

Electronic Supplementary Material (ESI) for Nanoscale
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Supporting Information

Thermally induced silane dehydrocoupling on porous silicon nanoparticles for ultra-long-acting drug release

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1. Materials and Methods

General information

The chemical reagents were purchased from Sigma-Aldrich (St. Louis, MO, USA), Merck (Darmstadt, Germany), Gibco (Carlsbad, CA, USA), Hyclone (Logan, UT, USA), Virginia Semiconductor, Inc. (Fredericksburg, VA, USA), J.T. Baker (Phillipsburg, NJ, USA), and Samchun Chemicals (Seoul, Rep. of Korea). Commercially available reagents and anhydrous solvents were used without further purification. Octadecylsilane (Product No. 442291), Pluronic® F127 (F127, Product No. P2443-250G), Potassium hydroxide (KOH, Product No. 484016) were purchased from Sigma-Aldrich (St. Louis, MO, USA). Phosphate-buffered saline (PBS pH 7.4 (10×), Product No. 70011044), Trypsin-EDTA (Product No. 25200-056), and Penicillin streptomycin (Product No. 15140-122) were purchased from Gibco (Carlsbad, CA, USA). Fetal bovine serum (FBS, SH30084.03), Dulbecco's modified eagle's medium (DMEM, Product No. SH30243.01), and Dulbecco's phosphate-buffered saline (DPBS, Product No. SH30028.02) were purchased from Hyclone (Logan, UT, USA). Single crystal silicon wafers were obtained from Virginia Semiconductor, Inc. (Fredericksburg, VA, USA). 48% Hydrofluoric acid was purchased from J.T. Baker (Product No. 9564-04, Phillipsburg, NJ, USA). 7-Ethyl-10-hydroxycamptothecin (SN-38, Product No. FE29579) was purchased from Carbosynth (Berkshire, UK). Dimethyl sulfoxide (DMSO, Product No. 1.02952.1000) was purchased from Merck (Darmstadt, Germany). Sea-sand (10~20 mesh, Product No. 23014S1550) was purchased from Junsei (Tokyo, Japan). Carbon tape (Type-B) and copper grid (Product No. 01813-F) were purchased from Ted Pella, Inc. (Redding, CA, USA). Cell counting Kit-8 (CCK-8, Product No. CK04-13) was purchased from Dojindo (Kumamoto, Japan). Corning® Matrigel® Basement Membrane Matrix (Phenol red-free, LDEV-free, 10 mL, Product No. 356237) was purchased from Corning Incorporated (Bedford, MA, USA). Ethanol (Ethyl alcohol, 94.5%) was purchased from Samchun Chemicals (Product No. E0223, Seoul, Rep. of Korea). 4% Paraformaldehyde (Product No. PC2031-100-00) was purchased from Chembio (Seoul, Rep. of Korea).

Preparation of porous silicon nanoparticles (named pSiNPs)

Porous silicon nanoparticles (pSiNPs) were fabricated by electrochemical etching; galvanostatic anodization of p⁺⁺-type single-crystal silicon wafers (heavily boron-doped) in an aqueous ethanolic hydrofluoric acid electrolyte consisting of 3 : 1 (v : v) of 48% aqueous HF : absolute ethanol (EtOH).^{1, 2} [CAUTION: HF is highly toxic and proper care should be used to avoid contact with skin or lungs.] Before the preparation of the pSi layers, the silicon wafer was anodized to generate a thin porous layer, commonly referred to as a “sacrificial layer”, in the HF-containing electrolyte, and the resulting porous layer was then dissolved by treatment with aqueous potassium hydroxide (KOH, 2 M). The etching waveform was applied with a lower current density of 44 mA cm⁻² and was applied for 1.8 s, followed by a higher current density pulse of 321 mA cm⁻² for 0.4 s. This waveform was repeated for 300 cycles generating a porous silicon (pSi) film with “perforations” repeating approximately every 200 nm through the porous layer. The pSi film was removed from the silicon substrate by applying a current density of 3.0 mA cm⁻² for 300 s in a solution containing 1 : 12 (v : v) of 48% aqueous HF : EtOH. The collected pSi film was placed in EtOH (6 mL) in a sealed glass vial (22.18 mL size, VWR, Product No. 66011-143, Radnor, PA, USA) and fractured into nanoparticles in an ultrasonic bath (VWR, PA, USA) for 24 h. Then, the resulting pSiNPs were collected using a centrifuge (Eppendorf Centrifuge model 5418, 5418FQ924939, Eppendorf AG, US) at 14000 rpm for 30 min and washed 3 times with EtOH.

Preparation of ODS-grafted pSiNPs (named pSiNPs-ODS)

As-prepared pSiNPs (~1 mg) were dispersed in the mixture of octadecylsilane (ODS, 100 μL) and EtOH (500 μL). The mixture was stirred (Digital Hotplate Stirrer, DH.WMH03020, Daihan, Rep. of Korea) at 80 °C for 16 h (300 rpm). After the stirring, the ODS-grafted pSiNPs (**pSiNPs-ODS**) were sequentially washed 3 times by centrifugation (14000 rpm, 15 min) with 100% *n*-hexane, 1 : 1 (v : v) *n*-hexane:EtOH, and 100% EtOH.³

For the SN-38 drug loading, **pSiNPs** or **pSiNPs-ODS** (~1 mg) was dispersed in 575 μL of EtOH and then added with the SN-38 stock solution (40 mg mL^{-1} in DMSO, 25 μL). To evaporate the solvent, the mixture was heated in a sea-sand bath at 50 $^{\circ}\text{C}$ for 24 h. The resulting pSiNPs were then washed (3 times) with EtOH (1 mL) by centrifugation (14000 rpm, 15 min) to remove the unloaded free SN-38. To analyze the SN-38 loading efficiency, supernatants from each washing step were analyzed by measuring the fluorescence intensity spectra spectrofluorophotometer (Shimadzu Corp., Kyoto, Japan) using a 1 cm standard quartz cell (internal volume of 3 mL, Hellma Analytics, Germany) at 25 $^{\circ}\text{C}$. The loading efficiency of SN-38 was determined using the following equation 1. The concentration of the unloaded SN-38 was calculated from the standard fluorescence curve (SN-38 concentration: 0.98–31.25 $\mu\text{g mL}^{-1}$) using OriginPro software (Northampton, MA, USA).⁴

$$\text{Loading efficiency (\%)} = \frac{W_{\text{total SN-38}} - W_{\text{unloaded SN-38}}}{W_{\text{loaded SN-38}} + W_{\text{nanoparticles}}} \times 100 \quad \text{----- (equation 1)}$$

Preparation of SN-38 loaded/F127-coated pSiNPs (named pSiNPs(SN-38)/F127) and SN-38 loaded/ODS-grafted/F127-coated pSiNPs (named pSiNPs(SN-38)-ODS/F127)

