Electronic Supplementary Material (ESI) for Nanoscale Advances.

Supplementary Material

Cytotoxicity of mini gold nanorods: intersection with extracellular vesicles

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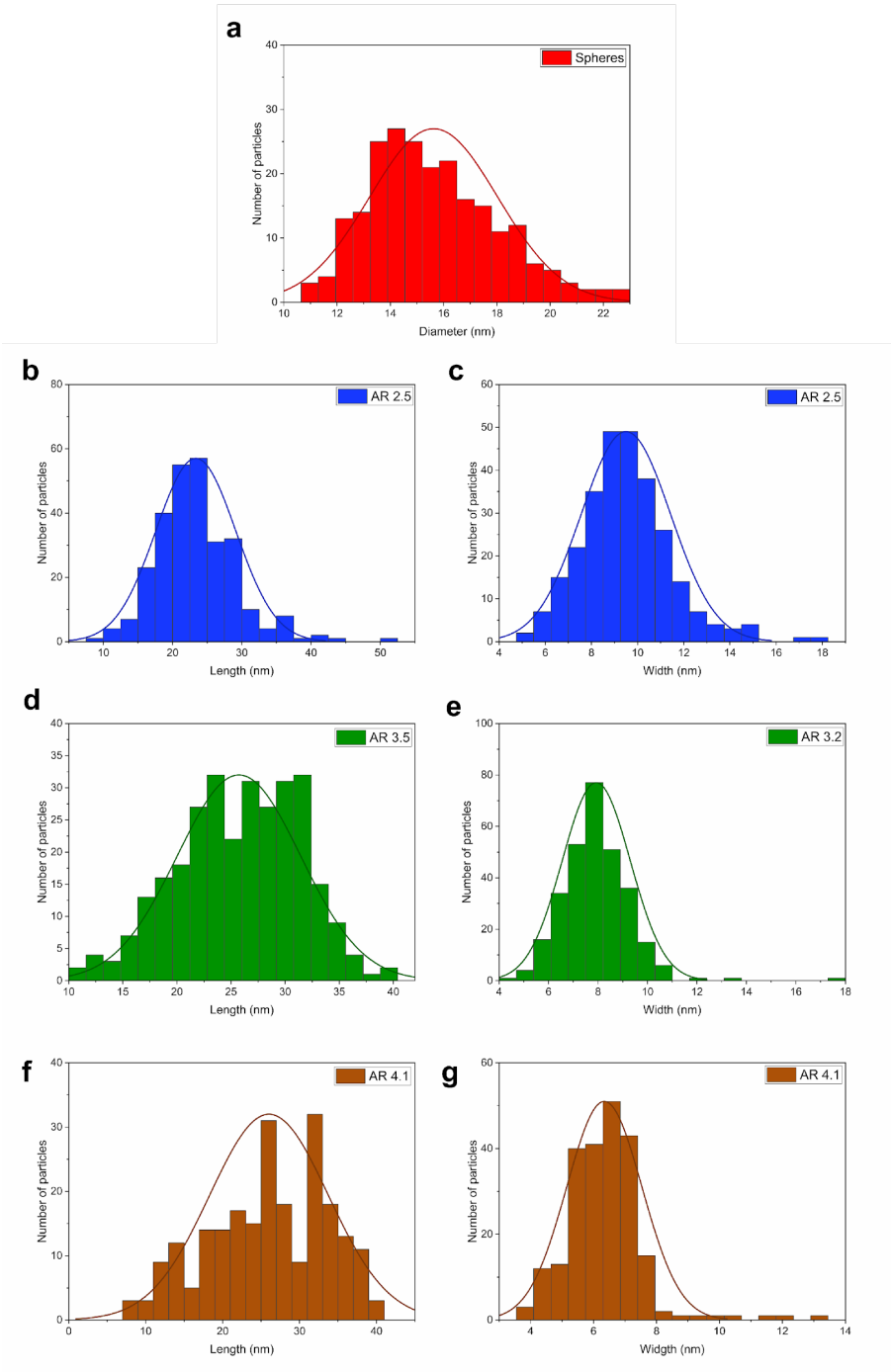


Figure S1. Histograms showing the particle size distribution pattern for the (a) spheres and mini gold nanorods: (b,c) AR 2.5; (d,e) AR 3.5; and (f,g) 4.1.

