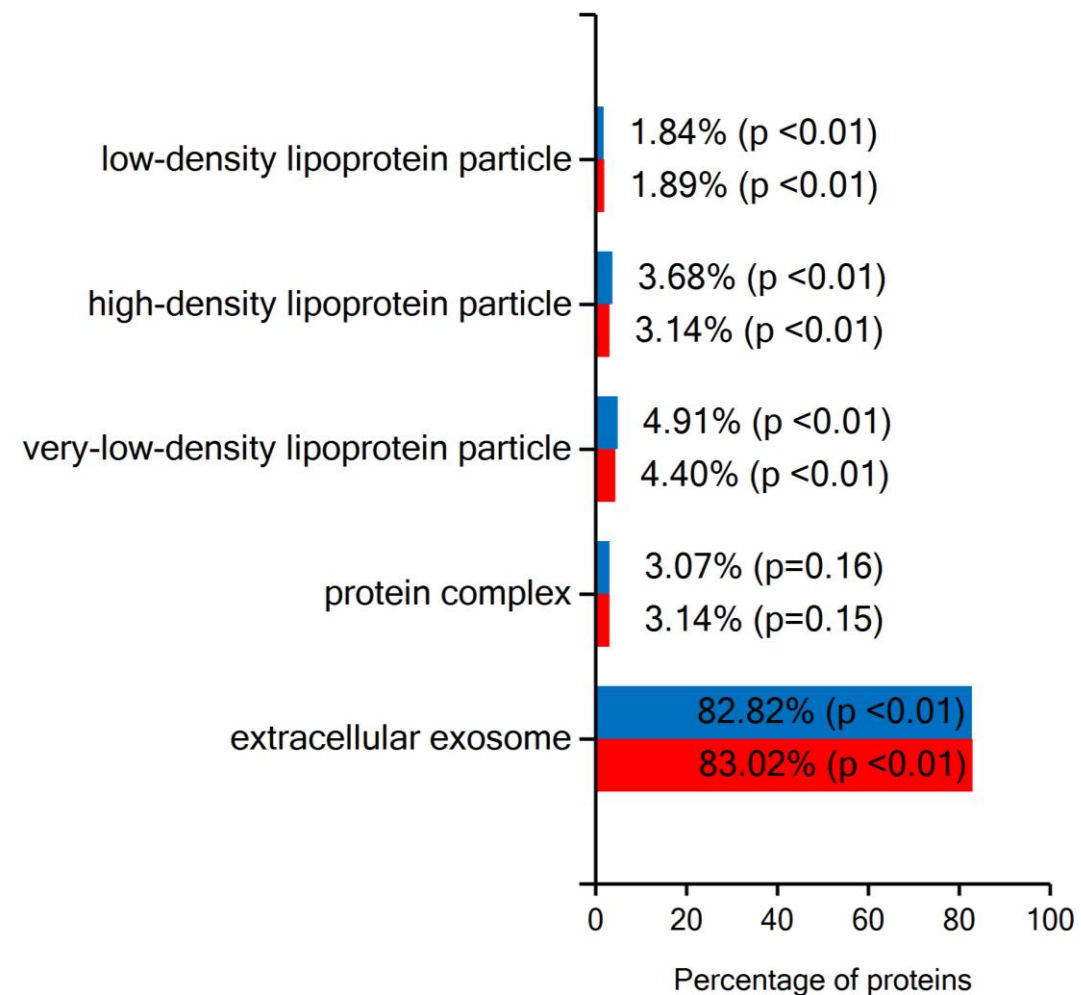
Biological process for HP 12.5K, PE 12.5K and Exocarta