For the hydrophilic coating of nanoparticles, pSiNPs(SN-38) or pSiNPs(SN-38)-ODS (~1 mg) was mixed with 2.5% Pluronic® F127 (2 mL) and sonicated in an ultrasonic bath for 3 h. After sonication, the resulting nanoparticles were washed 3 times by centrifugation (14000 rpm, 50 min) with deionized water ($\text{DI}\cdot\text{H}_2\text{O}$, 2 times) and EtOH (1 time). To quantify the amount of released SN-38 through the sonication process, the supernatants from each centrifugation step were analyzed by measuring the fluorescence intensity at 559 nm. The concentration of the released SN-38 was calculated from the standard fluorescence curve using OriginPro software (Northampton, MA, USA).⁵

Characterization of the pSiNPs formulations

Hydrodynamic size and zeta-potential of the pSiNPs formulations (pSiNPs, pSiNPs-ODS, pSiNPs(SN-38), pSiNPs(SN-38)-ODS, pSiNPs(SN-38)/F127, and pSiNPs(SN-38)-ODS/F127) were analyzed by Malvern Instruments Zetasizer Nano ZS90 (Worcester-shire, UK). The structural morphologies of pSiNPs formulations were visualized by transmission electron microscopy (TEM, Tecnai, G2 F30ST, FEI Company, OR, USA) imaging at the Korea Basic Science Center (Korea University, Seoul, Rep. of Korea). Attenuated total reflection Fourier transform infrared (ATR-FTIR) spectroscopy was performed using Thermo Scientific Nicolet™ iS™ 5 FT-IR spectrometer instrument (16 scans, Waltham, MA, USA). Raman spectra were measured using a Ntegra spectra II (NT-MDT spectrum instruments, Moscow, Russia) with a 100 mW 663 nm laser excitation source at the Korea Basic Science Center (Korea University, Seoul, Rep. of Korea).

Drug release rate analysis

Release kinetics of the loaded SN-38 from the pSiNPs formulations (pSiNPs(SN-38), pSiNPs(SN-38)/F127, and pSiNPs(SN-38)-ODS/F127) were measured using a spectro-fluorophotometer at 25 $^{\circ}\text{C}$. The pSiNPs formulations (0.5 mg mL^{-1}) were incubated in the phosphate-buffered saline (PBS) buffer (pH 7.4) at 37 $^{\circ}\text{C}$ for 14 days. At each time interval (0, 3, 6, 12, 24, 48 hours, 3, 4, 5, 6, 7, 8, 9, 10, 12, and 14 days), the pSiNPs formulations were removed from the aqueous phase by centrifugation at 14000 rpm for 15 min, and the fluorescence intensity (559 nm, $\lambda_{\text{ex}} = 364$ nm) of SN-38 left in the aqueous phase was measured using a spectro-fluorophotometer. The concentration of SN-38 remaining in the supernatant was calculated from the standard fluorescence curve using OriginPro software (Northampton, MA, USA).

Degradation rate analysis

The degradation of pSiNPs(SN-38)/F127 and pSiNPs(SN-38)-ODS/F127 was monitored by transmission electron microscopy (TEM) and the Attenuated total reflection Fourier transform infrared (ATR-FTIR) spectroscopy. Porous silicon nanoparticle samples (0.5 mg mL^{-1}) were dispersed in PBS (pH 7.4) solution at $37 \text{ }^{\circ}\text{C}$ for up to 14 days and then removed from the aqueous phase by centrifugation at 14000 rpm for 15 min at each time interval (0, 6, 24 hours, 7, and 14 days). TEM analysis porous silicon nanoparticle samples dispersed in EtOH (0.1 mg mL^{-1}) were applied to a copper grid before observation and dried for 1 day. ATR-FTIR analysis: nanoparticle samples (0.5 mg) dried with a freeze dryer for 1 day before measurement. Raman spectra analysis: pSiNPs/F127 and pSiNPs-ODS/F127 (1 mg mL^{-1} , without SN-38 for Raman spectra analysis) were incubated in PBS (pH 7.4) solution at $37 \text{ }^{\circ}\text{C}$ for 24 h and then collected by centrifugation at 14000 rpm for 15 min. Nanoparticle samples were dried using a freeze dryer for 1 day before measurement. X-ray diffraction (XRD) spectra analysis: pSiNPs/F127 and pSiNPs-ODS/F127 (1 mg mL^{-1}) were incubated in PBS (pH 7.4) solution at $37 \text{ }^{\circ}\text{C}$ for 24 h and then collected by centrifugation at 14000 rpm for 15 min. nanoparticle samples were dried using a freeze dryer for 1 day before measurement.

Cell culture

The HeLa cell lines used in this study were obtained from the American Type Culture Collection (ATCC, Manassas, VA, USA). The cells were placed in 5% CO_2 in Dulbecco's modified Eagle's medium (DMEM) at $37 \text{ }^{\circ}\text{C}$ and supplemented with 10% (v/v) fetal bovine serum, 1% (v/v) penicillin, and streptomycin.

Cell cytotoxicity analysis of the pSiNPs formulations and SN-38

The cytotoxicity of the pSiNPs formulations (pSiNPs, pSiNPs(SN-38), pSiNPs(SN-38)/F127, pSiNPs(SN-38)-ODS/F127) and SN-38 against the HeLa cells were analyzed using Cell-Counting Kit-8 (CCK-8) assays according to the manufacturer's instructions. The cells (5×10^4 cells per well, $500 \text{ }\mu\text{L}$) were seeded on 24-well plates and incubated for 24 h at $37 \text{ }^{\circ}\text{C}$ in a humidified 5% CO_2 incubator. The cells were then treated with 1 and $10 \text{ }\mu\text{M}$ SN-38 concentrations of the nanoparticles, and the cell toxicity was measured after 1, 2, 3, and 5 days of incubation. After incubation, $500 \text{ }\mu\text{L}$ of DMEM (10% WST-1) was added to each well, followed by a further 1 h incubation period. The reacted solution in the 24-well plate was transferred to a 96-well plate ($100 \text{ }\mu\text{L well}^{-1}$). Absorbance was measured at 450 nm using a microplate reader (Multiskan FC, Thermo Fisher, Waltham, MA, USA).

Animals

BALB/c nu/nu mice (female, five-week-old) were purchased from Daehan Biolink Co., Ltd. (Eumseong, Rep. of Korea). The mice were housed 5 per cage ($20 \times 26 \times 13 \text{ cm}$) with free access to food and water in the room that was kept at an ambient temperature of $23 \pm 1 \text{ }^{\circ}\text{C}$ and relative humidity of $60 \pm 10\%$ under a 12/12 h light/dark cycle. All of the experiments performed with mice were carried out under the approved guidelines by the National Institute of Health Guide for the Care and Use of Laboratory Animals (NIH publications No. 80-23, revised in 1996) and protocols approved by the Institutional Animal Care and Use Committee of Kyung Hee University for each experiment (KHSASP-20-224).