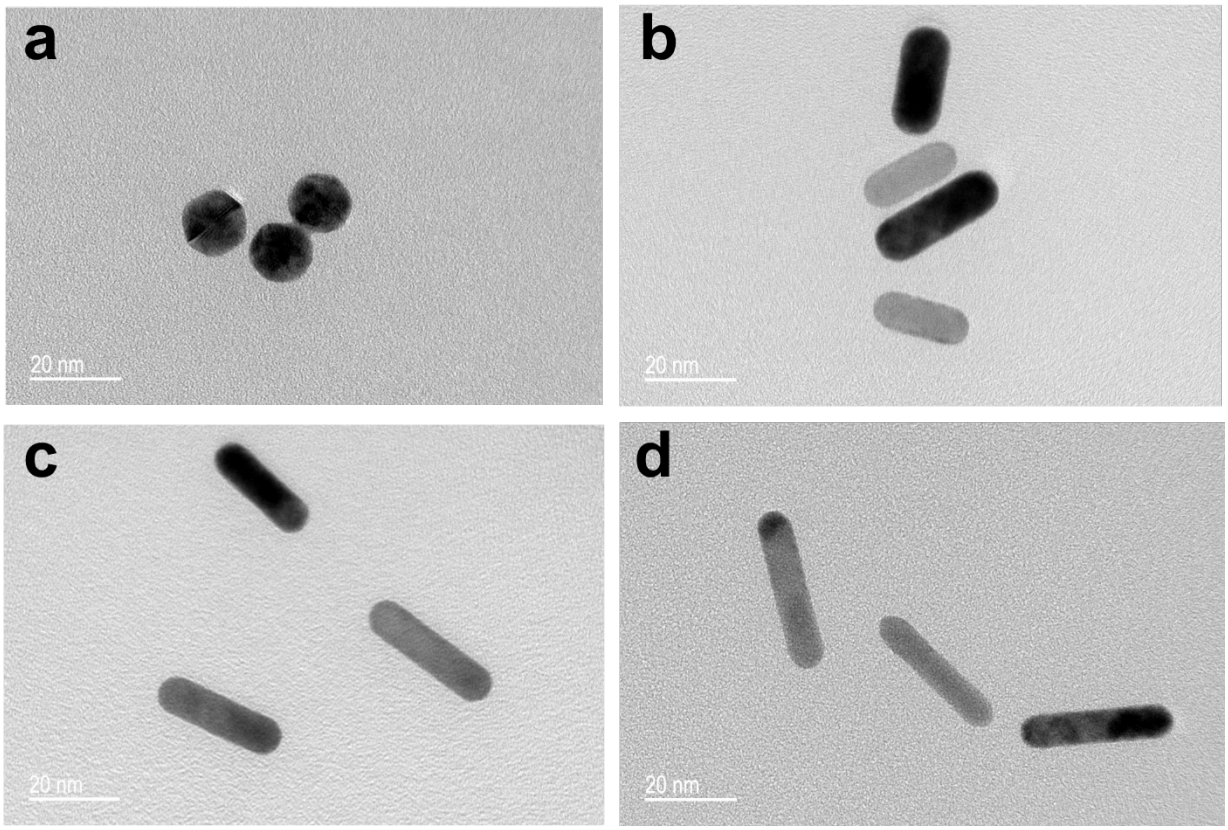
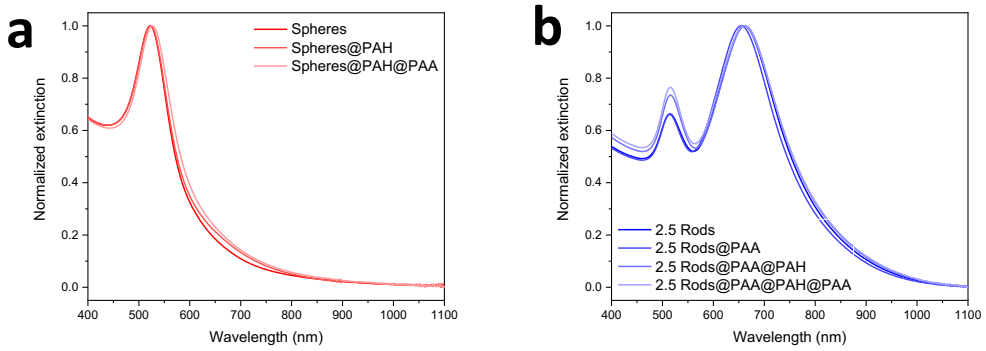