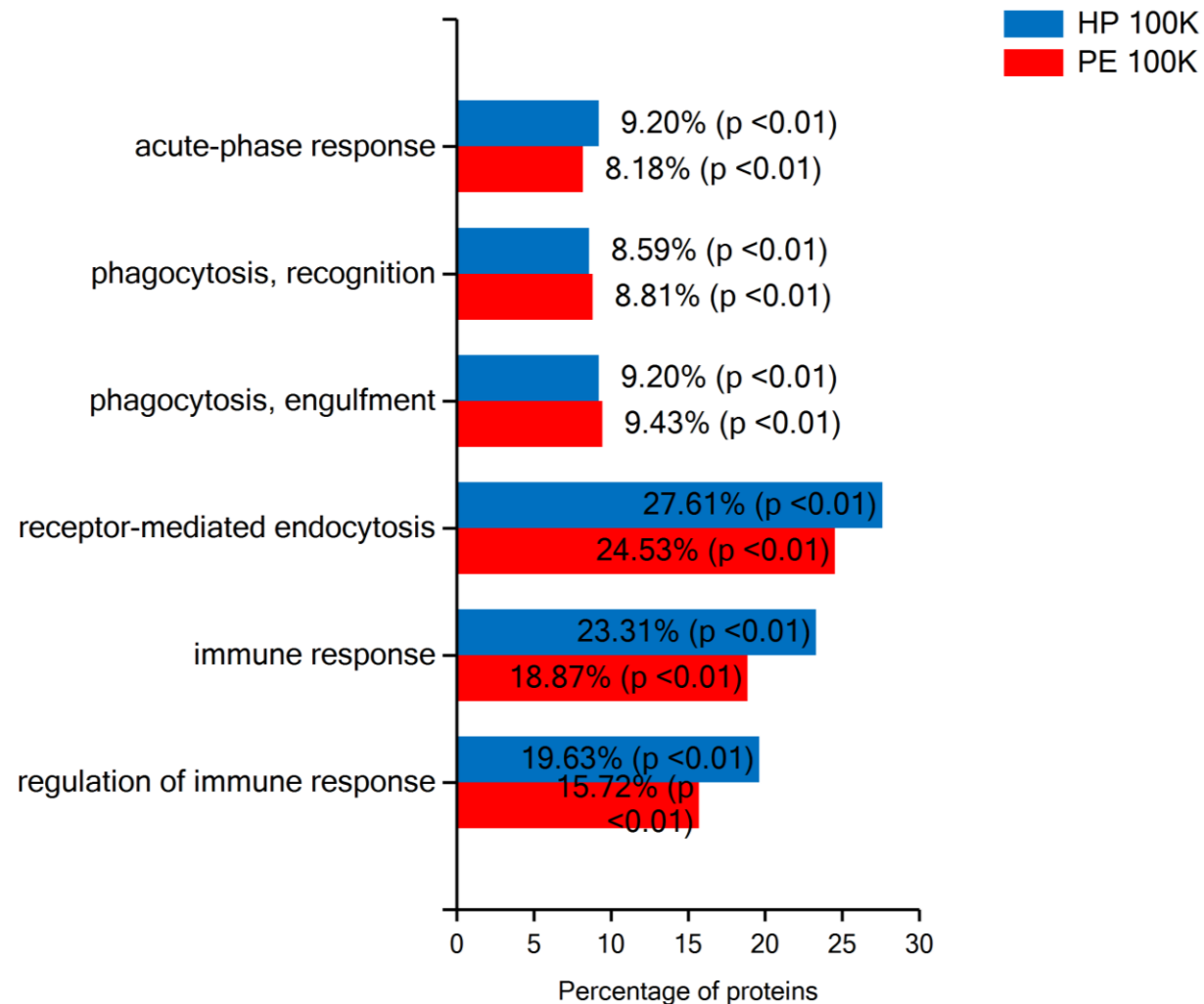
100K pellet (n=7 per group)



