Supporting Information

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SI Materials and Methods

Materials. Maleimide-polyethylene glycol 1000-*N*-hydroxysuccinimide ester (MAL-PEG1000-NHS) was purchased from Quanta BioDesign, Ltd. Polyethylene glycol 2000-*N*-hydroxysuccinimide ester (PEG2000-NHS) was purchased from Laysan Bio. The lipids and lipid derivatives including 1,2-distearoyl-*sn*-glycero-3-phosphoethanolamine-*N*-[methoxy(polyethylene glycol)-1000] (ammonium salt) (PEG1000-PE), 1,2-dioleoyl-sn-glycero-3-phosphoethanolamine (DOPE), 1,2-dioleoylsn- glycero-3-phosphoethanolamine-*N*-(lissamine rhodamine B sulfonyl) (ammonium salt) (rhodamine-PE), and 1,2-dipalmitoyl-*sn*-glycero-3-phosphoethanolamine-*N*-(7-nitro-2–1,3-benzoxadiazol-4-yl) (ammonium salt) (NBD-PE) were purchased from Avanti Polar Lipids. Cysteine-modified transactivating transcriptional activator peptide (Cys-TATp) and MMP2-cleavable (GPLGIAGQ) and uncleavable (GGGPALIQ) octapeptides were synthesized by the Tufts University Core Facility. The BCA Protein Assay Kit, *N*-hydroxysuccinimide (NHS), triethylamine (TEA), chloroform, dichloromethane (DCM) and methanol were purchased from Thermo Fisher Scientific. *N,N′*-Dicyclohexylcarbodiimide (DCC), 4-Dimethylaminopyridine (DMAP), Ninhydrin Spray reagent, Molybdenum Blue Spray reagent, HISTOPAQUE-1083, Triton X-100 (for electrophoresis), sodium salicylate, and propidium iodide were purchased from Sigma-Aldrich Chemicals. Human active MMP2 protein (molecular mass 66,000 Da) and TLC plate (silica gel 60 F254) were from EMD Biosciences. Dialysis tubing [molecular weight cutoff (MWCO) 2,000 Da] was purchased from Spectrum Laboratories, Inc. DMEM, penicillin streptomycin solution (PS) (100×), Simply Blue Safe Stain (Coomassie G-250), Hoechst 33342, Annexin V Alexa Fluor 488 Conjugate, normal mouse sera, and trypsin-EDTA were from Invitrogen Corporation. FBS was purchased from Atlanta Biologicals. SDS/PAGE precast gel (4–20% acrylamide) was purchased from Expedeon Ltd. Ready Gel Zymogram Gel (10% polyacrylamide gel with gelatin), Zymogram Renaturation Buffer, Zymogram Development Buffer and the broad range molecular weight standards were purchased from Bio-Rad. Amicon Ultra-0.5 centrifugal filter device (30K MWCO) and Fluorescent FragEL DNA Fragmentation Detection Kit were purchased from EMD Millipore Corporation. Cytotox 96 Non-Radioactive Cytotoxicity kit was purchased from Promega. Collagenase D was from Roche Diagnostics. Alanine transaminase (ALT) and aspartate transaminase (AST) assay kits were from Biomedical Research Service. ECV304 cell lysate and mouse monoclonal anti-β-tubulin antibody were purchased from Santa Cruz Biotechnology. Human MMP2 ELISA Kit was purchased from Boster Immunoleader. The mouse plasma was purchased from Bioreclamation. The donkey anti-mouse IgG FITC conjugated antibody was purchased from Jackson ImmunoResearch Laboratories. The BCA Protein Assay Kit was purchased from Pierce.

Synthesis, Purification, and Characterization of PEG2000-peptide-PTX. There are two steps in the synthesis of PEG2000-peptide-paclitaxel (PTX) (Scheme 1*A*). The MMP2-cleavable octapeptide and PEG2000-NHS (1.2:1, molar ratio) were first mixed and stirred in carbonate buffer (pH 8.5) under nitrogen protection at 4 °C overnight. The unreacted peptides was removed by dialysis (MWCO 2,000 Da) against distilled water and checked by RP-HPLC as described in a previous study (1).