Figure S2. TEM images of (a) spheres and mini AuNRs of different aspect ratios: (b) 2.5, (c) 3.2 and (d) 4.1. Magnification 200kx, scale bars = 20 nm.



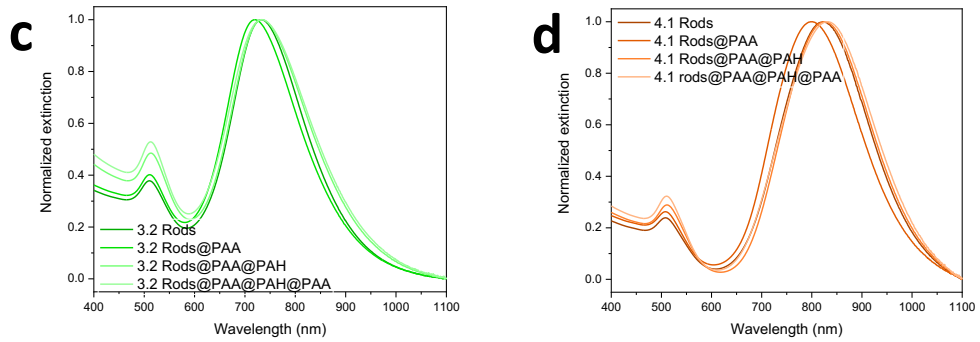


Figure S3. UV-vis-NIR spectra of each step of the coating procedure gold nanoparticles: (a) Spheres and mini AuNRs (b) 2.5, (c) 3.2 and (d) 4.1.

Table S1. Characterization of each step of the coating procedure of gold nanoparticles.

Particle	**Coating**	**SPR (nm)[a]**	**Hydrodynamic diameter (nm)**	**Zeta Potential (mV)**
Spheres	Citrate	522	45.1 ± 2.2	-28.3 ± 2.1
	@PAH	523	58.7 ± 1.0	+47.6 ± 2.3
	@PAA	526	67.5 ± 1.8	-47.0 ± 0.2
2.5 Rods	CTAB	660	30.8 ± 0.3	+38.3 ± 1.6
	@PAA	655	42.7 ± 0.5	-54.8 ± 0.8
	@PAH	662	66.4 ± 1.5	+37.9 ± 1.2
	@PAA	661	64.7 ± 0.3	-28.7 ± 1.8
3.2 Rods	CTAB	733	35.4 ± 0.3	+36.6 ± 0.7
	@PAA	719	58.7 ± 1.1	-51.0 ± 0.5
	@PAH	734	125.4 ± 0.4	+53.3 ± 5.6
	@PAA	734	119.6 ± 3.3	-35.9 ± 3.3
4.1 Rods	CTAB	821	43.8 ± 1.1	+31.4 ± 2.2
	@PAA	799	70.7 ± 3.6	-51.9 ± 1.2
	@PAH	826	109.5 ± 2.7	+35.9 ± 2.8
	@PAA	833	104.3 ± 0.2	-42.4 ± 3.0

[a] Longitudinal surface plasmon resonance (LSPR) for rods.

