## Supporting Information

## Bioengineered Human Serum Albumin Fusion Protein as Target/Enzyme/pH Three-Stage Propulsive Drug Vehicle for Tumor Therapy

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Table S1.	Primer see	quence	and the	condition	of PCI	<i>system</i>

Primer name	Primer sequence
	5'- ccctcgagaaaagaCGCGGAGATCGCGGAGATCGCGGAGATGATG
RHMH18	ATGATGATAAGGATGCACAAGAGAGTGAGGTTGCTCATCGA -3'
	5'- ttgcggccgcTTAATGGTGATGGTGATGGTGATGGTGATGGTGATGGTGATGGTGATGG
	TGTGCCCATAATCCTAATGGCTTATCATCATCATCATCTAAGCCTAAGGCAGCTTGACTTGCAGCAAC -3'
RHMH6	5'- ccctcgagaaaagaCGCGGAGATCGCGGAGATCGCGGAGATGAT
	GATGATGATAAGGATGCACAAGAGTGAGGTTGCTCATCGA -3'
	5'- ttgcggccgcTTAATGGTGATGGTGATGGTGTGCCCATAATCCTAAT
	_GGCTTATCATCATCATCTAAGCCTAAGGCAGCTTGACTTGCAGCAAC -3'
HMH18	5'-ccctcgagaaaagaGATGCACACAAGAGTGAGGTTGCTCATCGA -3'
	5'- ttgcggccgcTTAATGGTGATGGTGATGGTGATGGTGATGGTGATGGTGATGGTGATGG
	TGTGCCCATAATCCTAATGGCTTATCATCATCATCTAAGCCTAAGGCAGCTTGACTTGCAGCAAC -3'
RH	5'- ccctcgagaaaagaCGCGGAGATCGCGGAGATCGCGGAGATGATGC
	ACACAAGAGTGAGGTTGCTCATCGATTTAAAGATTTGGGAGAAG -3'
	5'- ttgcggccgcTTAATGGTGATGGTGATGGTGATGGTGATGGTGATGGTGATGGTGATGGTGATG
	GTGTGCCCATAATCCTAATGGTAAGCCTAAGGCAGCTTGACTTGCAGCAACAAGTTTTTTAC -3'

Component	Volume
Primers Star Max (2X)	25 µL
Upstream primer	2 µL
Downstream primer	2 µL
Ultra-pure water	20.5 µL
HSA Template	2.5 µL

 Table S2. The condition of double enzyme digestion

Component	Volume
DNA	3 µL
FastDigest Buffer	2 µL
Xho I	1 µL
Not I	1 µL
Ultra-pure water	13 µL

 Table S3. The condition of plasmid linkage

Component	Volume
T4 ligase	1 μL
FastDigest Buffer	1 μL
pPICZaA (after digestion)	1 μL
DNA (after digestion)	1 μL
Ultra-pure water	16 µL

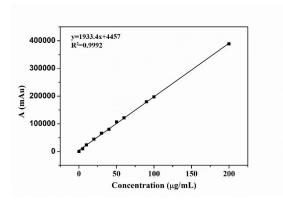


Figure S1 The concentration-absorption curve of PTX in PBS: methanol=45:55.

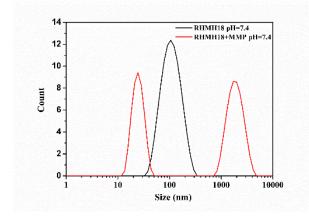
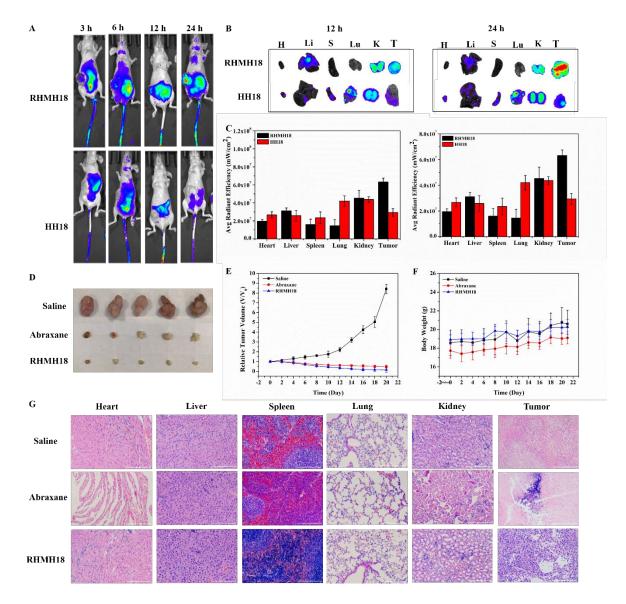


Figure S2 Size difference of RHMH18 NPs before and after adding MMP-2 for 24 h



**Figure S3** (A) NIR fluorescence images of A549 tumor bearing mice after injection of fusion HSA nanomedicine at different time intervals. (B) *Ex vivo* NIR fluorescence images of major organs and tumors after different treatments at 12 h and 24 h. (C) Quantitative analysis of NIR fluorescence intensities of major organs and tumors after different treatments at 12 h and 24 h. (D) Optical images of tumor status stripped from mouse. (E) Relative volumes of tumors. (F) Body weight changes. (G) H&E images of the excised tumor and normal organs.

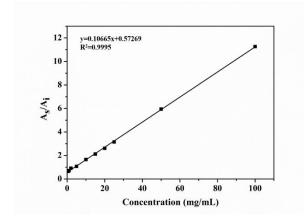


Figure S4 Standard curve of paclitaxel concentration and peak area in plasma samples.