PEG2000-peptide was then activated with a 20-fold molar excess of DCC/DMAP in DCM. Then, a twofold molar excess of paclitaxel was added and the reaction was carried out under nitrogen in the dark at room temperature overnight. The reaction was monitored by using analytical TLC (chloroform/methanol, 6:4, vol/vol) and visualized by UV at 254 nm and Dragendorff's reagent (a self-preparation using a US Pharmacopeia protocol) staining. The product was purified by preparative TLC (same conditions as analytical TLC) and characterized by ^{1}H-NMR spectroscopy on a Varian 400 MHz spectroscope with $CDCl_3$ and D_2O as solvents. To prepare the uncleavable PTX conjugate, the scramble peptide (GGGPALIQ) was used.

Synthesis, Purification, and Characterization of TATp-PEG1000-PE. Two steps are involved in the synthesis of TATp-PEG1000-PE (Scheme 1*B*). First, the heterobifunctional PEG derivative, NHS-PEG1000-MAL, reacted with DOPE (1:1.4, molar ratio) in DCM. The DOPE-PEG1000-MAL was purified by preparative TLC (same conditions as above). Then, the DOPE-PEG1000-MAL and Cys-TATp (CYGRKKRRQRRR) (1:1.2, molar ratio) were mixed in a pH 7.2 Hepes buffer and the reaction was carried out at 4 °C under nitrogen protection overnight, followed by dialysis (MWCO 2,000 Da) against distilled water to remove unreacted TATp. The reaction and purification processes were monitored by TLC (chloroform/methanol, 8:2, vol/vol) and visualized by using Dragendorff's reagent staining for PEG, Ninhydrin Spray reagent staining for peptides, and Molybdenum Blue Spray reagent staining for phospholipids.

Determination of Critical Micelle Concentration. The critical micelle concentration (CMC) was determined by fluorescence spectroscopy using pyrene as a hydrophobic fluorescent probe (2–4). Briefly, pyrene chloroform solution was added to the testing tube at the final concentration of 8×10^{-5} M and dried on a freeze-dryer overnight. Then, the nanopreparation in Hank's balanced salt solution (HBSS) was added to the tubes at the 10-fold serial dilutions (from 1 to 10^{-7} mg/mL) and incubated with shaking at room temperature for 24 h before measurement. To study the influence of the serum on the CMC, the nanopreparation was hydrated by HBSS containing 50% mouse serum. The fluorescence intensity was measured on an F-2000 fluorescence spectrometer (Hitachi) with the excitation wavelengths (λ_{ex}) of 338 nm (I_3) and 334 nm (I_1) and an emission wavelength (λ_{em}) of 390 nm. The intensity ratio (I338 / I334) was calculated and plotted against the logarithm of the micelle concentration. The CMC value was obtained as the crossover point of the two tangents of the curves.

Cleavage Study of PEG2000-peptide-PTX. The cleavability of PEG2000-peptide-PTX before and after incorporation into the PEG1000-PE micelles was evaluated by enzymatic digestion (1). Briefly, 2.5 mg/mL PEG2000-peptide-PTX was incubated with active human MMP2 (5 ng/μL) in pH 7.4 HBS containing 10 mM $CaCl_2$ at 37 °C overnight. The reaction mixture was analyzed by TLC (chloroform/methanol, 8:2, vol/vol) followed by Dragendorff's reagent staining. In parallel, the PTX conjugate was incubated with the mouse plasma at 37 °C overnight and checked by TLC and visualized by Dragendorff's staining and UV at 254 nm.

Determination of MMP2 Levels in Cell Cultures and Tissues. The human non-small cell lung cancer cells (A549 cells) and normal rat cardiomyocytes (H9C2 cells) were seeded in a six-well plate at 1×10^5 cell per well and maintained in complete growth media for 3 d. Then, the cell media was collected and concentrated by using an Amicon Ultra-0.5 centrifugal filter device (30K MWCO) at 7,200 × *g* for 20 min. The concentrated media was loaded and

run on a 4–20% SDS/PAGE followed by Simply Blue Safe Stain staining. The broad range molecular weight standards were used as molecular weight markers. The gelatinase activity of the secreted proteins was determined by gelatin zymography using a precast 10% zymogram gel followed by Simply Blue Safe Stain staining. ECV304 cell lysate was used as positive control for MMP2. For quantitative detection of MMP2, an MMP2 ELISA (sensitivity < 10 pg/mL) was performed to detect the MMP2 concentration in the original cell media (without concentration process) according to the manufacturer's instruction.