Drug release monitoring in a xenograft mouse model

5-week-old mice were stored for 4 weeks, and the HeLa cells (4.5×10^6 cells, suspended in a $200 \text{ }\mu\text{L}$ mixture of PBS and Matrigel (1 : 1, v/v)) were subcutaneously injected behind the flank of each mouse when 9-weeks-old. The tumor size reached approximately 259.8 mm^3 after 3 weeks, and the mice were then randomly divided into 3 groups ($n=3$) for the study. PBS (control), pSiNPs(SN-38)/F127, and pSiNPs(SN-38)-ODS/F127 (dose: 20 mg kg^{-1}) were directly injected into the tumor site. After 6 hours, all the mice were sacrificed, and the tumors were excised. The excised tumors were immersed in a pH 12 buffer (1 h) and fixed with 4% paraformaldehyde followed by washing

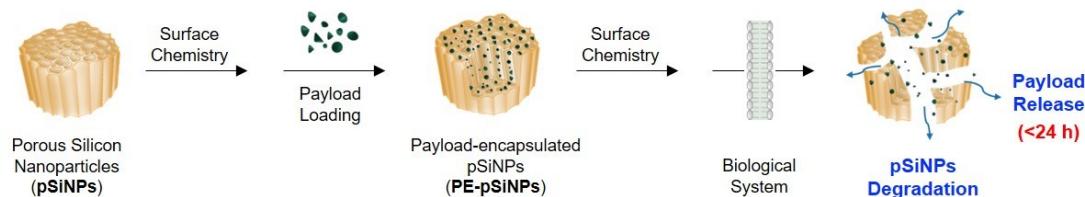
in PBS (pH 7.4).^{6, 7} The fluorescence intensity of SN-38 in tissue samples was measured with an ex vivo fluorescence tissue imaging system (FTIS, VISQUE® InVivo Elite, Vieworks Co., Ltd., Rep. of Korea) to analyze the amount of SN-38 remaining in the tumor. GFP excitation filter (λ_{ex} : 390–490 nm) and the GFP emission filter (λ_{em} : 500–550 nm) were used for the signal collection.

Statistical analysis

The cell cytotoxicity experimental results were expressed as mean \pm standard deviation (S.D.). Data were analyzed using one-way ANOVA (for multiple comparisons) followed by Newman-Keuls's multiple comparison test. All statistical analyses were performed with Prism 8.0.1 software (GraphPad, La Jolla, CA, USA). **** $p < 0.0001$, when compared with pSiNPs. The data of the HeLa tumor FTIS results are presented as mean \pm S.D. The results of the ex vivo imaging data were analyzed by one-way analysis of variance (ANOVA) with the Newman-Keuls's multiple comparison test. All statistical analyses were performed with Prism 8.0.1 software (GraphPad, La Jolla, CA, USA). (* $p < 0.05$, ** $p < 0.01$, when compared to the PBS group, # $p < 0.05$, when compared to the pSiNPs(SN-38)/F127 group.) [F (2, 6) = 20.60, P = 0.0021]. Relative fluorescence intensity plot of SN-38 from tumors experimental results were presented as means \pm standard deviation. Data were analyzed by significance testing and Student's t-test was used for data analysis. The analysis was carried out using SPSS 20 (IBM Corp., USA). ** $p \leq 0.01$, compared with the pSiNPs(SN-38)/F127 control.

2. Supporting Figures

(a) pSiNPs-based drug delivery systems (DDS)



(b) Typical surface functionalization methods

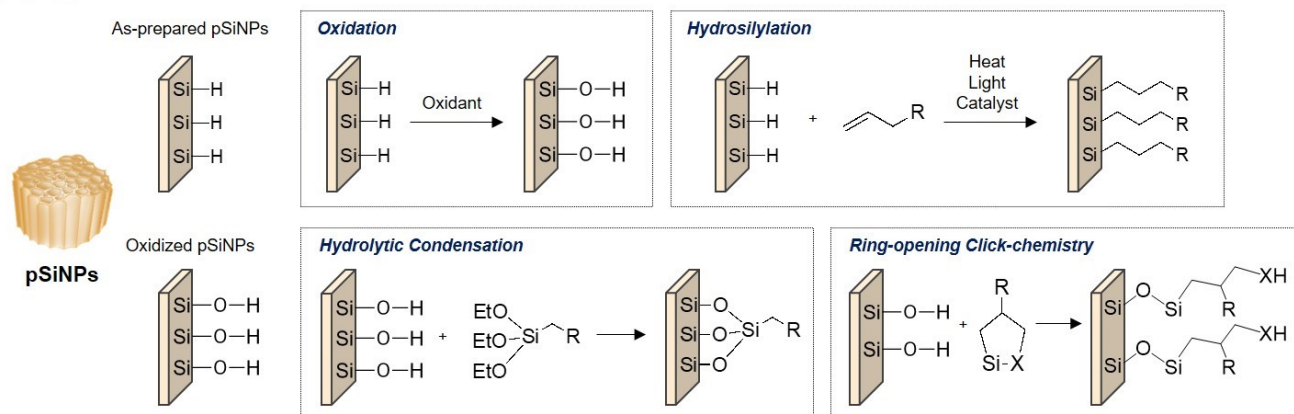


Fig. S1. (a) Typical pSiNPs-based drug delivery systems. The systems were prepared through surface chemistry and payload loading process (payload-encapsulated pSiNPs; PE-pSiNPs). The payload could be typically released from pSiNPs within 24 h in biological systems via particle degradation. (b) Typical surface functionalization methods of pSiNPs. As-prepared pSiNPs have silicon hydride (Si-H) functionality. (i) Oxidation: Si-H on the surface of pSiNPs was oxidized by treating with oxidants (DI-H₂O, base, etc.) and the oxidized silicon surfaces (Si-OH) were formed. (ii) Hydrosilylation: Si-H on the surface of as-prepared pSiNPs reacted with unsaturated carbons such as alkene (C=C) in a specific environment (high temperature, light, catalyst, etc.). (iii) Hydrolytic condensation: Si-OH of oxidized pSiNPs were reacted with organo-silane reagents (X₃-Si-R, X=methoxy, ethoxy) and generated a new Si-O-Si bond. (iv) Ring-opening click chemistry: Si-OH of oxidized pSiNPs reacted with 5-membered heterocyclic silane reagents (Si-X (X=S, N)) and generated a new Si-O-Si bond.

