Table. S1 Concentration of endocytosis inhibitors and markers used in this study

Drug/ treatment	Final Conc.	Pathway targeted/marked	Mode of action
NAV2729	25uM (in vitro) 5mM (in vivo)	Arf1/Arf6	Blocks ARNO- and GEP100-mediated guanine nucleotide exchange on Arf6, and inhibits the activation of △17Arf1 by BRAG2Sec7PH
5-(N-ethyl-N-isopropyi)-amiloride (EIPA)	21uM (in vitro) 5mM (in vivo)	Macropinocytosis	Inhibits Na+/H+ exchange; may affect actin
Chlorpromazine (CPZ)	4ug/ml	CME	Translocate clathrin and AP2 from the cell surface to intracellular endosomes. Inhibits CIE in some cells
Methyl-b-cyclodextrin (MBCD)	1mM	Lipid raft; CME; fluid phase endocytosis	Removes cholesterol from the plasma membrane
Tfn-AF647	25μg/ml	CME	
Dextran-AF647	4mg/ml	Macropinocytosis	
CTxB-AF647	10μg/ml	CIE, CvME	

Abbreviations: CIE, clathrin-independent endocytosis; CME, clathrin-mediated endocytosis; CvME, caveolae-mediated endocytosis.

Table. S2 Particle size and zeta potential of Tat/pDNA/C16TAB nanoparticles

Formulation	N/P ratio	Size/nm	Zeta Potential/mV
Tat/pGL3/C16TAB	1	209±0.8	+11±4.7
	10	82±6.0	+28±5.9
Tat/TF/C16TAB	1	214±2.3	+9±2.2
	10	83±9.1	+22±6.4
Tat/DiYO-1-pGL3/C16TAB	1	301±8.2	+18±5.7
	10	109±7.6	+31±6.3
Tat/ DiYO-1-TF/C16TAB	1	350±5.7	+20±3.4
	10	112±4.0	+36±4.8
Tat/DiYO-3-pGL3/C16TAB	1	303±9.0	+18±4.2
	10	108±5.3	+32±5.5
Tat/ DiYO-3-TF/C16TAB	1	356±9.0	+23±8.4
	10	112±6.3	+35±5.1