To determine the MMP2 levels in tissues, the tumor-bearing mice were killed and the tumors and major organs were collected. The tissues were then homogenized in PBS containing 0.5% Triton X-100 by a TissueRuptor (QIAGEN) on ice. The homogenates were centrifuged at 911 × *g* for 20 min and then analyzed by the MMP2 ELISA. The concentration of the total protein was measured with a BCA Protein Assay Kit.

Preparation of the MMP2-Sensitive Nanopreparation. To prepare the MMP2-sensitive nanopreparation (TATp-PEG1000-PE/PEG2000-peptide-PTX), PEG2000-peptide-PTX (50 mol%), PEG1000-PE (40 mol%), and TATp-PEG1000-PE (10 mol%) were dissolved in chloroform and dried on a freeze-dryer overnight, followed by hydration with HBSS at room temperature. The nonsensitive nanopreparation (TATp-PEG1000-PE/PEG2000-peptide-PTX uncleavable), nanopreparations without TATp modification (PEG1000-PE/PEG2000-peptide-PTX and PEG1000-PE/PEG2000-peptide-PTX uncleavable), and the empty micelle (TATp-PEG1000-PE) were prepared by using the same method. The particle size and morphology of these nanopreparations were analyzed by transmission electron microscopy (model XR-41B) (Advanced Microscopy Techniques) using negative staining with 1% phosphotungstic acid.

Stability Study of the MMP2-Sensitive Nanopreparation. The MMP2-sensitive nanopreparations were incubated with HBSS at 37 °C for 4 h before size measurement. To study the long-term stability of the nanopreparations, the samples were kept in HBSS at 4 °C for 3 wk. To evaluate the in vivo stability, the nanopreparations were incubated with the normal mouse serum (1:10, vol/vol) at 37 °C for 4 h. The particle size was analyzed by dynamic light scattering (DLS) on a Coulter N4-Plus Submicron Particle Sizer (Beckman Coulter).

Determination of PTX by RP-HPLC. The samples were analyzed by RP-HPLC on a reverse-phase C18 column (250 mm × 4.6 mm; Alltech) by the LaChrom Elite HPLC system (Hitachi). The chromatograms were collected at 227 nm by using acetonitrile/water (45:55, vol/vol) as the mobile phase at a flow rate of 1.0 mL/min.

In Vitro Drug Release. To study whether the MMP2 cleavage influences the encapsulation of paclitaxel, the drug release study was performed. Briefly, TATp-PEG1000-PE/PEG2000-peptide-PTX micelles were pretreated with MMP2 at 37 °C overnight. The samples (0.8 mL) were then dialyzed (MWCO 2,000 Da) against 20 mL of water containing 1 M sodium salicylate to maintain the sink condition (5). The PTX in a methanol/water (1:4, vol/vol) mixture and TATp-PEG1000-PE/PEG2000-peptide-PTX micelles without MMP2 pretreatment were used as controls. The PTX in the outside and inside media was determined by RP-HPLC. To obtain the naked PTX (without amino acid residues), the samples were trypsinized at 37 °C for 1 h before HPLC.

Tubulin Immunostaining. Cells (1.2×10^5) were seeded on glass coverslips in a 12-well plate and incubated for 24 h before treatment. Cells were then incubated with 24 nM of PTX formulations at 37 °C overnight. The treated cells were washed with PBS and fixed with 4% paraformaldehyde followed by the permeabilization with 0.5% Triton X-100 at room temperature. After washing with PBS, the cells were incubated with a mouse monoclonal anti–β-tubulin antibody at a 1:10 dilution in PBS containing 1% BSA at room temperature for 1 h. Cells were then washed three times with PBS before staining with a donkey anti-mouse IgG FITC conjugated antibody at a 1:100 dilution at room temperature for 1 h. Finally, the cells were washed with PBS and stained with Hoechst 33342 before confocal microscopy.

Establishment of A549 Tumor Cell Spheroids. A549 multicellular spheroids were formed according to the method in refs. 6 and 7. Briefly, A549 cells were grown in DMEM supplemented with 50 U/mL penicillin, 50 μg/mL streptomycin, and 10% FBS at 37 °C. The 96-well plates were coated with 1.5% agarose in DMEM to prevent cell adhesion and seeded with 1×10^4 cells per well. The plates were then centrifuged at 405 × *g* for 15 min, and the multicellular aggregates were maintained at 37 °C for spheroid formation. The spheroid was identified by its size and shape. The 6-d spheroids with a diameter of 700–900 μm were used as the in vitro tumor model.

