

Electronic Supporting Information

Optimization and characterization of miRNA-129-5p-encapsulated poly (lactic-co-glycolic acid) nanoparticle to reprogram activated microglia

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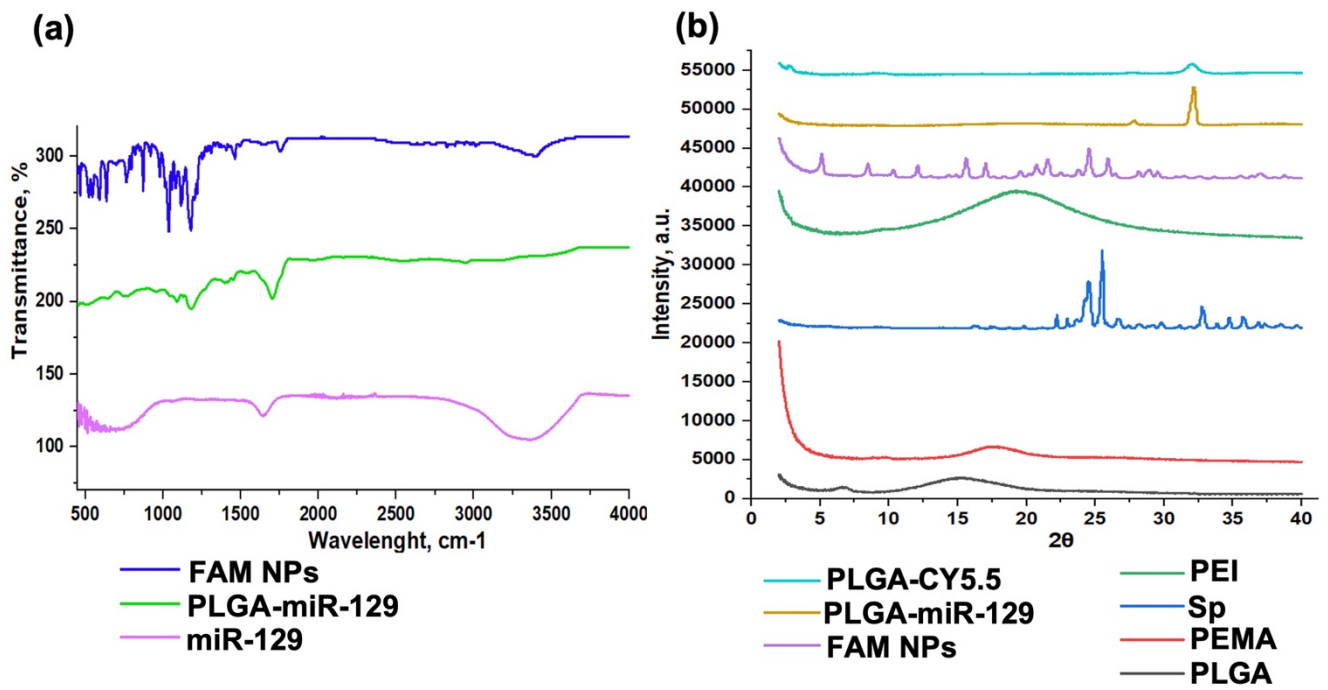


Fig S1. Nanoformulation characterization by FTIR **(a)** and X-ray diffraction patterns **(b)**.