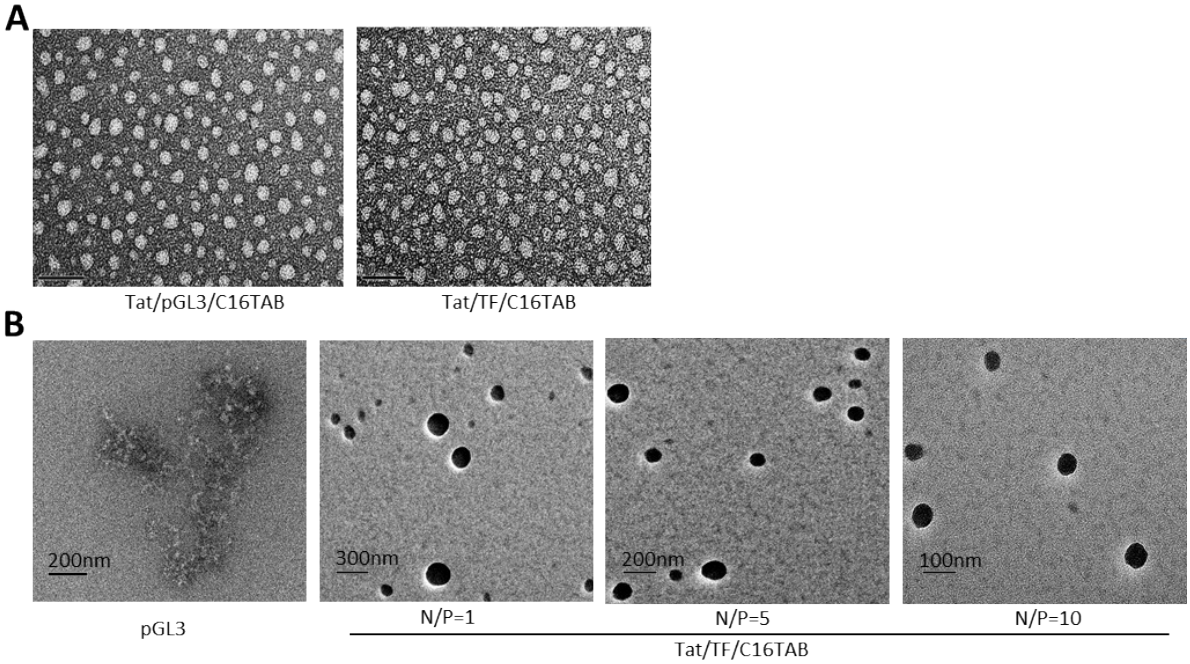

Fig. S1 Representative TEM images of T-P-C nanoparticles. (**A**) No difference on the formulation of Tat/pDNA/C16TAB nanoparticles based on distinct plasmids. N/P=10. (**B**) Smaller size of T-P-C nanoparticles was formulated with increased N/P ratios.

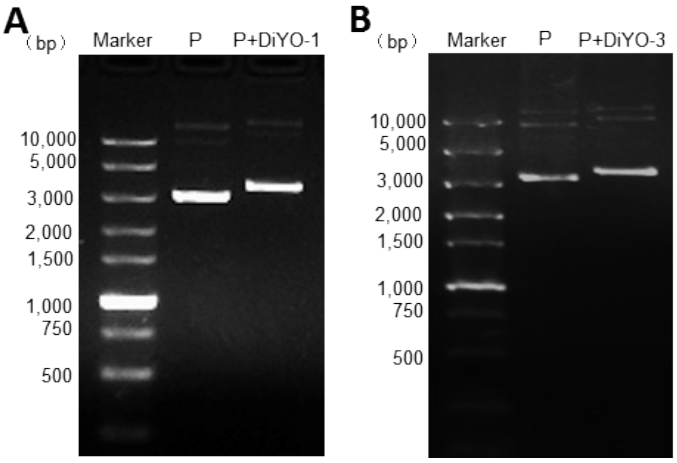

Fig. S2 Labeling efficiency of DiYO-1 or DiYO-3 to pDNA is Similar. Agarose electrophoresis analysis of the mobility difference of pDNA (P: pGL3 plasmid) labeled with DiYO-1 (**A**) or DiYO-3 (**B**).