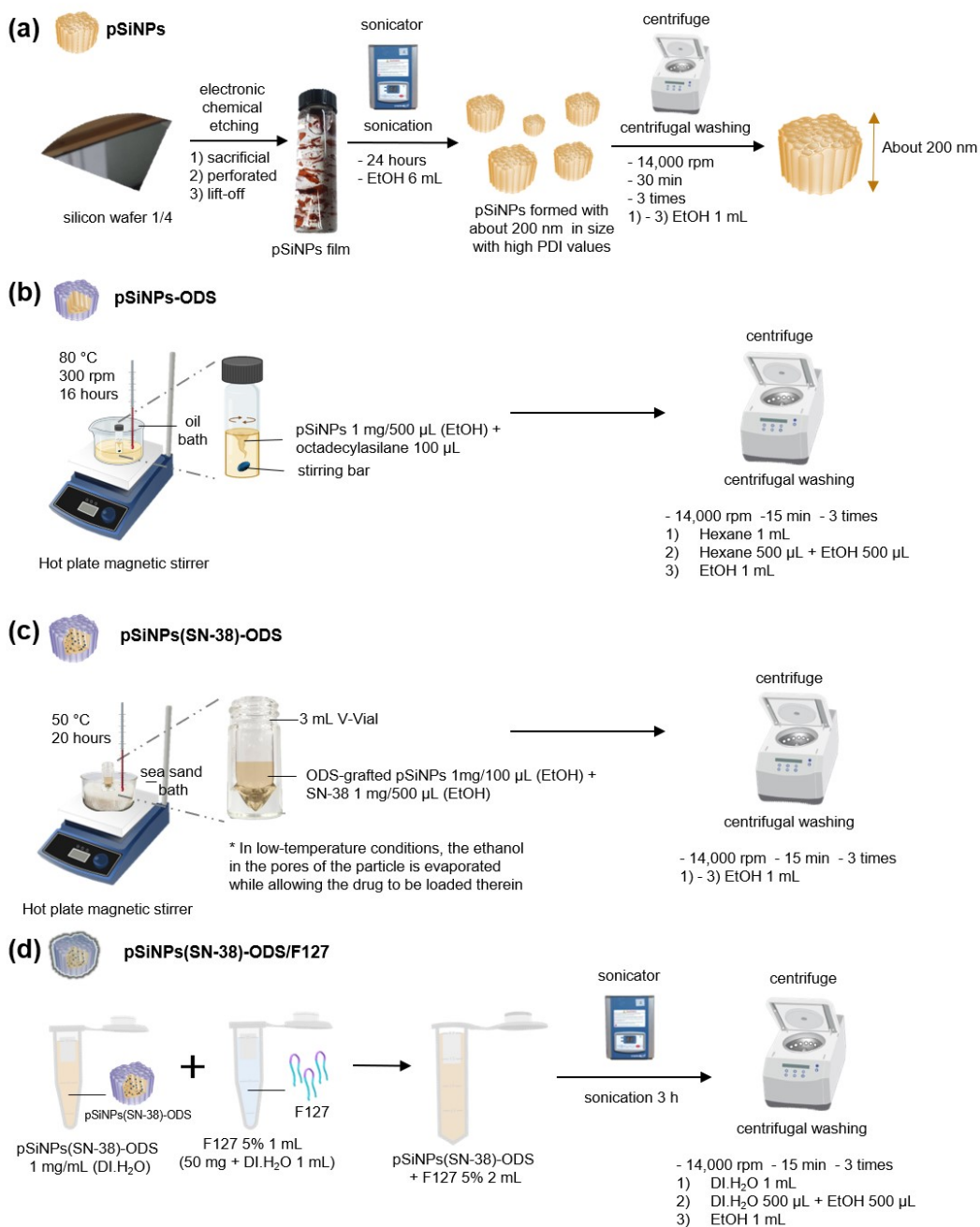


Fig. S2. Schematic diagram of (a) pSiNPs, (b) pSiNPs-ODS, (c) pSiNPs(SN-38)-ODS, and (d) pSiNPs(SN-38)-ODS/F127. Etching conditions for pSiNPs: (i) 44 mA cm^{-2} for 60 s. (ii) perforated etching: 44 mA cm^{-2} for 1.8 s, 321 mA cm^{-2} for 0.4 s. (iii) lift-off: 3 mA cm^{-2} for 60 s. Hexane: *n*-hexane. SN-38: 7-Ethyl-10-hydroxycamptothecin. F127: Pluronic F127.

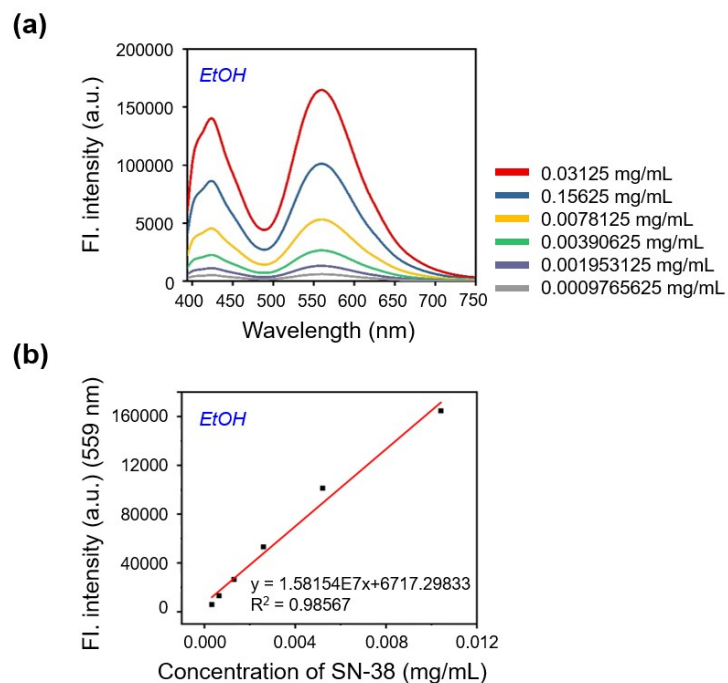


Fig. S3. (a) Fluorescence intensity spectra of SN-38 ($0.0009765625\text{--}0.03125\text{ mg mL}^{-1}$) in ethanol. (b) Concentration-dependent fluorescence intensity plot of SN-38 at 559 nm. Excitation wavelength: 384 nm. Fluorescence intensity of SN-38 (λ_{em} : 559 nm, solvent: ethanol) was linear plotted using OriginPro software, $R^2=0.98567$.