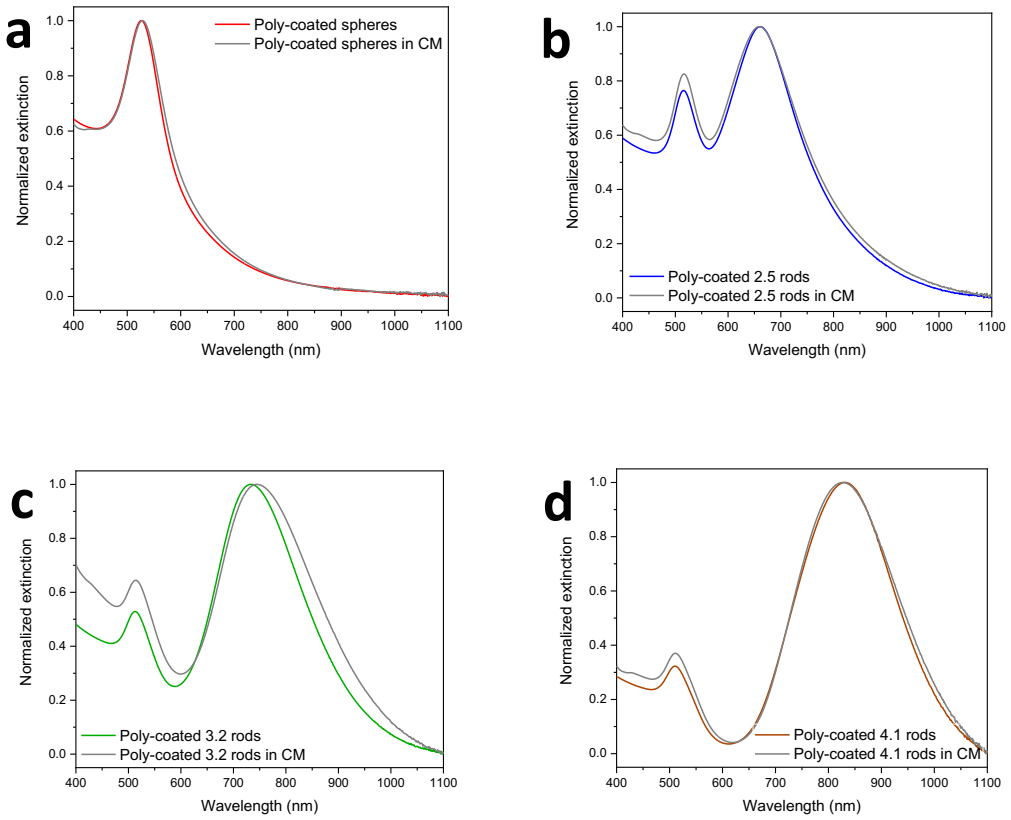


Figure S4. UV-vis-NIR spectra of gold nanoparticles in water and cell culture media: (a) Spheres and mini AuNRs (b) 2.5, (c) 3.2 and (d) 4.1.

Table S2. Characterization of the gold nanoparticle in water and cell culture media.

Particle	**Solvent**	**SPR (nm)[a]**	**Hydrodynamic diameter (nm)**	**Zeta Potential (mV)**
Poly-coated spheres	Water	526	67.5 ± 1.8	-47.0 ± 0.2
	Cell Medium	529	96.2 ± 1.5	-17.4 ± 1.4
Poly-coated 2.5 rods	Water	661	64.6 ± 0.3	-28.7 ± 1.8
	Cell Medium	660	111.3 ± 1.0	-18.26 ± 1.0
Poly-coated 3.2 rods	Water	734	119.6 ± 3.3	-35.9 ± 3.3
	Cell Medium	745	214.6 ± 6.5	-18.4 ± 0
Poly-coated 4.1 rods	Water	833	104.3 ± 0.2	-42.4 ± 3.0
	Cell Medium	828	155.6 ± 2.5	-20.8 ± 1.8