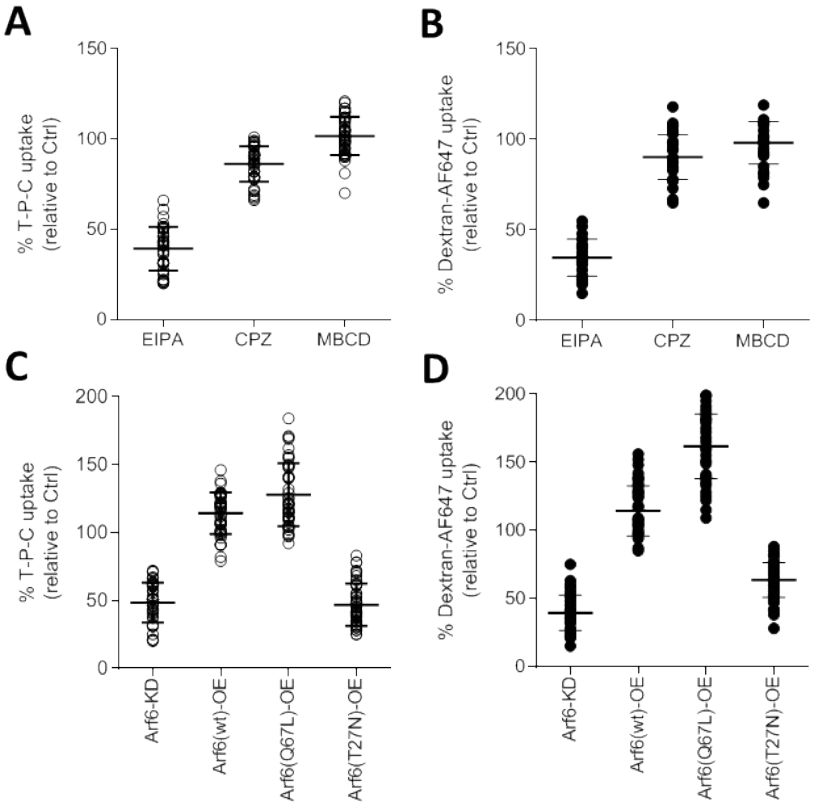

Fig. S3 Scattershot depicts the uptake of indicating ligands in single cell populations. Uptake of DiYO-1-labeled T-P-C nanoparticles (**A**, **C**) and Dextran-AF647 (**B**, **D**) in inhibitor-treated (**A**, **B**) or Arf6-interfered (**C**, **D**) Skov3 cells were quantified by confocal imaging. At least 60 cells were counted for each treatment/transfection. Undisturbed Skov3 cells (Ctrl) were set as 100% for comparison.

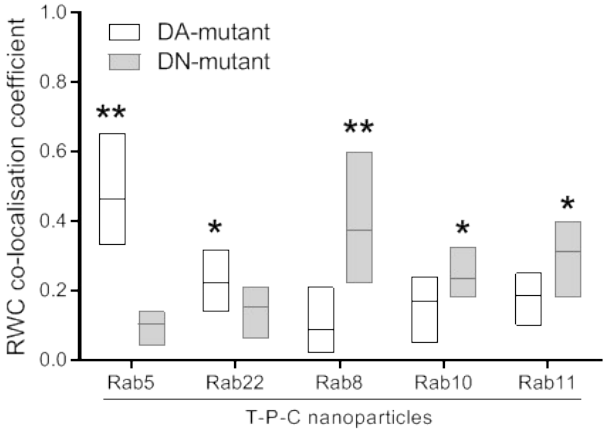

Fig. S4 Co-localization analysis of T-P-C nanoparticles with expressed Rab mutants in cell populations. Box-and-whiskers plots depicting the co-localization of distinct Rab mutants with internalized T-P-C nanoparticles. Rank weighted coefficient (RWC) values were quantified. ≥60 cells were counted for each transfection. n = 3. Significant differences are shown. $^{**}p < 0.01$, $^{*}p < 0.05$.

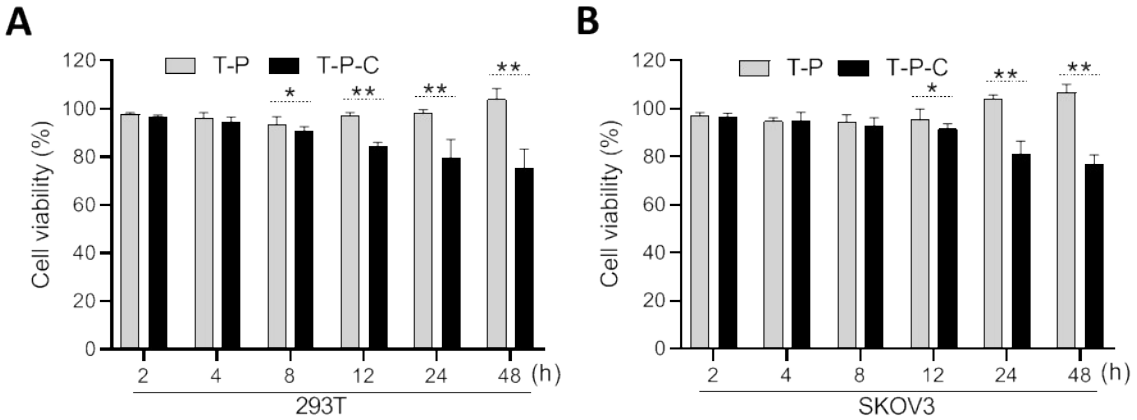

Fig. S5 No cytotoxicity of T-P-C nanoparticles was observed after short-term incubation. Cytotoxicity of nanoparticles was checked by MTT assays on nontumorous 293T cells (**A**) and OC SKOV3 cells (**B**). Both T-P and T-P-C nanoparticles present no cytotoxicity after short time of incubation. A little cytotoxicity of T-P-C nanoparticles was observed after long time of incubation. n=3, ** P <0.01, * P <0.05.