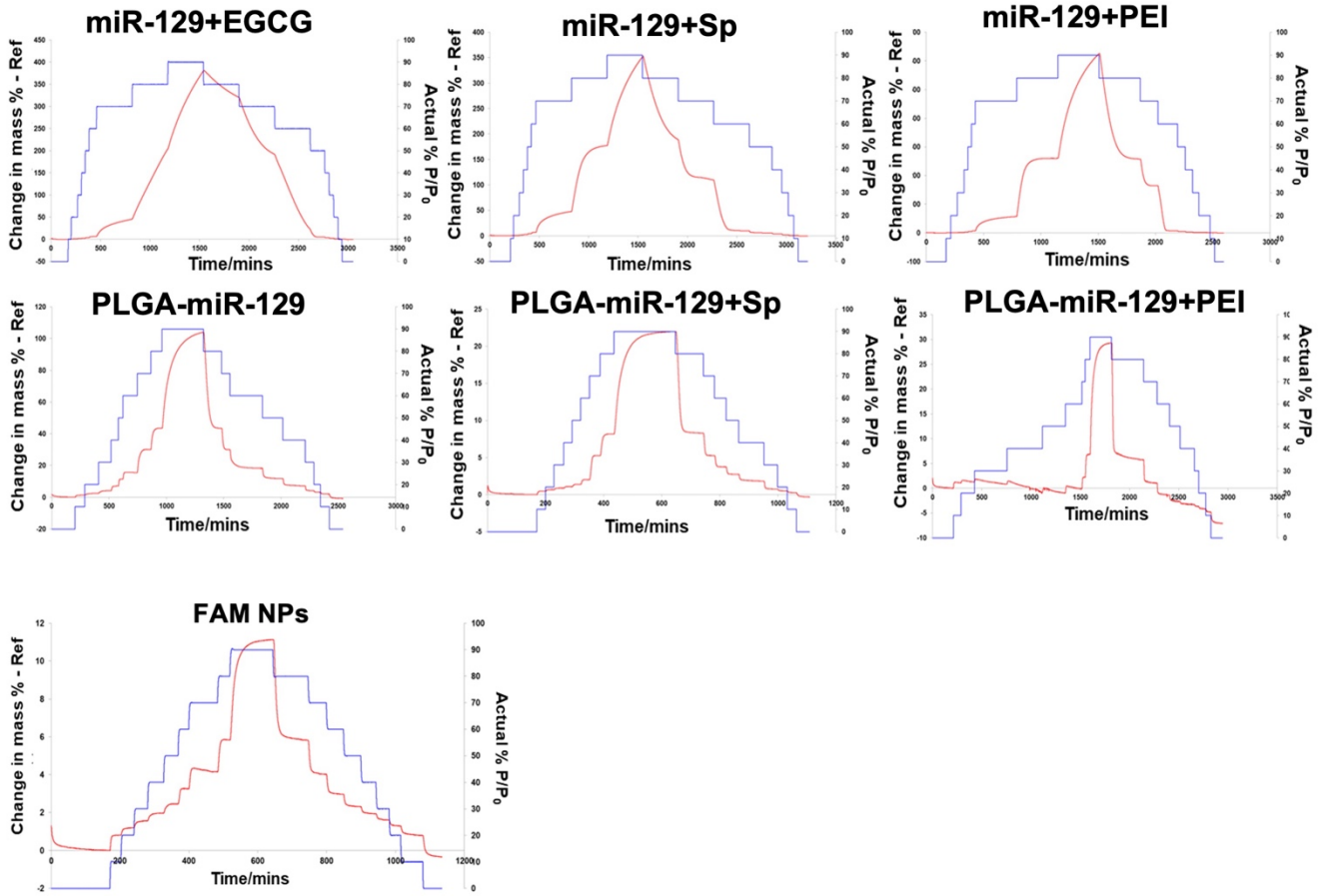