Cellular Uptake of PEG2000-peptide-PTX and Its Nanopreparations. To study the cellular uptake in monolayer cells, A549 cells were seeded in 24-well plates at 1.6×10^5 cells per well. After 24 h, the cells were washed twice with pH 7.4 PBS and then treated with various NBD-PE–labeled formulations in 300 μL per well of serum-free media. After 2-h incubation with the formulations, the media was removed and the cells were washed with serum-free media three times. Cells were then analyzed by FACS and confocal microscopy. For FACS analysis, the cells were trypsinized and collected by centrifugation at 720 × *g* for 4 min. After washing with ice-cold PBS, the cells were resuspended in 400 μL of PBS and immediately applied on a BD FACS Calibur flow cytometer (BD Biosciences). The cells were gated upon acquisition by using forward versus side scatter to exclude debris and dead cells. The data were collected (10,000 cell counts) and analyzed with BD Cell Quest Pro Software. For confocal microscopy, the cells were fixed by 4% paraformaldehyde and stained with 5 μM Hoechst 33342 for 15 min at room temperature. The photos were taken with a Zeiss LSM 700 confocal microscope system and analyzed by using Zeiss Image Browser software.

To study the cellular uptake in spheroids, the 6-d spheroids were transferred to eight-well glass Lab-Tek chamber slides (Thermo Scientific) and incubated with various rhodamine-PE–labeled formulations in 100 μL per well of serum-free media for 2 h. Then, the spheroids were washed with PBS and imaged by confocal microscopy. The spheroids were further sectioned with a HM550 Cryostats microtome (Thermo Scientific) and stained by Hoechst 33342. The spheroid sections were observed by using a Nikon Eclipse E400 microscope and a Spot Advanced software (Spot Imaging).

TATp Competition Study. A549 cells were seeded in 24-well plates at 1.6×10^5 cells per well in 300 μL per well of complete growth media. After 24 h, the free TATp (0.35, 3.5, or 35 μM) was added into the cell media. Then, 10 μL of the rhodamine-PE–labeled MMP2-sensitive nanopreparation (1 mg/mL) was immediately added and incubated for 2 h. The media was removed and the cells were washed with serum-free media three times. Cells were then analyzed by FACS.

Cytotoxicity of PEG2000-peptide-PTX and Its Nanopreparations. A549 cells and H9C2 cells were seeded in 96-well plates at 2×10^3 cells per well and incubated for 24 h before treatments. To study cytotoxicity of PTX and its conjugates, a series of diluted samples were added to A549 or H9C2 cells and incubated for 72 h. To study the cytotoxicity of the PTX-containing nanopreparations,

various formulations were added to A549 cells and incubated for 72 h. The cell viability was determined by CellTiter-Blue Cell Viability Assay. Briefly, 15 μL of CellTiter-Blue Reagent was diluted with 100 μL of complete growth medium per well and incubated with treated cells at 37 °C for 2 h. Thereafter, the fluorescence was recorded at $\lambda_{ex} = 560$ nm and $\lambda_{em} = 590$ nm by using a Labsystems Multiskan MCC/340 microplate reader.

To study the response of spheroids to PTX conjugates and its nanopreparations, various formulations were used to treat 6-d A549 spheroids at the dose of 29.5 ng/mL every other day for 6 d. The spheroids were maintained in the 96-well plate with 100 μL of complete growth media. The additional 50 μL of fresh media was added to compensate for evaporation when the treatment was given. On day 12, the LDH release was measured with a Cytotox 96 Non-Radioactive Cytotoxicity kit to evaluate the cytotoxicity of the formulations on spheroids as reported by refs. 7 and 8. The LDH release of spheroids was normalized to total LDH content from both spheroids and their growing media.

Apoptosis Analysis. The apoptosis of A549 cells was determined by FACS using annexin V/propidium iodide double staining after treatment with PTX or its conjugate at 29.5 ng/mL for 72 h according to the manufacturer's instruction. Briefly, treated cells were trypsinized and collected by centrifugation at 405 × *g* for 5 min. The cells were washed and resuspended with 100 μL of PBS. The cells were first stained with annexin V (25 μg/mL) for 15 min on ice and then incubated with propidium iodide (50 μg/mL) for 5 min before FACS analysis.

