Supplementary Information

Enhanced Chemotherapy Efficacy by Co-delivery of shABCG2 and Doxorubicin with a pH-responsive Charge-reversible Layered Graphene Oxide Nanocomplex

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KEYWORKDS: *GO nanoparticle, pH-responsive, charge-reversible, co-delivery, polyehtylenimine*

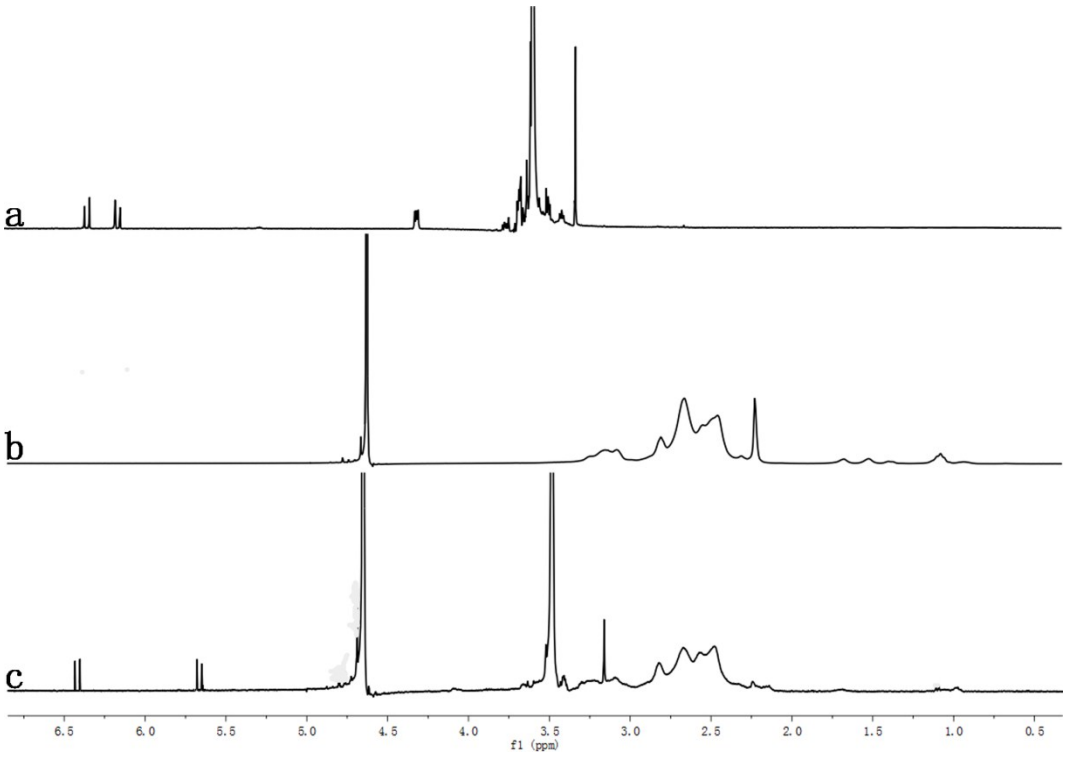

Figure S1. ^{1}H NMR spectra of NHS-terminated mPEG in $CDCl_3$ (a), GO-PEI (b) and GO-PEI-PEG (c) in D_2O (400 MHz). The peak at 6.2 and 6.4 ppm was attributed to -CH=CH of mPEG-NHS in the Figure S1(a); 2.5-3.0 ppm was $-CH_2-$ of PEI in Figure S1 (b); The peak at 5.6-6.3 ppm, 3.5ppm and 2.5-3.0 ppm were attributed to -CH=CH of mPEG-NHS, $-CH_3$ of mPEG-NHS and $-CH_2-$ of PEI respectively in the Figure S1(c).