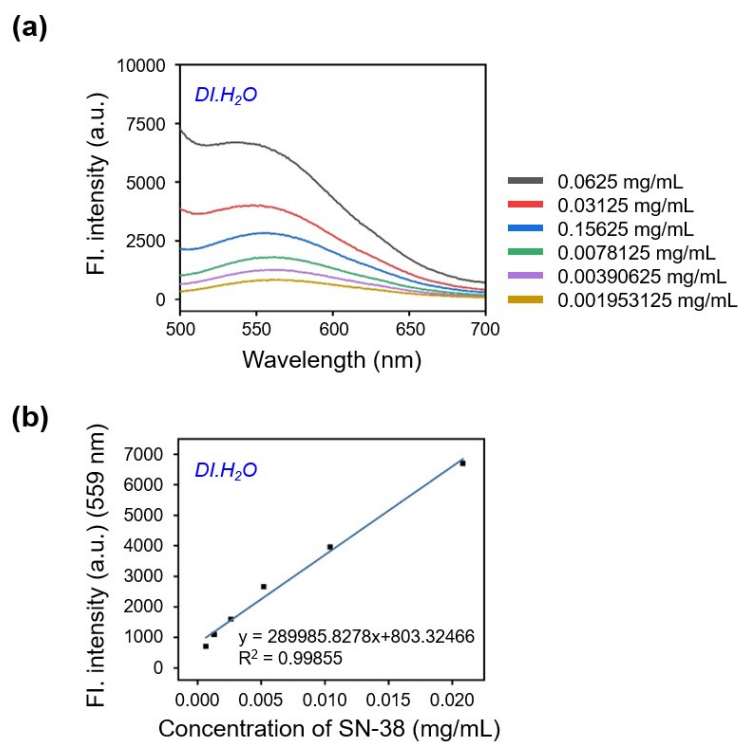


Fig. S4. (a) Fluorescence intensity spectra of SN-38 ($0.001953125\text{--}0.0625\text{ mg mL}^{-1}$) in deionized water ($\text{DI}\cdot\text{H}_2\text{O}$). (b) Concentration-dependent fluorescence intensity plot of SN-38 at 559 nm. Excitation wavelength: 364 nm. Fluorescence intensity of SN-38 (λ_{em} : 559 nm, solvent: $\text{DI}\cdot\text{H}_2\text{O}$) was linear plotted using OriginPro software, $R^2=0.99855$.

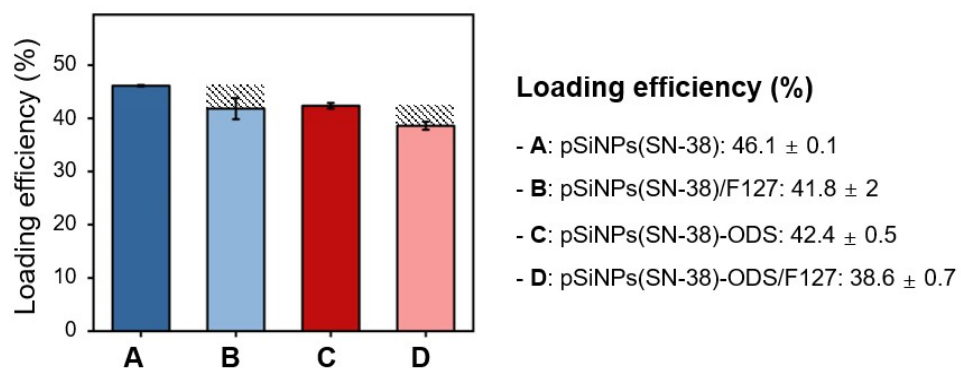
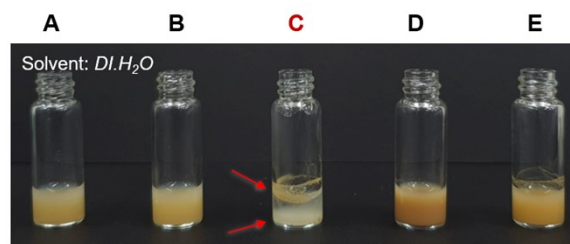


Fig. S5. Loading efficiency of SN-38 in pSiNPs(SN-38), pSiNPs(SN-38)/F127, pSiNPs(SN-38)-ODS, and pSiNPs(SN-38)-ODS/F127.



- A: pSiNPs
- B: pSiNPs(SN-38)
- C: pSiNPs(SN-38)-ODS
- D: pSiNPs(SN-38)/F127
- E: pSiNPs(SN-38)-ODS/F127

Fig. S6. Colloidal stability of pSiNP nanoformulations (pSiNPs, pSiNPs(SN-38), pSiNPs(SN-38)-ODS, pSiNPs(SN-38)/F127, and pSiNPs(SN-38)-ODS/F127) in $DI-H_2O$. The red arrows indicated aggregated nanoformulations.

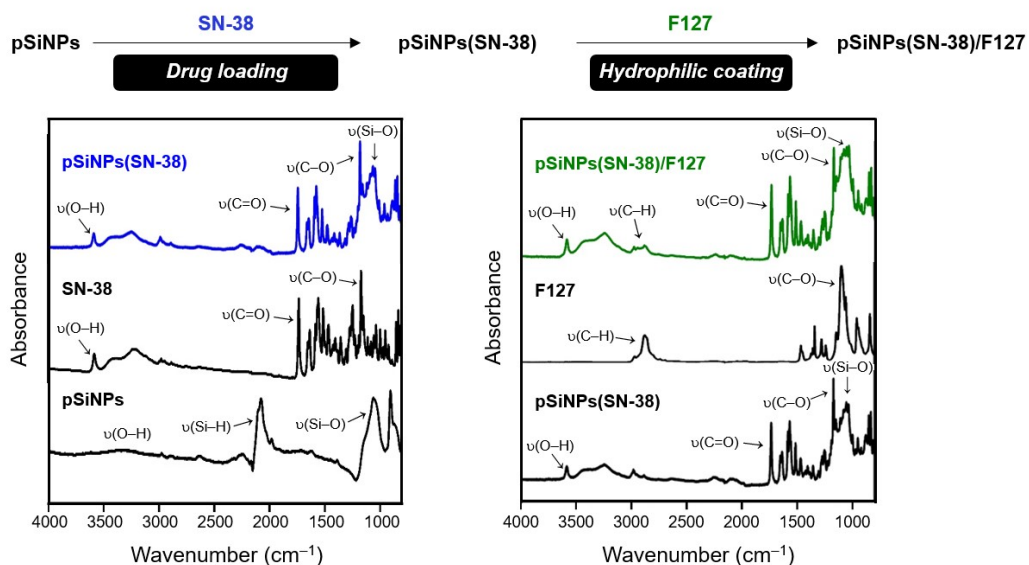


Fig. S7. Attenuated total reflectance Fourier-transform infrared (ATR-FTIR) spectra of the pSiNP, pSiNPs(SN-38), and pSiNPs(SN-38)/F127. Symbols: ν = stretching. (i) pSiNPs showed three characteristic peaks at $3200\text{--}3500\text{ cm}^{-1}$ ($\nu(\text{O-H})$), 2087 cm^{-1} ($\nu(\text{Si-H})$), and 1033 cm^{-1} ($\nu(\text{Si-O})$). (ii) SN-38 had three bands at 3590 cm^{-1} ($\nu(\text{O-H})$), 1750 cm^{-1} ($\nu(\text{C=O})$), and 1170 cm^{-1} ($\nu(\text{C-O})$). (iii) F127 displayed two peaks at 2882 cm^{-1} ($\nu(\text{C-H})$) and 1100 cm^{-1} ($\nu(\text{C-O})$).