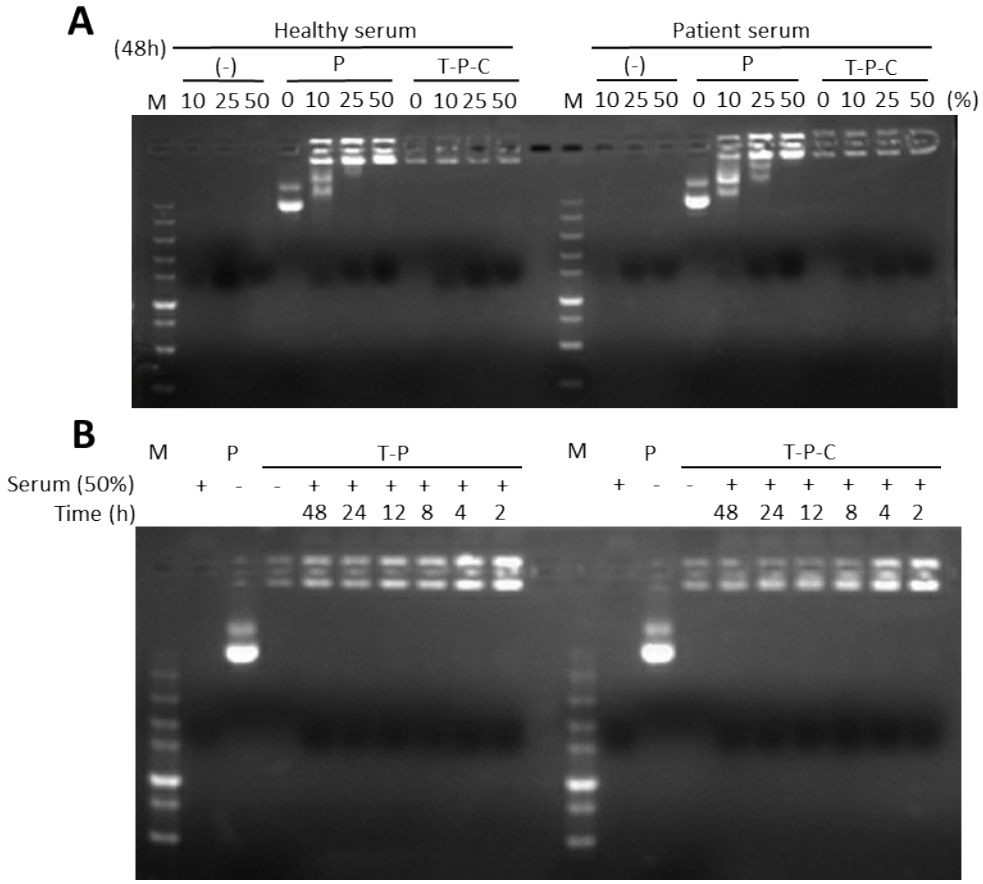

Fig. S6 Characteristics of serum stability for T-P-C nanoparticles. (**A**) Agarose electrophoresis analysis of the stability of T-P-C nanoparticles incubated with healthy human serum and OC patient serum, respectively. P: TF pDNA. (-) only serum was loaded. (**B**) Time-course checking the stability of T-P and T-P-C nanoparticles by agarose electrophoresis analysis. N/P ratios=10. M: DNA ladder.