Establishment of the NSCLC Xenograft Mouse Model. Female nude mice (NU/NU, 4–6 wk old) were purchased from Charles River Laboratories. All animal procedures were performed according to an animal care protocol approved by Northeastern University Institutional Animal Care and Use Committee. Mice were housed in groups of five at 19–23 °C with a 12-h light–dark cycle and allowed free access to food and water.

Approximately 2×10^6 A549 cells suspended in 50 μL of HBSS were mixed with the phenol-red free high concentration Matrigel (1:1, vol/vol) and inoculated in nude mice by s.c. injection over their right flanks. The tumor was monitored for length (l) and width (w) by caliper and calculated by the equation $V = lw^2/2$.

Biodistribution and Intratumor Localization Study. HBSS, the rhodamine-PE labeled/loaded MMP2-sensitive nanopreparation, and its nonsensitive counterpart were i.v. injected in tumor-bearing mice with a tumor size of approximately 400 mm^3 at the dose of 5 mg/kg PTX via tail vein. At 2 h after injection, mice were anesthetized and killed. The tumor and major organs (heart, liver, spleen, lung, and kidney) were collected. To determine the in vivo cellular uptake, the fresh tissues were minced into small pieces and incubated in 400 U/mL collagenase D solution for 30 min at 37 °C to dissociate cells (9). The single-cell suspension was analyzed immediately by FACS. The rest of the tissue was stored at −80 °C and sectioned with a microtome, followed by Hoechst 33342 staining. The tumor sections were analyzed by confocal microscopy. To determine the PTX's tissue accumulation, in addition to the tumor and organs, the whole blood was collected from the retroorbital plexus. Portions of the tissues and blood were weighed and homogenized in PBS by a TissueRuptor on ice. The homogenates were mixed with 10 volumes of t-butyl methyl ether and extracted for 30 min on a shaker at room temperature. The mixture was centrifuged at 911 × *g* for 20 min. The organic layer was transferred to a fresh tube and blown to dryness with a stream of nitrogen. The residue was reconstituted by methanol and analyzed by RP-HPLC. The standard curves were made by spiking PTX into the blank tissues and blood, followed by the same extraction procedure.

Antitumor Efficacy and Side Toxicity Study. Tumor-bearing mice were randomized to five groups (seven mice per group), when the tumor size was approximately 200 mm^3. HBSS, PTX in Cremophor EL/ethanol (1:1, vol/vol), PEG2000-PE/PTX, TATp-PEG1000-PE/PEG2000-peptide-PTX, and the nonsensitive nanopreparation were injected i.v. in mice at the dose of 5 mg/kg PTX twice a week for 4 wk. The tumor volume and body weight were monitored every 3 d until the end of the experiment. At the end of the experiment, whole blood was collected from the retroorbital plexus. White blood cells were counted by using a hemocytometer by microscopy. The activities of alanine transaminase (ALT) and aspartate transaminase (AST) were measured by the ALT and AST assay kits. The tumor and major organs were collected and sectioned. Tumor sections were stained by Hoechst 33342, and the tumor cell apoptosis was analyzed by terminal deoxynucleotidyl transferase dUTP nick end labeling assay using a FragEL DNA Fragmentation Detection Kit. The pictures were taken by confocal microscopy. Hematoxylin and eosin staining was conducted to identify the necrotic areas of tumors and to examine the pathological change of major vital organs. Briefly, frozen tumor and tissue sections were stained with Harris modified hematoxylin solution followed by rinsing in running tap water and then restained with eosin Y, dehydrated, cleared and slide mounted. The slides were visualized by light microscopy at 20× magnification.

Statistical Analysis. Data were presented as mean ± SD. The difference between the groups was analyzed by using a one-way ANOVA analysis by the commercial software PASW Statistics 18 (SPSS). $P < 0.05$ was considered statistically significant.

