Supporting Information

Esterase-activated celastrol delivery system suppressed steatosis-related hepatocellular carcinoma progression

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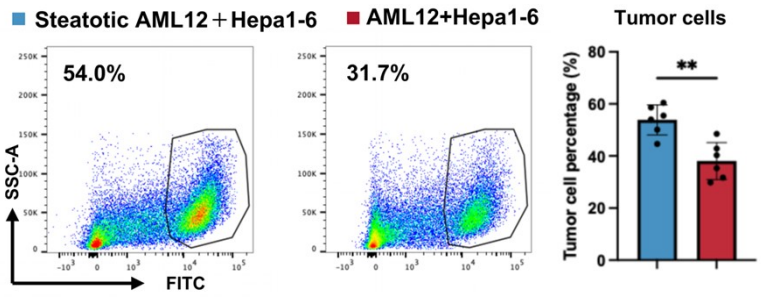

Figure S1. Flow cytometry profiles and comparison of quantification of Hepa 1-6-GFP tumor cells in steatotic and conventional tumors. Data are presented as mean ± SD of biological replicates (n = 6). (****p < 0. 0001; ***P <0.001; ** P <0.01; * P <0.05; n.s. no significance)

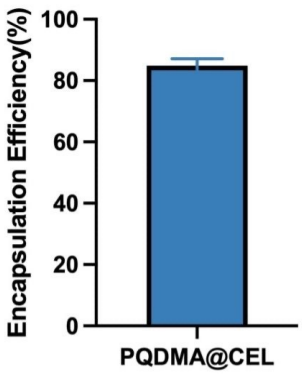

Figure S2. Encapsulation efficiency of the optimized PQDMA@CEL NPs.

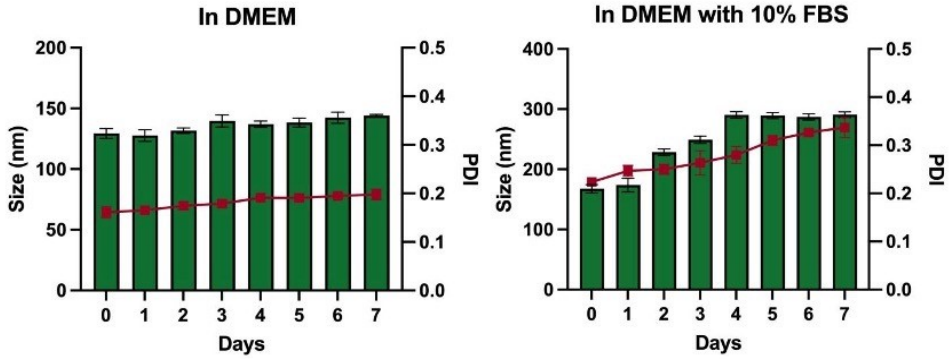

Figure S3. Stabilities of PQDMA@CEL NPs in DMEM with or without 10% FBS.

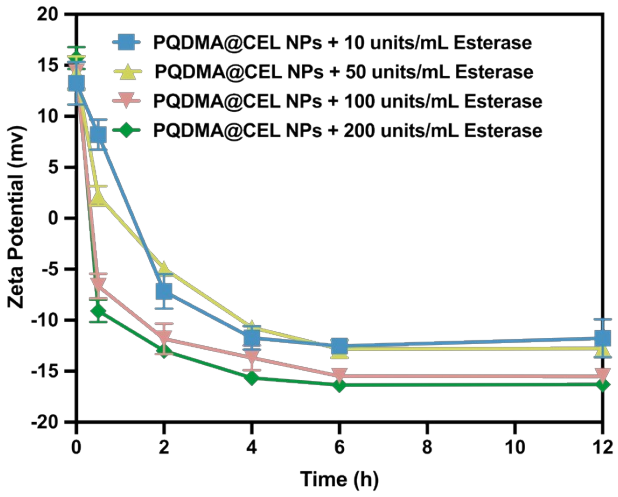

Figure S4. Zeta potential measurement of PQDMA@CEL NPs in the presence of different esterase conditions.