[a] Longitudinal surface plasmon resonance (LSPR) for rods

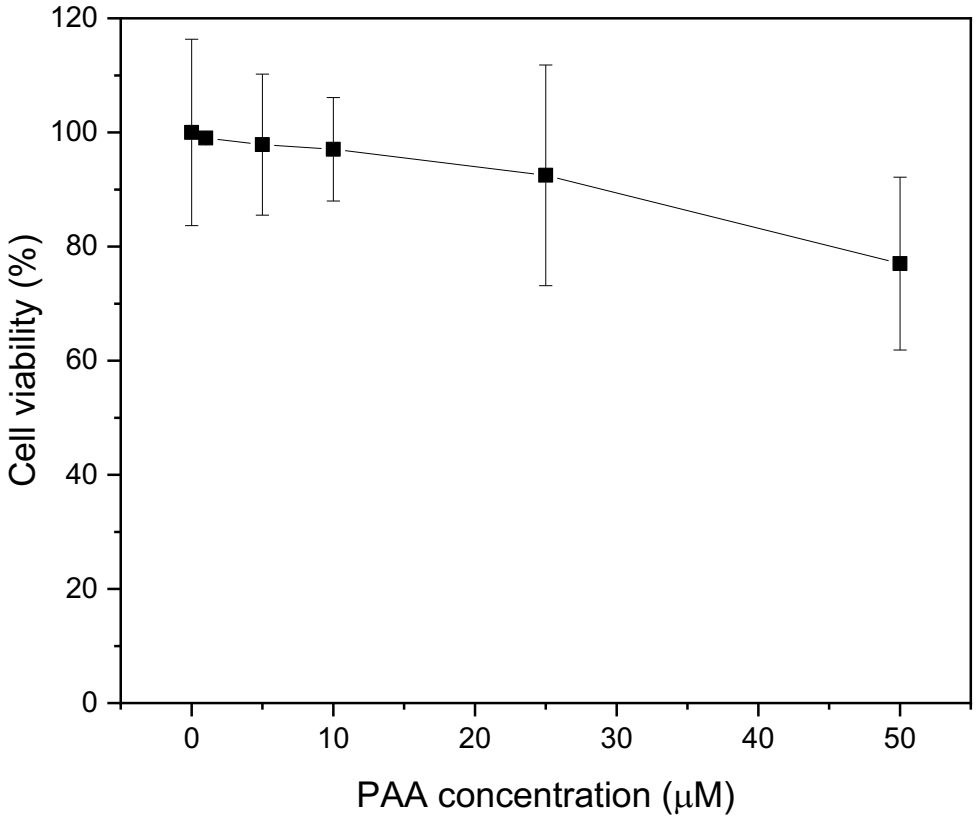


Figure S5. Cell viability assay for HDF cells exposed to PAA after 72 h Data were analyzed by one-way analysis of variance (ANOVA) followed by the Dunnett post hoc test, and $p < 0.05$ was considered statistically significant, $n = 3$.