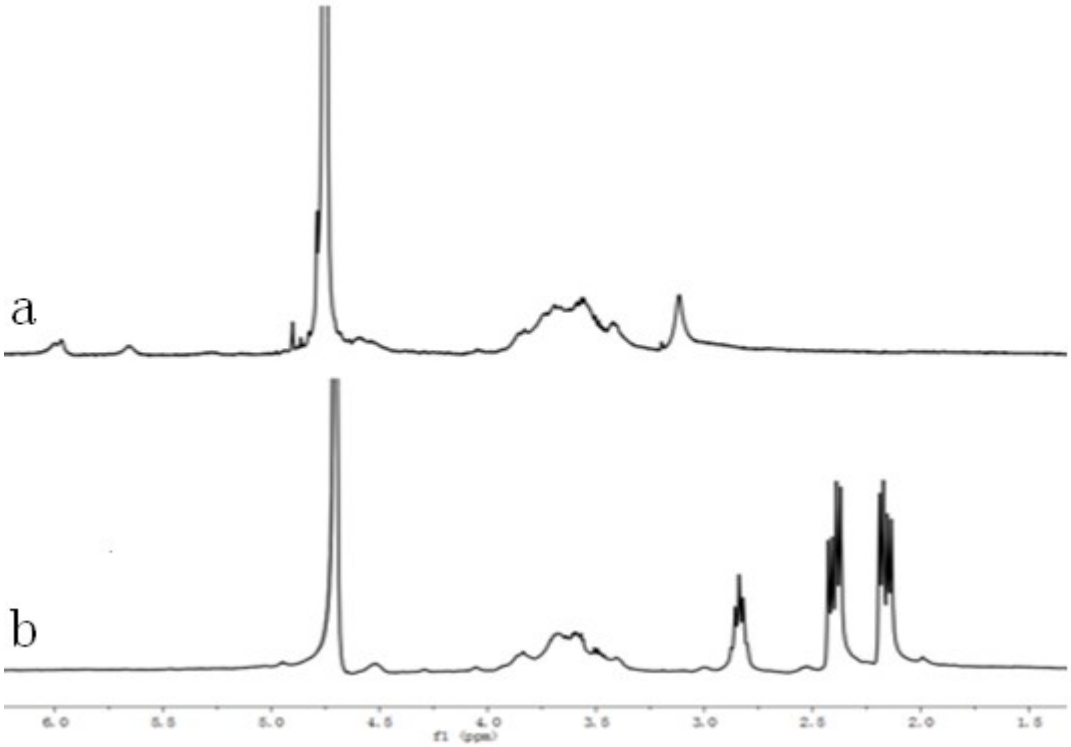

Figure S2. ^{1}H NMR spectrum of CS-Aco (a) and CS-Car (b) in D_2O (400 MHz). (a) The peak at 5.9 ppm and 2.9-3.7 ppm were attributed to –CH=C of Aco and –H of CS, respectively. (b) The peak at 2.9-3.7 ppm was attributed to –H of CS, and the peak at 2.86 ppm, 2.41 ppm and 2.19 ppm were attributed to –CH, CH_2, CH_2 of Car, respectively.

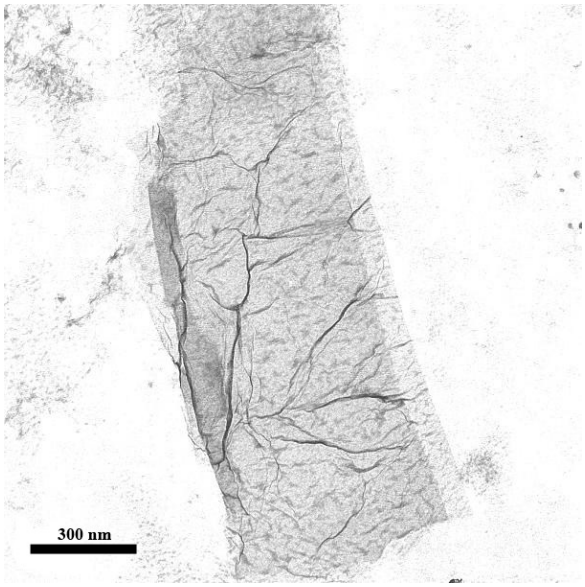

Figure S3. Transmission electron microscopy image of GO sheet. The size of GO was about 700 nm.

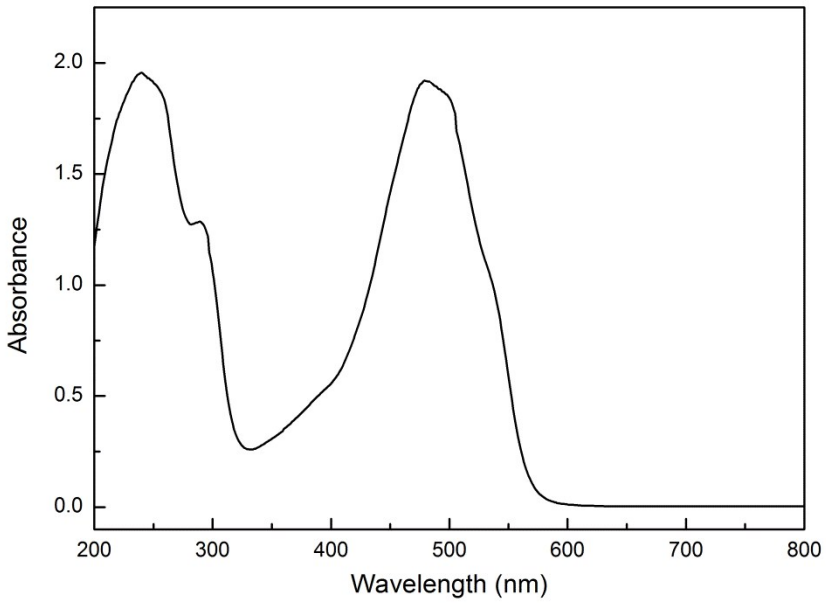

Figure S4. UV-vis spectroscopy of DOX in water.

Experimental Part

Gene silencing efficiency. shABCG2 cloning: The ABCG2 siRNA sequence 5'-GGAUAAGCCA-CUCAUAGAA-3' was cloned into the pSicoR vector with standard procedures. Appropriate primers were designed and purchased from Sangon (Shanghai, China), boiled for 5 min, allowed to cool slowly, and inserted into the pSicoR vector. After the identity of the insert was confirmed by DNA sequencing, shABCG2 or shRNA was assembled into the nanoparticles. The RNA was extracted in the HepG2 cells as described previously. Briefly, the collected cells were dispersed in TRIzol Reagent, followed by the addition of chloroform. The tubes were centrifuged and the aqueous phase was removed. Isopropanol was added to the aqueous phase to precipitate the total RNA, and the precipitates were washed with 75% of ethanol. The RNA pellet was stored in RNase-free distilled.