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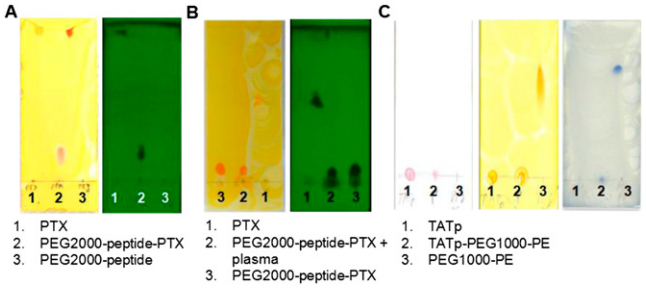


Fig. S1. Characterization of PEG2000-peptide-PTX (*A*), stability of PEG2000-peptide-PTX in plasma (*B*), and TATp-PEG1000-PE (*C*) by TLC. For characterization of PEG2000-peptide-PTX, the samples were run in chloroform/methanol (6:4, vol/vol) and visualized with Dragendorff's reagent (*Left*) and UV 254 nm (*Right*). For characterization of TATp-PEG1000-PE, the samples were run in chloroform/methanol (8:2, vol/vol) and visualized by Ninhydrin reagent (*Left*), Dragendorff's reagent (*Center*) and Molybdenum blue (*Right*).

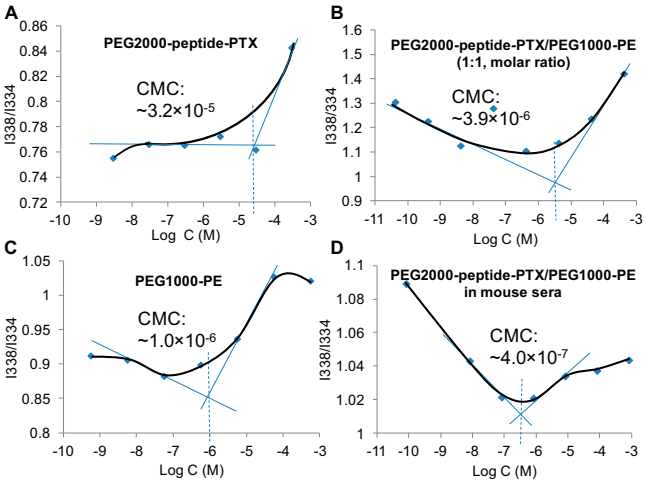


Fig. S2. Determination of the critical micelle concentration (CMC). The CMC of the nanopreparations was determined by fluorescence spectroscopy using pyrene as a hydrophobic fluorescent probe. The samples were hydrated by HBSS (*A*–*C*) or HBSS containing 50% mouse serum (*D*) at a 10-fold serial dilution and incubated with shaking at room temperature for 24 h before measurement. The intensity ratio (I338 / I334) was calculated and plotted against the logarithm of the micelle concentration. The CMC value was obtained as the crossover point of two tangents of the curves.

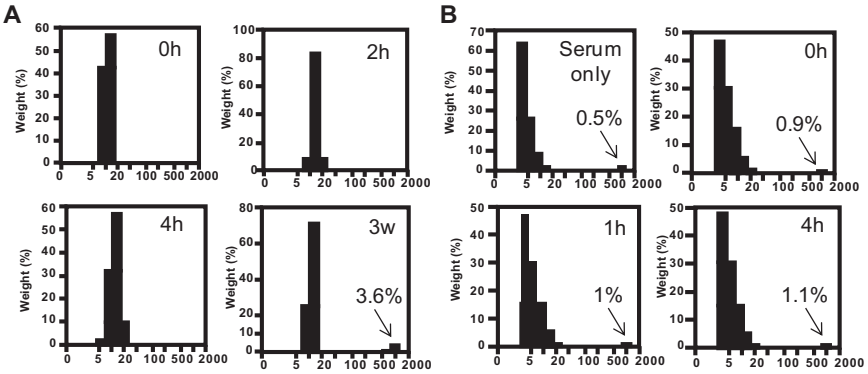


Fig. S3. Stability study by the DLS. (*A*) The stability of the nanopreparations in HBSS for 0–4 h at 37 °C and for 3 wk at 4 °C. (*B*) To evaluate the serum stability, the nanopreparations were incubated with normal mouse sera (1:10, vol/vol) at 37 °C for 0–4 h. The percentage of the particles with the size >500 nm was determined and indicated on the histogram.