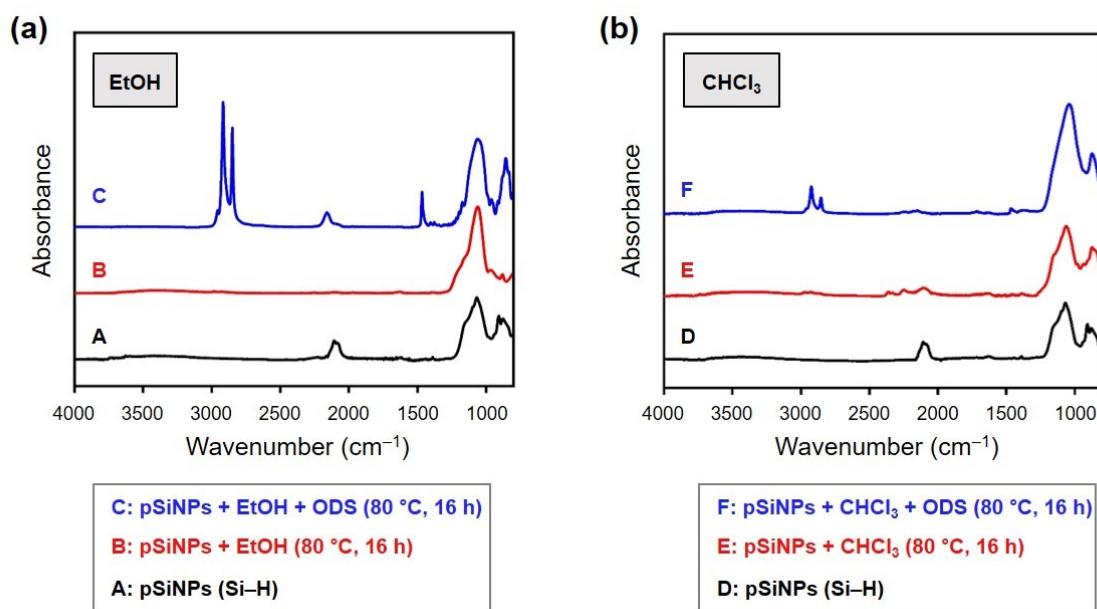


Fig. S8. Attenuated total reflectance Fourier-transform infrared (ATR-FTIR) spectra of the pSiNPs particles that prepared via dehydrocoupling reaction within (a) ethanol (EtOH) and (b) chloroform (CHCl₃). A&D: as-prepared pSiNP, B&E: reaction without ODS, and C&F: reaction with ODS.

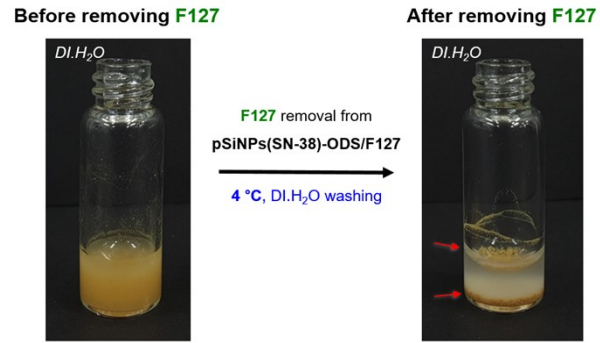


Fig. S9. Verification of F127 coating on the pSiNPs nanoformulations. Before removing the F127, the pSiNPs(SN-38)-ODS/F127 showed high colloidal stability in DI·H₂O (left). After removing the F127, the nanoformulations aggregated and settled on the bottom. The red arrows indicated aggregated nanoformulations.

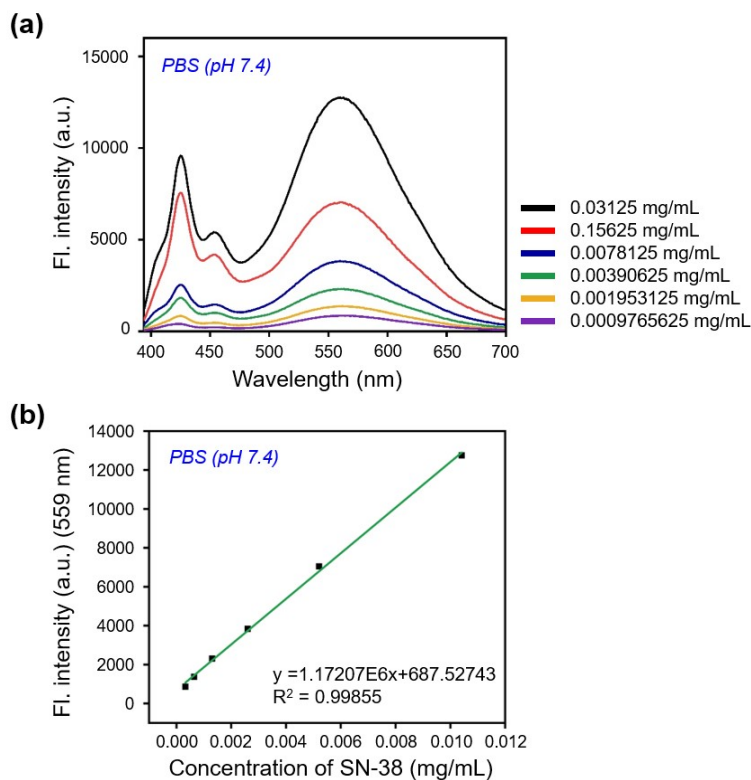
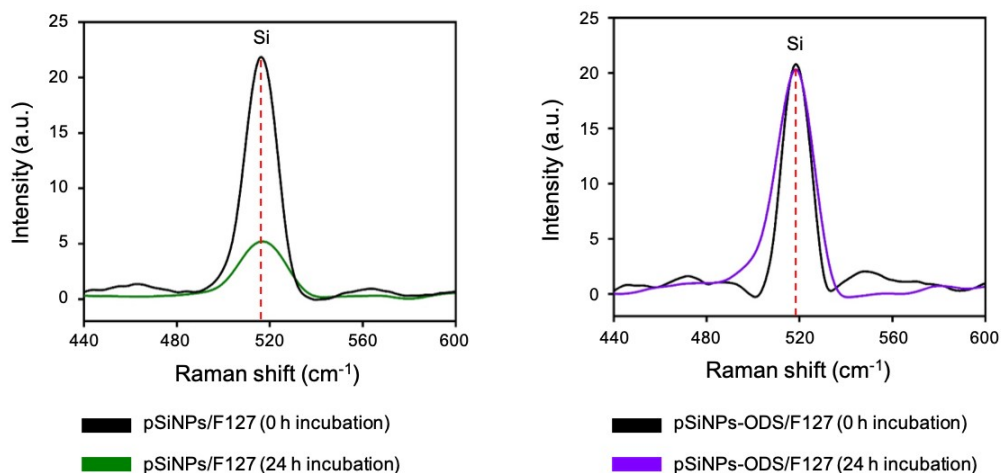


Fig. S10. (a) Fluorescence intensity spectra of SN-38 (0.0009765625–0.03125 mg mL⁻¹) in PBS buffer (pH 7.4). (b) Concentration-dependent fluorescence intensity plot of SN-38 at 559 nm. Excitation wavelength: 364 nm. Fluorescence intensity of SN-38 (λ_{em} : 559 nm, solvent: pH 7.4 PBS) was linear plotted using OriginPro software, $R^2=0.99855$.