Cellular component for HP 100K and PE 100K



Biological process for HP 100K and PE 100K



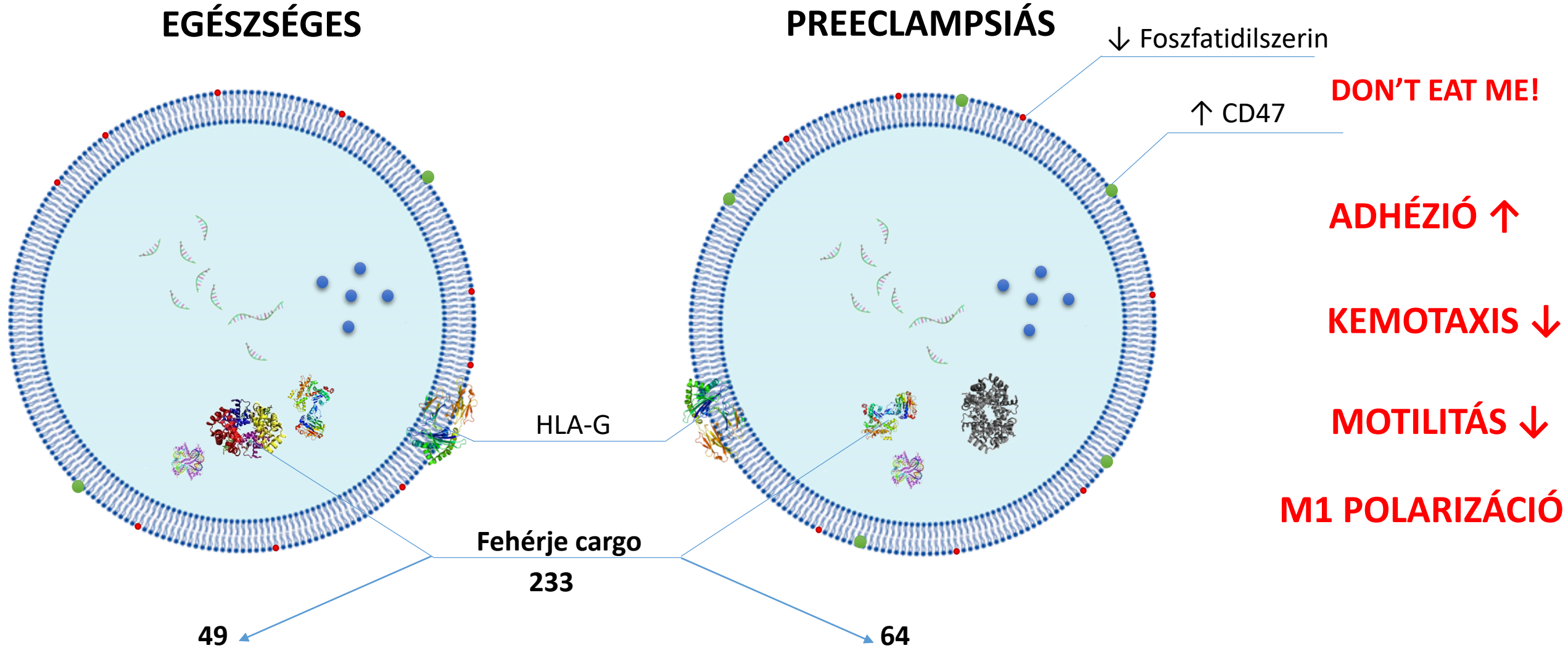
Clinical relevance

	Healthy 3 rd trimester pregnant (n=20)	Preeclamptic 3 rd trimester pregnant (n=25)
Maternal age (mean± SD)	33.8± 4.7 years	31.7± 4.6 years
Gestational age at sampling (mean± SD)	33.8± 3.5 weeks	32.0± 4.3 weeks
Gestational age at birth (mean± SD)	38.9± 1.2 weeks	32.5 ± 4.3 weeks
Birth weight (mean± SD)	3560± 416 g	1688± 922 g
AGA (%)	75%	38%
SGA (%)	5%	54%
LGA (%)	20%	8%
Early onset PE (%)	N.A	48 %
HELLP syndrome (%)	0 %	19 %
BMI (mean ± SD)	22.3± 3.1	26.3± 7.6
Systolic/diastolic blood pressure (mean ± SD)	115.8± 5.3 / 71.4± 3.1 mm Hg	155.0± 18.4 / 94.1± 13.9 mm Hg
Urine protein (mean ± SD)	N.D.	4063± 2329 mg/24 h