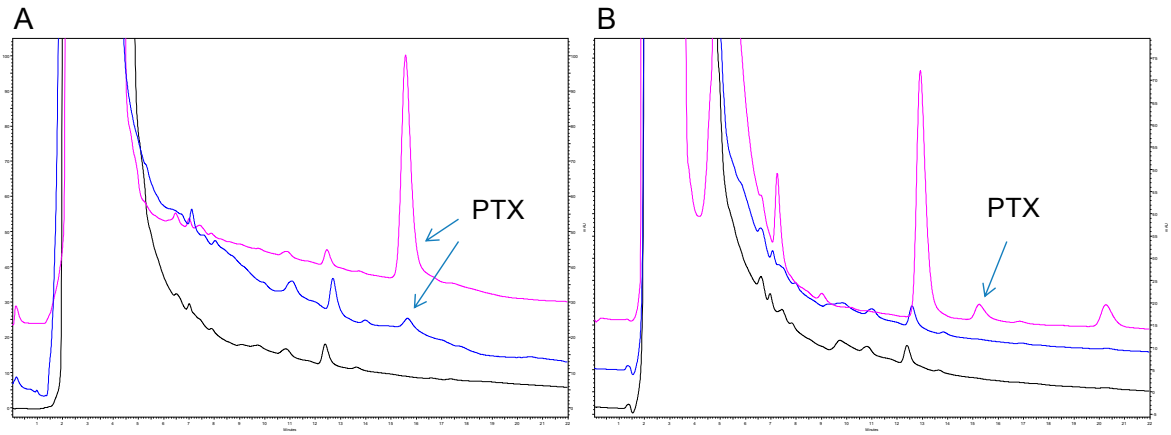


Fig. S4. In vitro drug release. The PTX release was measured by RP-HPLC after dialysis (MWCO 2,000 Da) against 1 M sodium salicylate for 24 h at 37 °C. All samples were trypsinized at 37 °C for 1 h before measurement. (*A*) PTX/methanol/water mixture. Black, the blank media containing trypsin; blue, the outside media; pink, the inside media. (*B*) TATp-PEG1000-PE/PEG2000-peptide-PTX micelles. Black, the outside media (TATp-PEG1000-PE/PEG2000-peptide-PTX inside the tube); blue, the outside media (TATp-PEG1000-PE/PEG2000-peptide-PTX + MMP2 overnight, inside the tube); pink, the inside media (TATp-PEG1000-PE/PEG2000-peptide-PTX + MMP2 overnight, inside the tube).

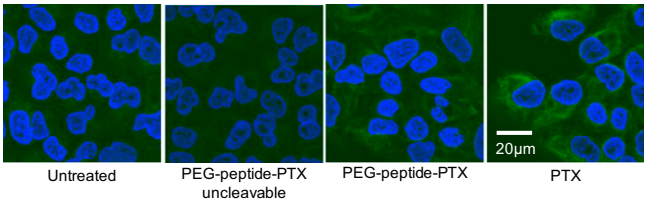


Fig. S5. Tubulin immunostaining. A549 cells were seeded on glass coverslips and treated with 24 nM of PTX formulations at 37 °C overnight. Then, the cells were fixed and permeabilized, followed by staining with a mouse monoclonal anti–β-tubulin antibody and a donkey anti-mouse IgG FITC conjugated antibody. Finally, the cell nuclei were visualized by Hoechst 33342 before confocal microscopy.

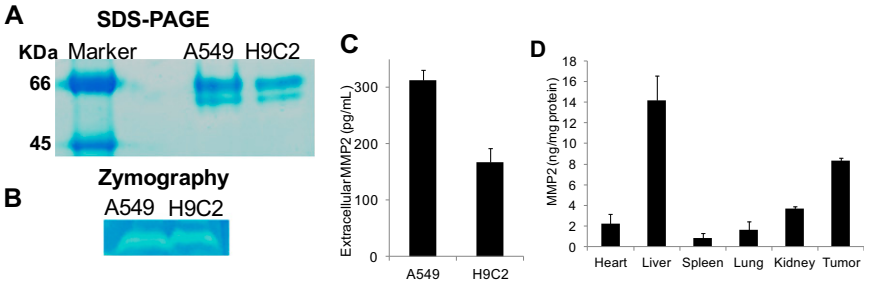


Fig. S6. MMP2 levels in cell culture media and mouse tissues. The same numbers of H9C2 and A549 cells were maintained in complete growth media for 3 d. The media was then collected and concentrated by ultrafiltration before SDS/PAGE (*A*) and zymography (*B*). (*C*) For quantitative detection of MMP2, an MMP2 ELISA was performed to detect the MMP2 concentration in the original cell media (without concentration process). To determine the MMP2 levels in tissues, tumors and major organs were collected and homogenized in PBS containing 0.5% Triton X-100. (*D*) The homogenates were analyzed by the MMP2 ELISA and normalized by the concentration of the total protein.