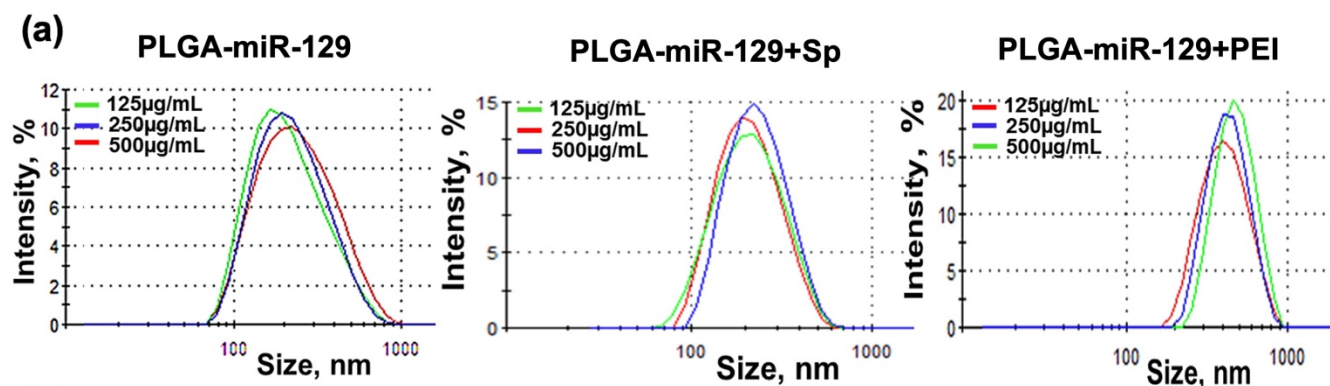