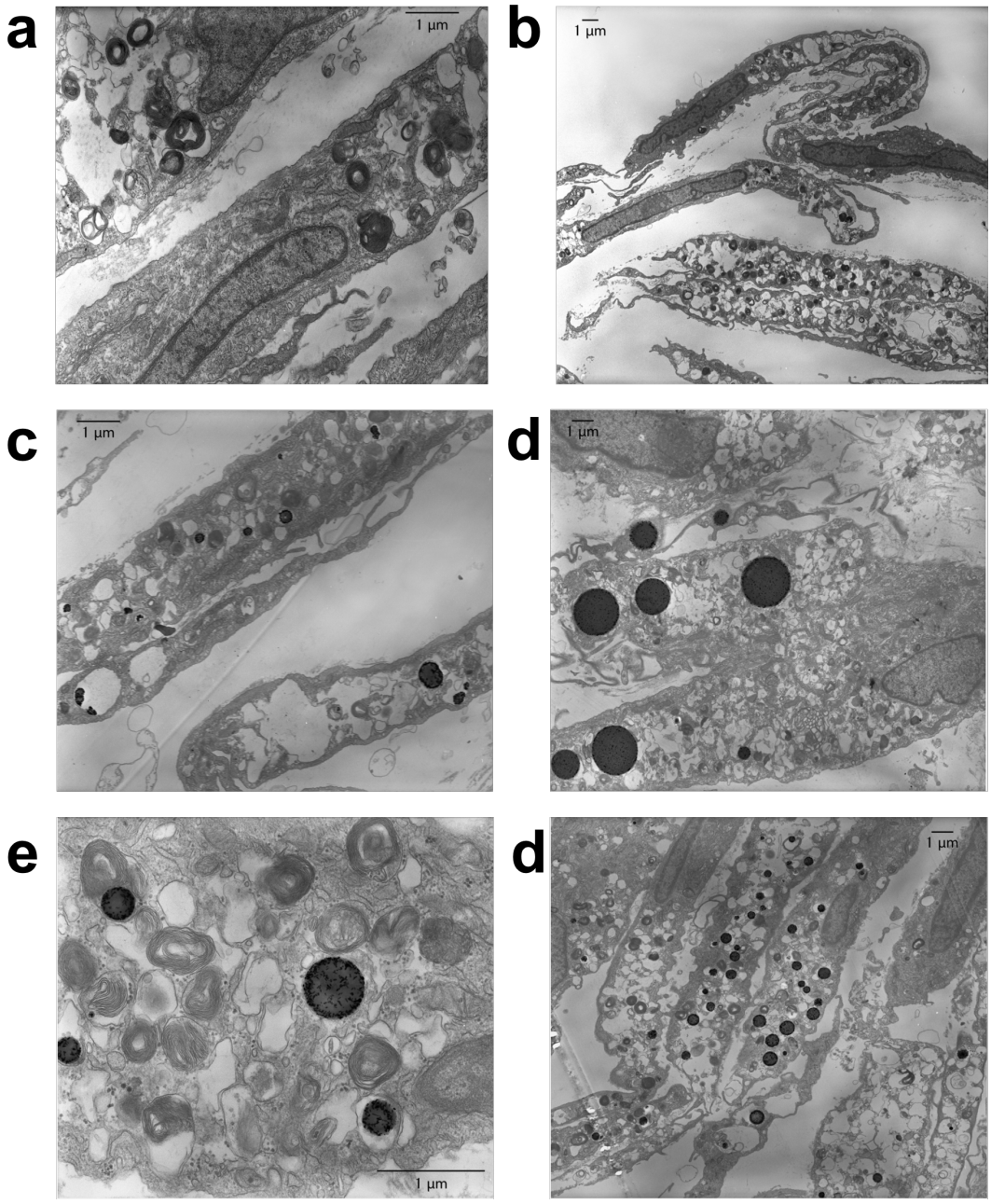


Figure S6. TEM images of HDF cells in the absence (a,b) and in presence of (c) spheres, and different aspect ratio mini gold nanorods 1nM: (d) 2.5 rods, (e) 3.2 rods, and (f) 4.1 rod.