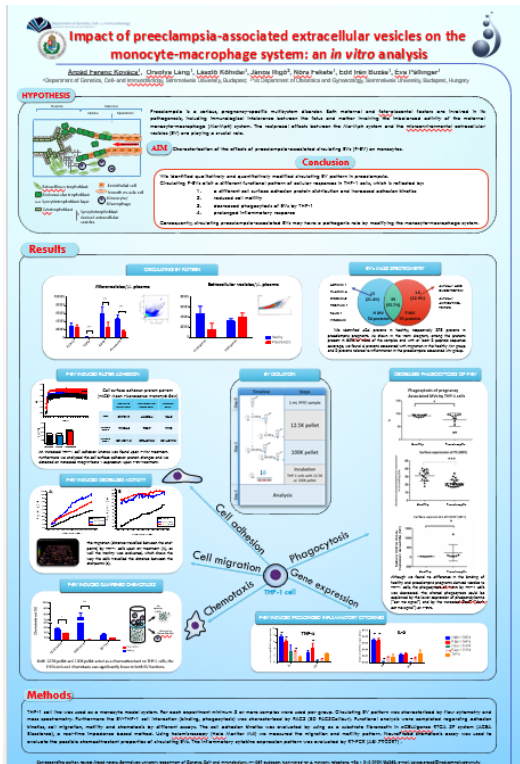
ÖSSZEFOGLALÁS

EGÉSZSÉGES

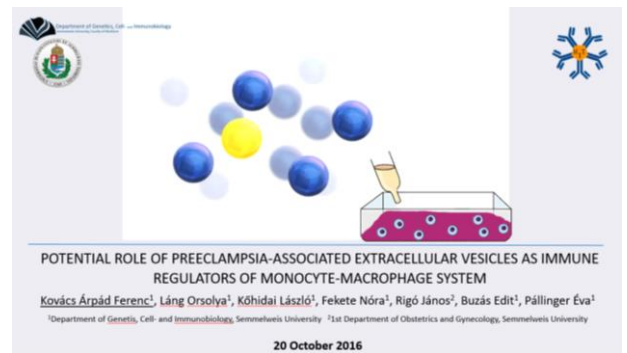
PREECLAMPSIÁS



Következő lépés: miR cargo (November-December 2016)



ISEV, Rotterdam



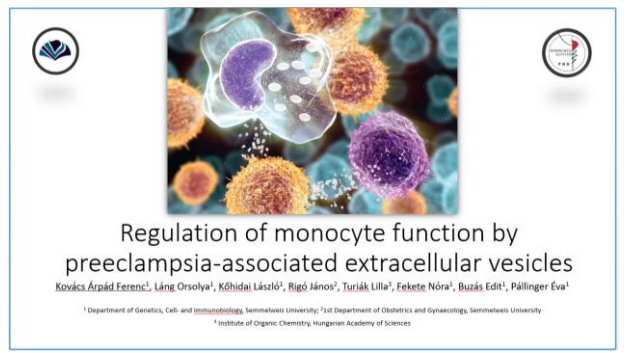
MIT, Velence



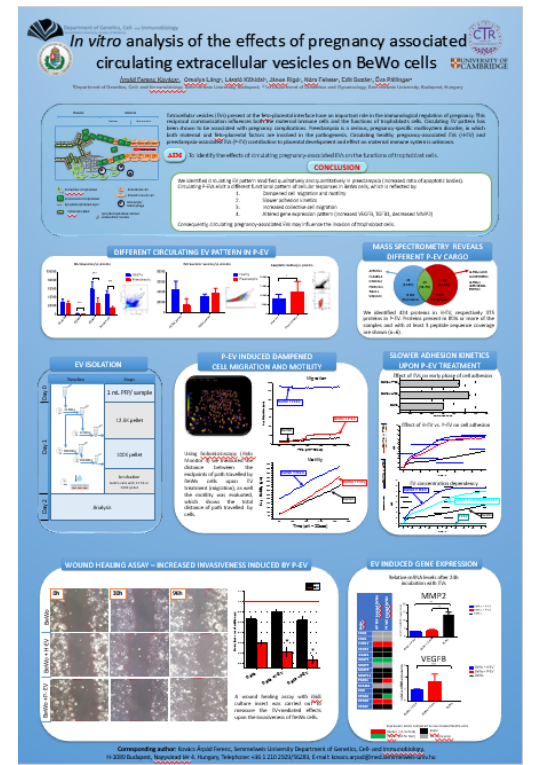
MAGE, Szeged



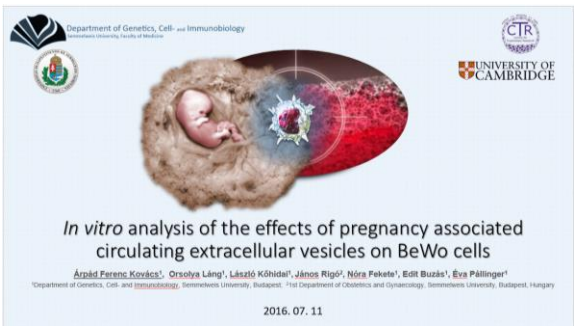
MHGT, Pécs



PhD Konferencia, Budapest



CTR, Cambridge





KÖSZÖNÖM SZÉPEN A FIGYELMET!