Fig S2. Dynamic vapor sorption analysis results. Water adsorption/desorption kinetics (blue line: %P/P₀, red line: mass change).



(b)

NPs name	miRNA loading, µg	Size, nm	Pdl	Zeta potential, mV
PLGA-miR-129	125	309.3±8.5	0.23±0.03	-28.6±0.3
	250	315.2±9.2	0.27±0.02	-28.8±0.5
	500	330.9±11.1	0.26±0.07	-29.2±1.5
PLGA-miR-129+Sp	125	364.7±4.2	0.18±0.04	-22.7±1.1
	250	375.2±9.7	0.15±0.01	-26.1±0.8
	500	393.6±10.5	0.18±0.01	-26.0±1.6
PLGA-miR-129+PEI	125	435.3±11.4	0.22±0.03	-24.8±1.2
	250	468.1±13.1	0.26±0.03	-25.2±0.9
	500	471.4±15.0	0.29±0.04	-26.6±1.4

Fig S3. Characterization of miR-129 nanoparticles obtained at different loading amounts of miR-129 (0.01-0.5µg per 1mg of NPs). **(a)** Representative average size based on the intensity (DLS) for the most promising miR-129-NPs formulations; **(b)** Summarized data obtained by DLS analysis from **(a)** (n=3).

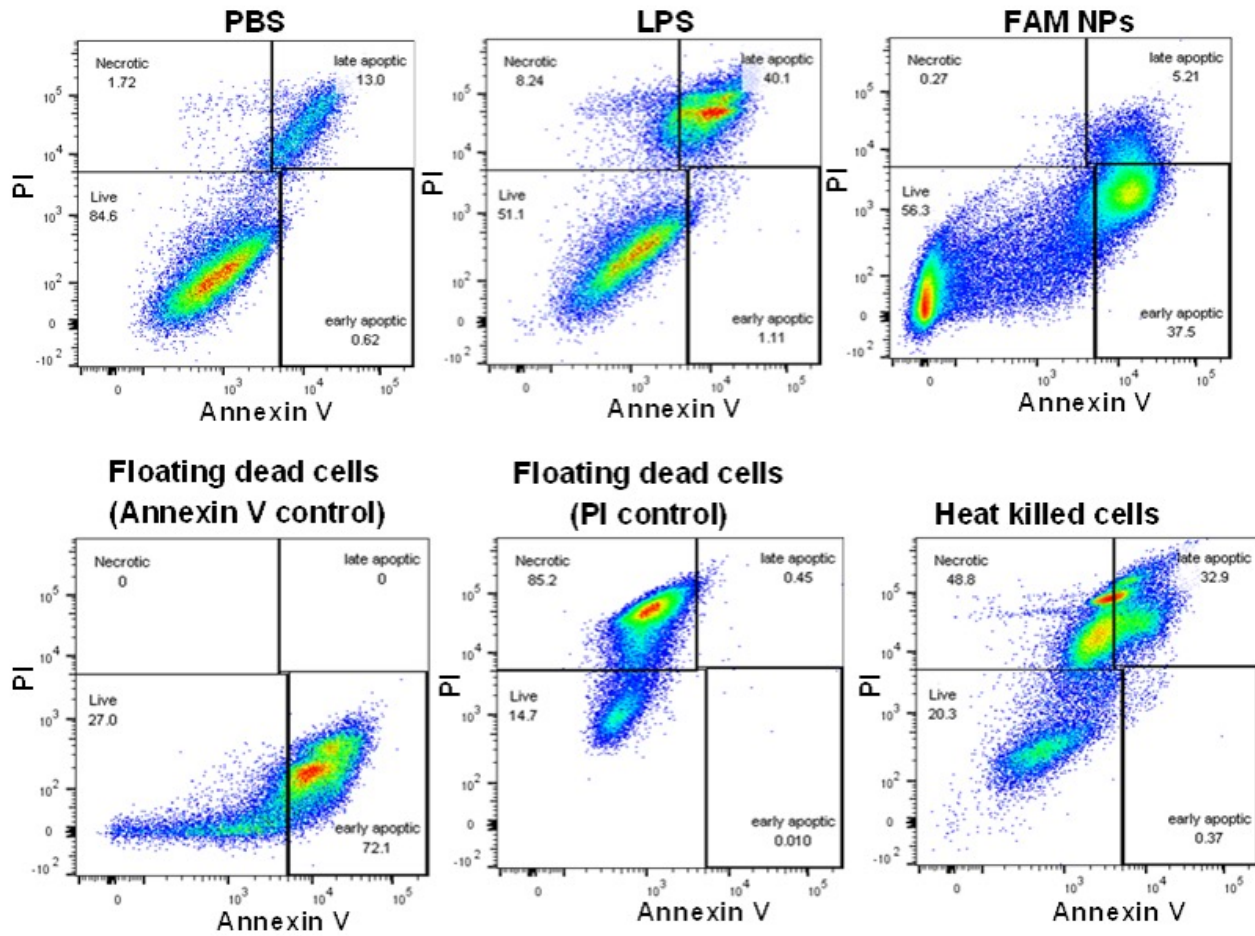


Fig S4. Flow cytometry data: controls for Annexin V/PI assay.