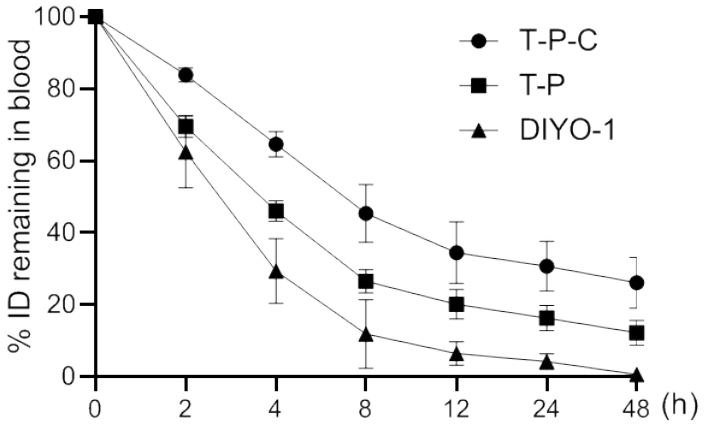

Fig. S7 In vivo blood clearance rate of T-P-C nanoparticles. Blood clearance curves of T-P-C, T-P and free DIYO-1 in mice over 48h after intravenous injection. Balb/c-nu mice were divided into three groups (n=3 per group). DIYO-1-labelled nanoparticles (1 mg/kg. free dye at dose corresponded to its incorporating in nanoparticles) were injected via tail vein. Blood were collected at predetermined time points, and stored into tubes pretreated with 2 % EDTA solution. The DIYO-1 fluorescence intensity in blood samples was analyzed by fluorescence microplate reader. Data expressed as the percent of injected dose (% ID). The initial 5 min fluorescence intensity in the blood was taken as 100 %. Meanwhile, fluorescence intensity in blood from none-injected mice was regarded as background signal.

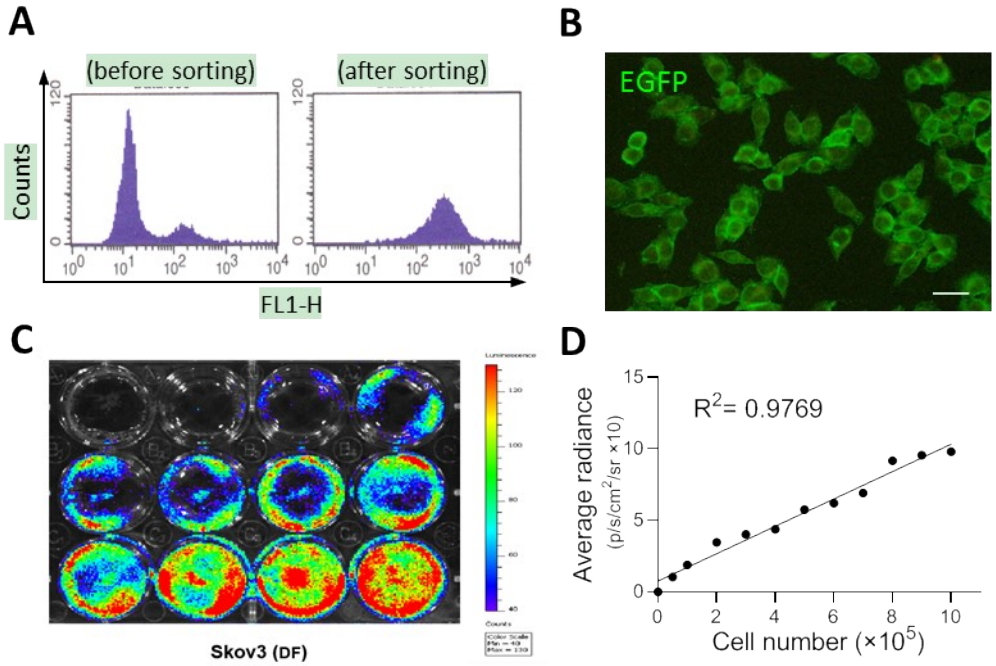

Fig. S8 Identification of lentivirus-transduced Skov3 cells with DF (double fusion) reporter gene. (**A**) Representative FACS results during GFP-signal-gated cell sorting. (**B**) Fluorescence microscope imaging of Skov3 (DF) cells. (**C**) Representative bioluminescence images of Skov3 (DF) cells, and (**D**) increased signals correlated with cell numbers.