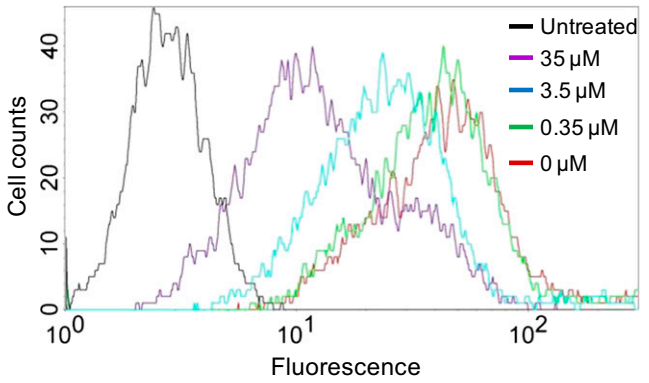


Fig. S7. TATp competition study. A549 cells were seeded in 24-well plates at 1.6×10^5 cells per well in 300 μL per well of complete growth media. After 24 h, the free TATp (0.35, 3.5, or 35 μM) was added into the cell media. Then, 10 μL of the rhodamine-PE–labeled MMP2-sensitive nanopreparation (1 mg/mL) was immediately added and incubated for 2 h. The cells were collected and analyzed by FACS.

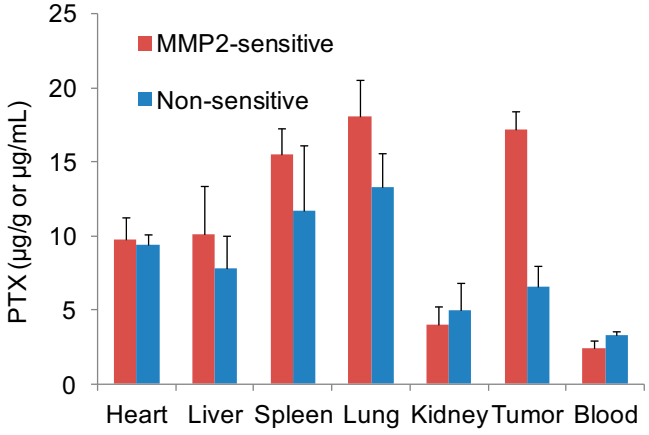
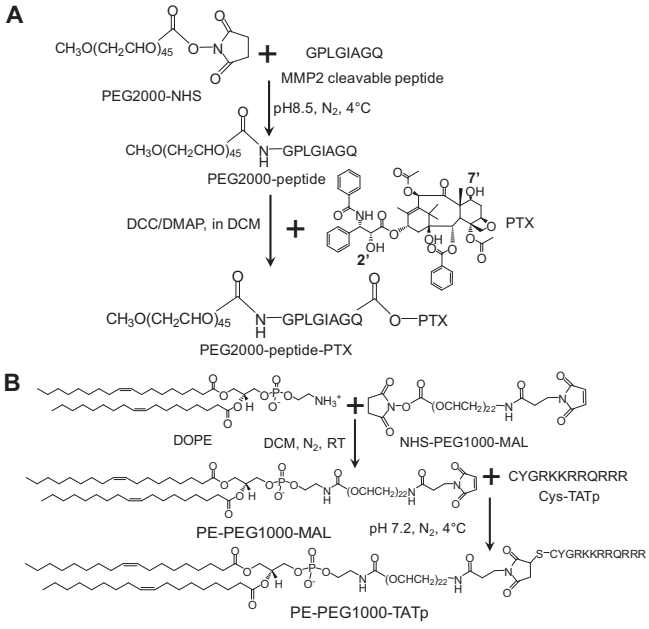


Fig. S8. Tissue distribution of PTX. At 2 h after i.v. injection of 5 mg/kg PTX formulations, the tumor, blood, and major organs were collected, weighed, and homogenized in PBS. The homogenates were extracted with 10 volumes of t-butyl methyl ether followed by centrifugation. The extracted PTX was reconstituted by methanol and analyzed by RP-HPLC. The unit for blood is micrograms per milliliter. The unit for other tissues is micrograms per gram.



Scheme 1. Synthesis of PEG2000-peptide-PTX (*A*) and PE-PEG1000-TATp (*B*).