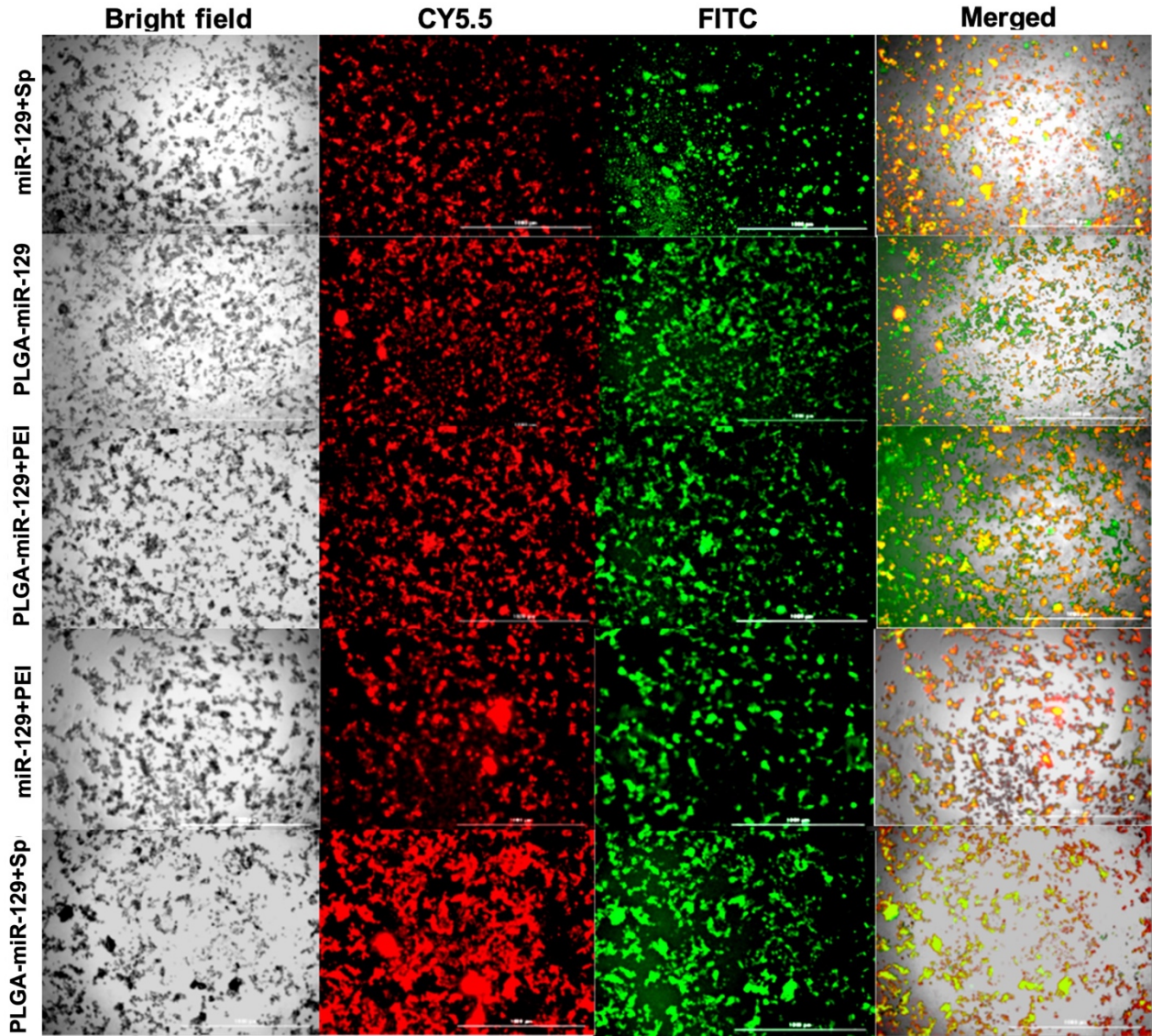


Fig S5. NPs uptake by BV-2 cells. BV-2 cells pretreated with LPS and then exposed to the NPs for 72h. Microphotographs of the cells were taken after 2x washes with PBS by Cytation 7 plate reader using bright field and filters for CY5.5 (for PLGA NPs) and FAM (for miR). Scale bar = 1 mm.