(a) Raman spectra



(b) XRD spectra

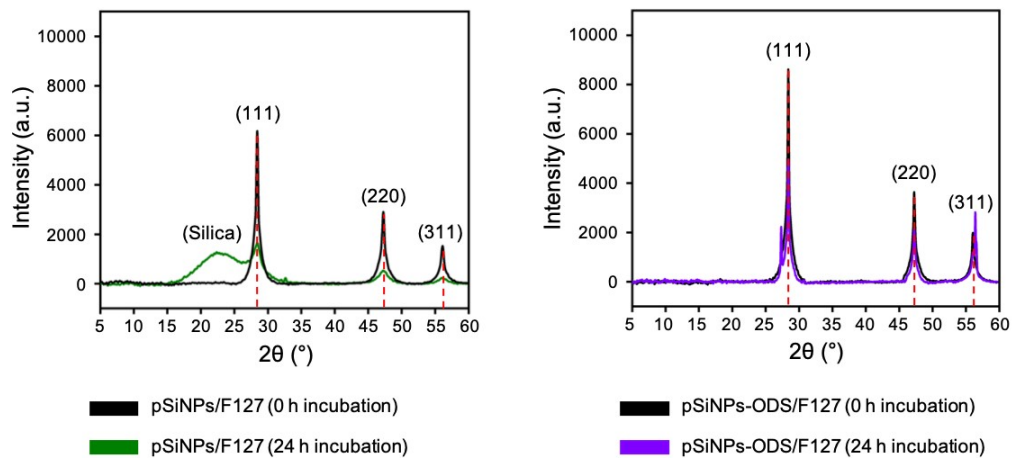


Fig. S11. (a) Raman spectra of pSiNPs/F127 and pSiNPs-ODS/F127 obtained before incubation (0 h) and after incubation (24 h). Solvent: pH 7.4 PBS. Si lattice mode: 515 cm^{-1} . (b) X-ray diffraction (XRD) spectra of pSiNPs/F127 and pSiNPs-ODS/F127 powders obtained before incubation (0 h) and after incubation (24 h) in PBS (pH 7.4) at 37 $^{\circ}\text{C}$.

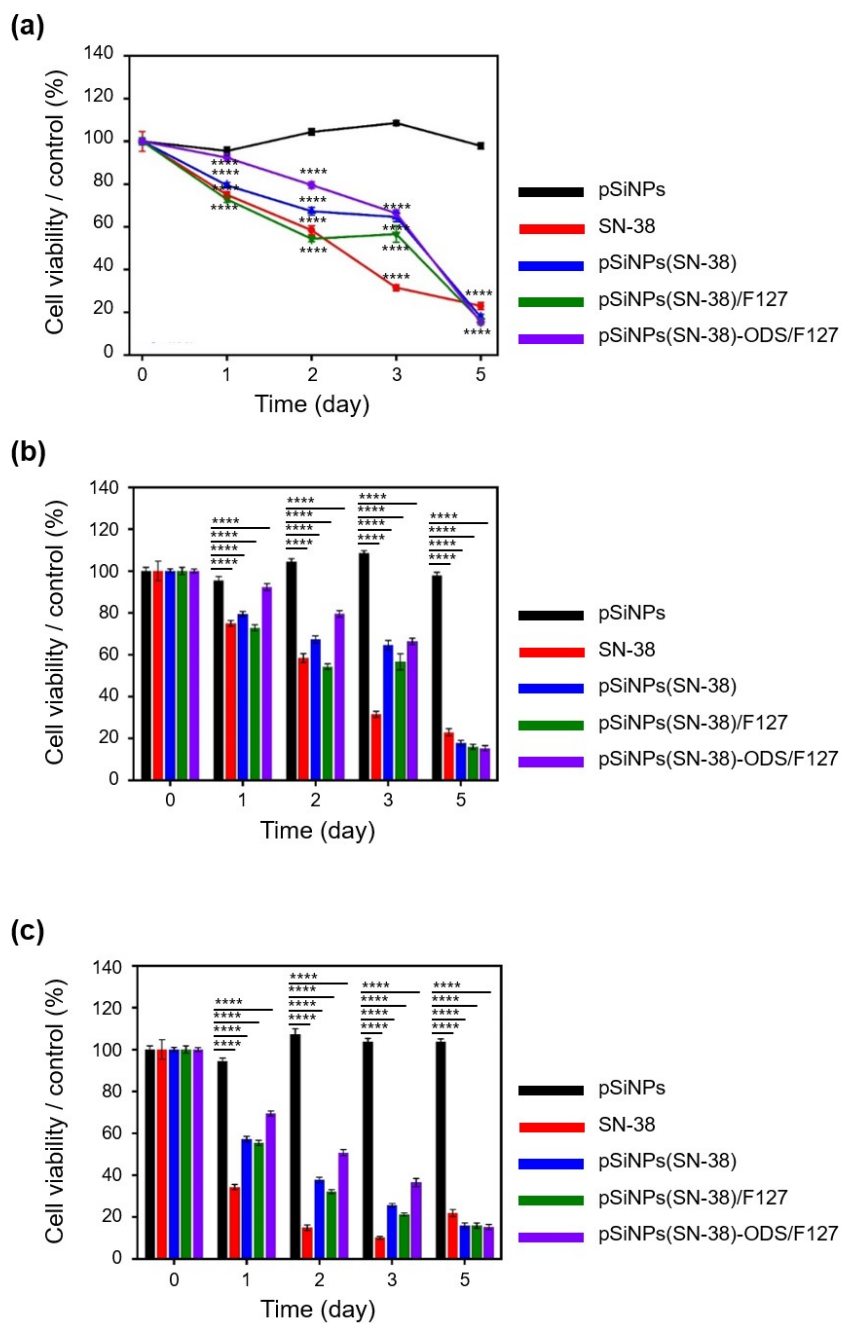


Fig. S12. (a, b) Cell viability of HeLa cells after treatment of 1 μM of SN-38 with incubation at 37 $^{\circ}\text{C}$ for 5 days. (c) Cell viability of HeLa cells after treatment of 10 μM of SN-38 with incubation at 37 $^{\circ}\text{C}$ for 5 days. Experimental results were expressed as mean \pm standard deviation (S.D.). Data were analyzed by significance testing using one-way ANOVA (for multiple comparisons) followed by Newman-Keuls's multiple comparison test. All statistical analyses were performed with Prism 8.0.1 software (GraphPad, La Jolla, CA, USA). **** $p < 0.0001$, when compared with pSiNPs.