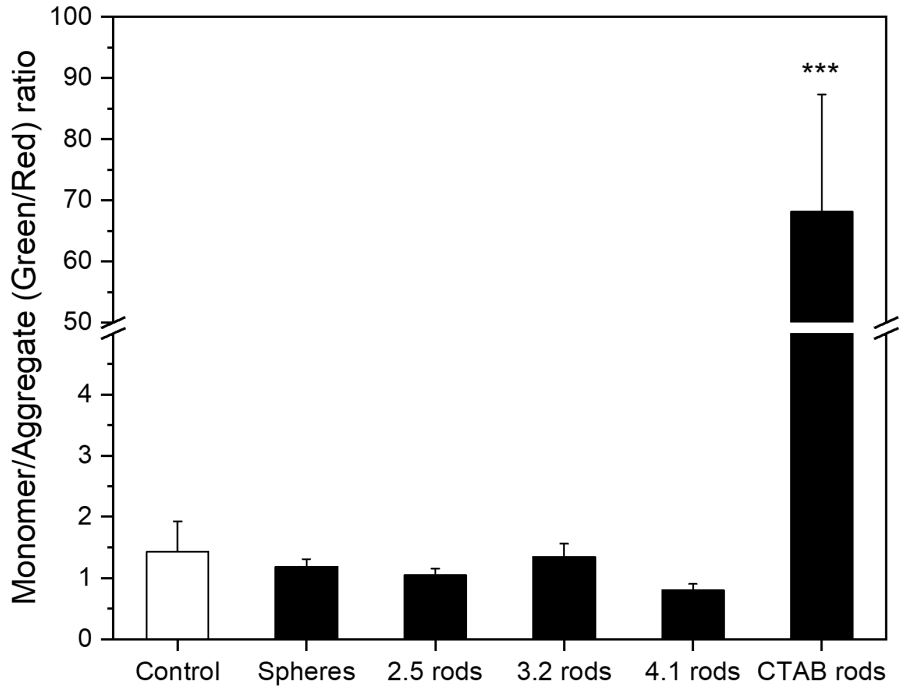


Figure S7. The effect of different polyelectrolyte-coated gold nanoparticles in the mitochondrial membrane potential (MMP) of HDF cells measured by JC1 dye. Quantification data of MMP in HDF cells. Data are means of three independent experiments. CTAB-rods (AR 2.5) were used to show the effect of non-coated AuNRs.