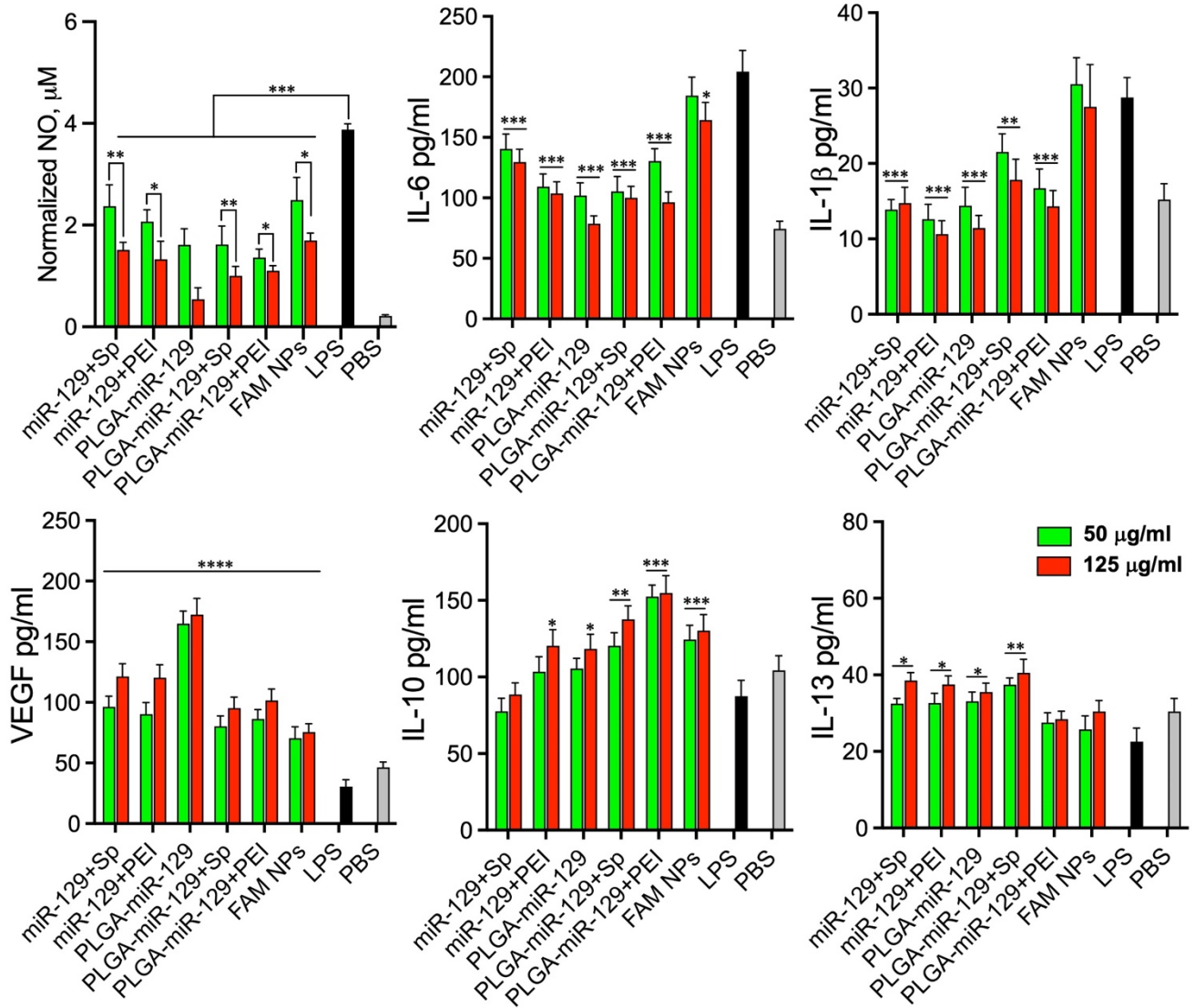


Fig S6. The immunomodulatory effects of miR-129 loaded nanoformulations were concentration-dependent. The expression levels of NO, and pro- and anti-inflammatory factors in response to nanoformulations were significantly altered compared to LPS group. N=5/group, error bars = SD, p-value: *p < 0.05, **p < 0.01, ***P < 0.001 and ****P < 0.0001 vs. LPS group.

NPs name	Size_{zs}, nm	Size_{SEM}, nm	Pdl	Zeta potential, mV
FAM NPs	330.5±8.4	270.6±28.8	0.27±0.00	-22.2±1.4
miR-129+EGCG	356.4±9.2	378.6±80.1	0.46±0.02	-28.5±0.4
miR-129+Sp	346.6±21.9	367.4±31.2	0.20±0.02	-23.8±0.9
miR-129+PEI	321.8±30.5	312.7±23.6	0.23±0.01	-27.1±0.5
PLGA-miR-129	310.8±6.9	258.8±41.2	0.16±0.01	-26.5±2.8
PLGA-miR-129+Sp	407.9±17.9	400.8±64.5	0.48±0.18	-21.7±0.6
PLGA-miR-129+PEI	401.1±25.9	327.2±71.3	0.31±0.04	-24.9±0.8

Table S1. Physicochemical properties of each of the nanoformulations by ZetaSizer and SEM.