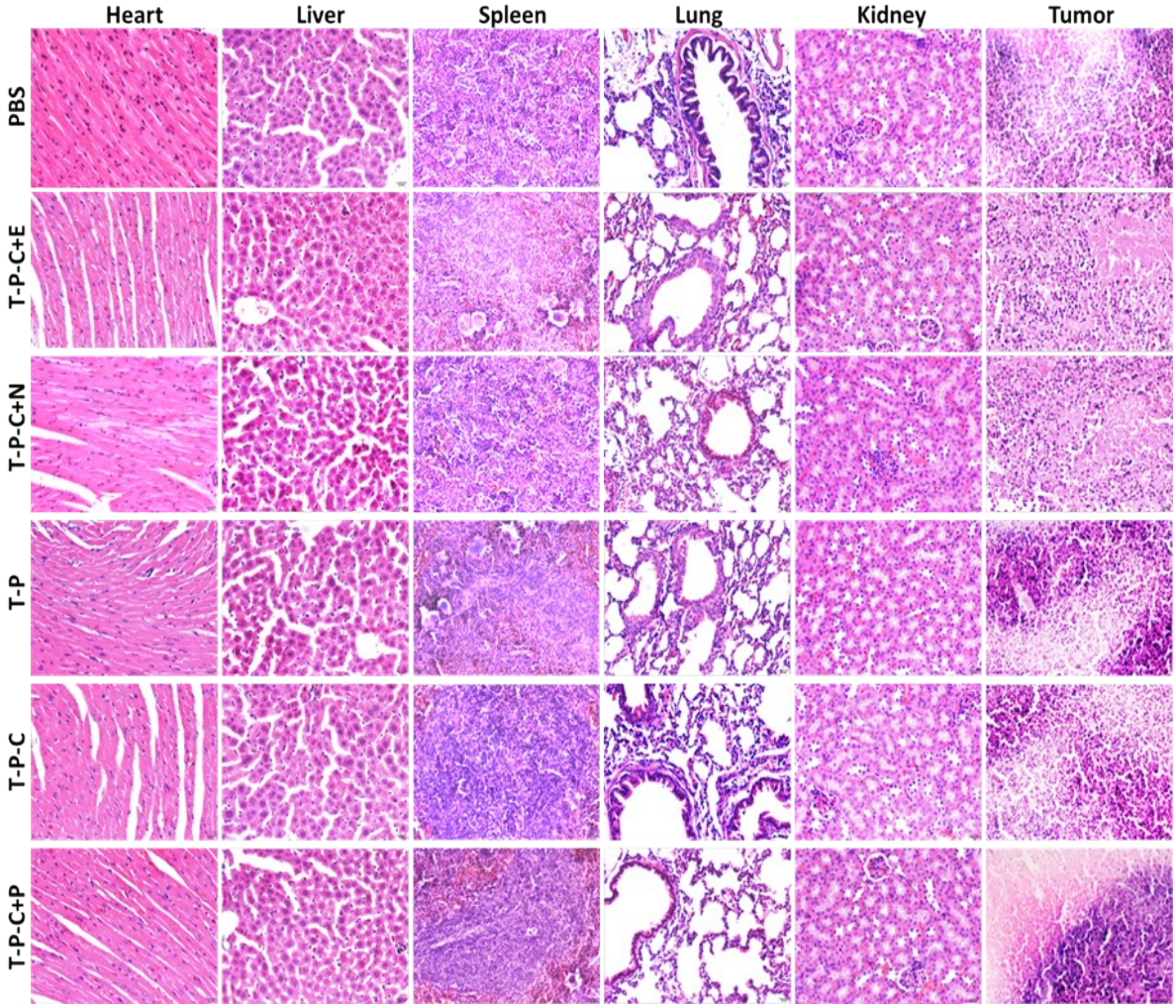

Fig. S9 H&E staining of hearts, livers, spleens, lungs, kidneys and tumor tissues from mice treated under various conditions. T-P-C+E: Tat-TF-C16TAB+EIPA. T-P-C+N: Tat-TF-C16TAB+NAV-2729. T-P: Tat-TF. T-P-C: Tat-TF-C16TAB. T-P-C+P: Tat-TF-C16TAB+PTX. Scale bar: 50 μm.