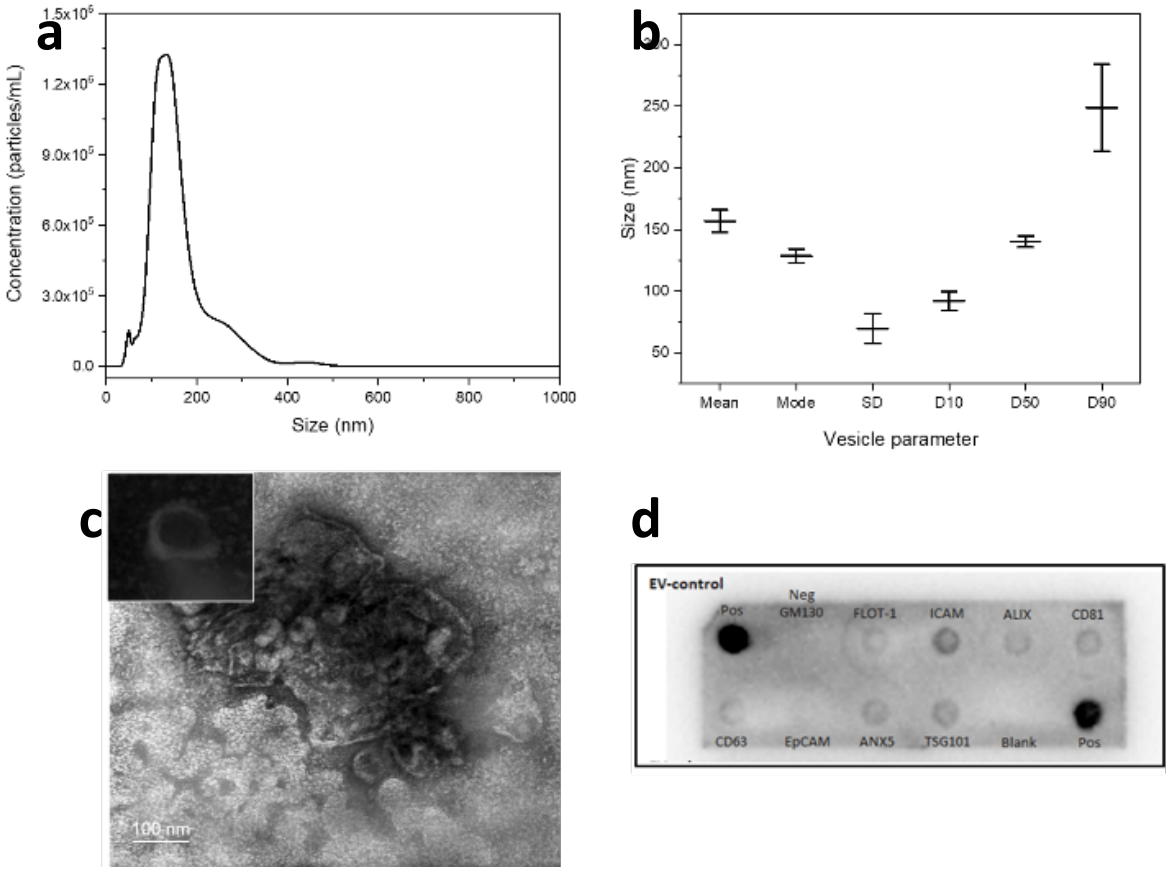


Figure S8. Extracellular Vesicles characterization. Nanoparticle tracking analysis showing size distribution of EVs (a) and (b) size characteristics: mean, mode, standard deviation, and different size distribution (D10, D50 and D90), n=3. Transmission electron micrographs of negative stained EVs (c) and EVs biomarkers verification (d).

Table S3. Sample absorbance at 260 and 280 nm

Sample	A260	A280	260/280
Control	2.20 ± 0.18	3.20 ± 0.33	0.68
Spheres	2.91 ± 0.91	4.14 ± 1.47	0.70
2.5 rods	2.65 ± 0.56	3.72 ± 0.97	0.71
3.2 rods	2.19 ± 0.33	3.04 ± 0.47	0.72
4.1 rods	2.19 ± 0.37	3.55 ± 1.27	0.61

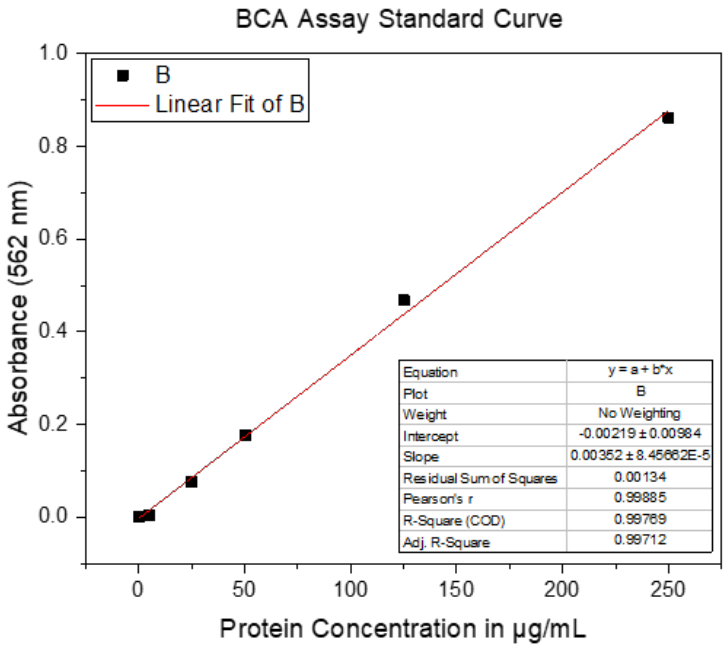


Figure S9. BCA assay standard curve (BSA 5 – 250 µg/mL)