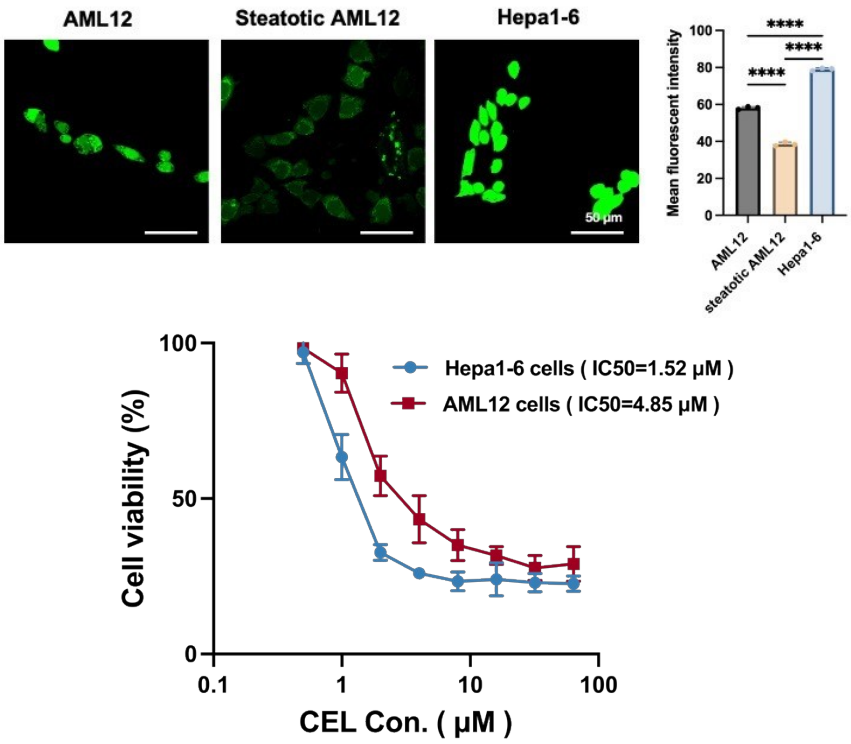

Figure S5. Esterase expression levels of AML12 cells, steatotic AML12 cells, and Hepa1-6 cells based on fluorescein diacetate method (esterase: 5 μg/mL) and cytotoxicities of PQDMA@CEL NPs on Hepa1-6 cells and AML12 cells of 24h incubation. Scale bar: 50 μm. Data are presented as mean ± SD of biological replicates (n = 3). (****p < 0. 0001; ***P <0.001; ** P <0.01; * P <0.05; n.s. no significance)

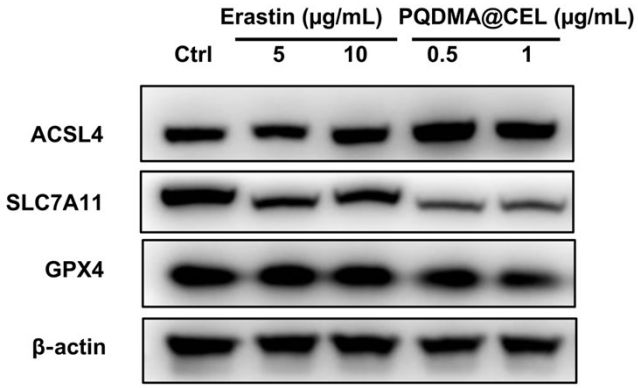

Figure S6. Western blot analysis of ACSL4, SLC7A11, and GPX4 protein expression in Hepa1-6 cells treated with PQDMA@CEL NPs, with erastin used as a positive control.

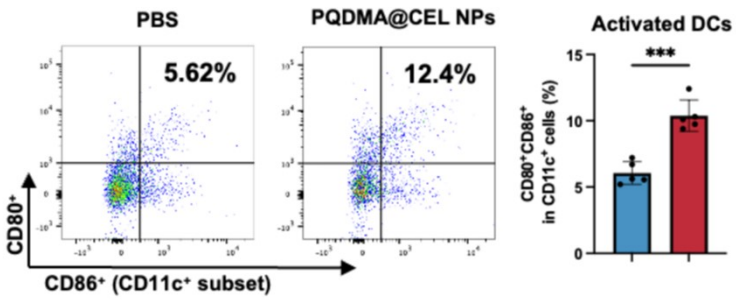

Figure S7. Representative flow cytometry profile and quantification of activated DCs in draining lymph nodes. Data are presented as mean ± SD of biological replicates (n = 5). (****p < 0. 0001; ***P <0.001; ** P <0.01; * P <0.05; n.s. no significance)

Supplementary table 1.

Antibodies	**Company**	**Catalog**	**Application**
Hoechst 33342	Invitrogen	H3570	IF
DiD cell label solution	Invitrogen	V22887	IF
Lyso-Tracker Green	Beyotime	C1047S	IF
Mito-Tracker Deep Red	Beyotime	C1034-50	IF
Annexin V /PI Kit	LIANKE BIO	A503441	FC
Cell Counting Kit 8	APE BIO	K1018	
Ms CD45 APC-Cy7 30-F11	BD Pharmingen	557659	Flow
Ms CD3ePE-Cy7 145-2C11	BD Pharmingen	552774	Flow
Ms CD4 FITC RM4-5	BD Pharmingen	553046	Flow
Ms CD8a BV605 53-6.7	BD Horizon	563152	Flow
CD11b Alexa 700 M1/70	BD Pharmingen	557960	Flow
Ms F4/80 BV421 T45-2342	BD Horizon	565411	Flow
Ms CD86 BB700 GL1	BD Pharmingen	BB700	Flow
Ms CD11C PE-Cy7 HL3	BD Pharmingen	558079	Flow
Ms CD25 BV421	BD Horizon	564370	Flow
Ms IFN-Gma R718	BD Horizon	567061	Flow