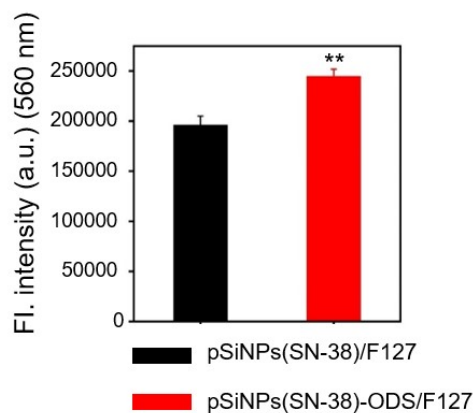


Fig. S13. Relative fluorescence intensity plot of SN-38 from tumors (n=3) after 6 h circulation of pSiNPs(SN-38)/F127 and pSiNPs(SN-38)-ODS/F127 (intratumoral injection with 20 mg kg⁻¹). Solution: pH 12 buffer. Excitation wavelength: 412 nm. The values in this graph were derived from the emission intensity of SN-38 in different buffers (pH 7 and pH 12, see Fig. S9–S11). Experimental results were presented as means \pm standard deviation (S.D.). Data were analyzed by significance testing and a Student's t-test was employed for data analysis. The analysis was carried out using SPSS 20 (IBM Corp., USA). ** $p \leq 0.01$, compared with pSiNPs(SN-38)/F127 control.

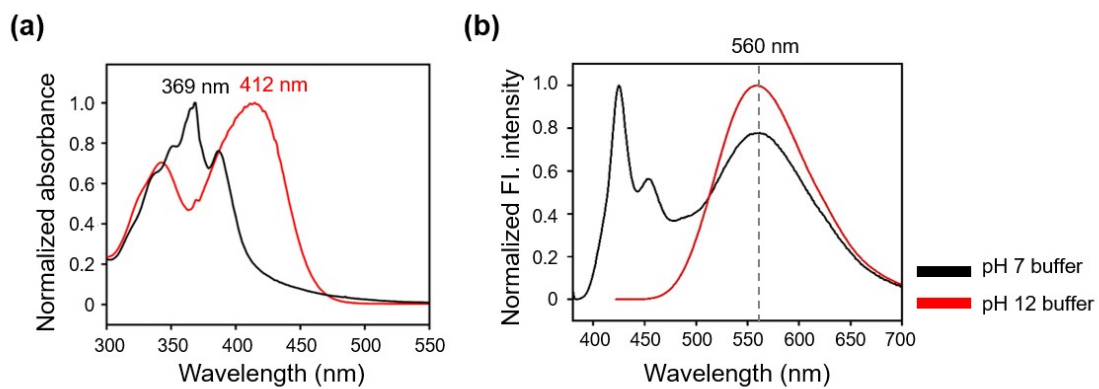


Fig. S14. (a) Normalized absorption and (b) emission spectra of SN-38 in pH 7 and 12 buffer. Excitation wavelength: (a) 369 nm, (b) 412 nm.

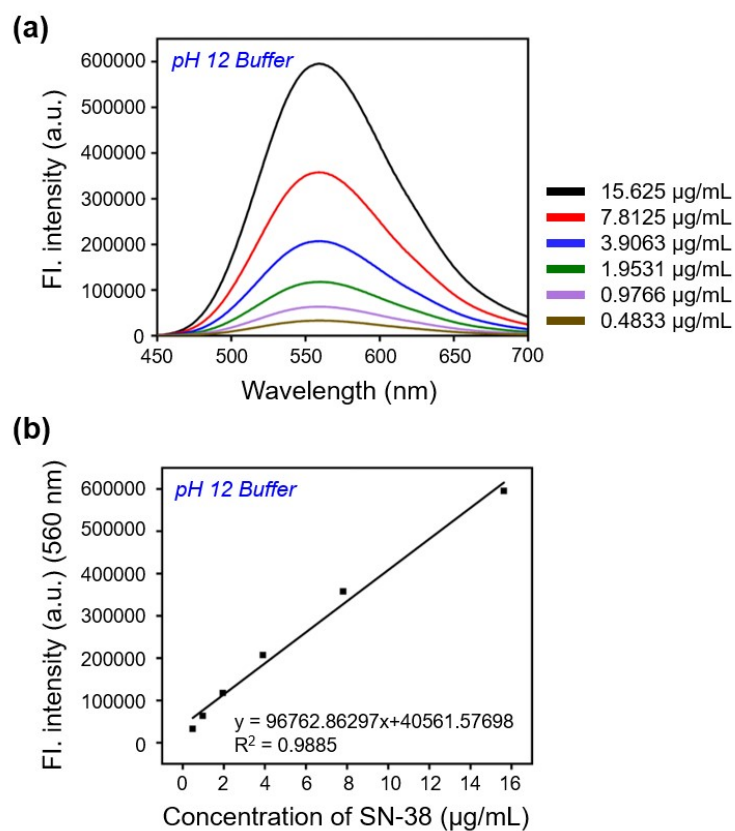


Fig. S15. (a) Fluorescence intensity spectra of SN-38 (0.4833–15.625 $\mu\text{g kg}^{-1}$) in pH 12 buffer. (b) Concentration-dependent fluorescence intensity plot of SN-38 at 560 nm. Excitation wavelength: 412 nm. Fluorescence intensity of SN-38 (λ_{em} : 559 nm, solvent: pH 12 buffer) was linear plotted using OriginPro software, $R^2=0.9885$.

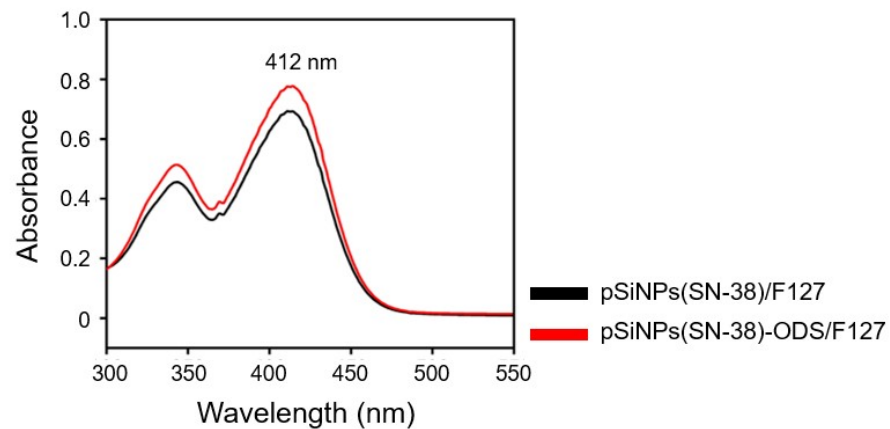


Fig. S16. Absorption spectra of SN-38 at pH 12 buffer extracted from tumors after the intratumoral injection of pSiNPs(SN-38)/F127 and pSiNPs(SN-38)-ODS/F127, respectively. Injection concentration of particles: 20 mg kg⁻¹, circulation time: 6 h, dissected tumor (size: 259.8 mm³) incubation time in pH 12 buffer: 72 h. See the experimental details in Method Section.

3. Supporting Tables

Table S1. Hydrodynamic size/PDI and zeta-potential value of nanoparticle formulations (pSiNPs, pSiNPs-ODS, pSiNPs(SN-38), pSiNPs(SN-38)-ODS). The values were measured in EtOH. Each experiment was performed in quintuplicate to determine the mean and standard deviation (S.D.).

Solvent	EtOH			
Sample	pSiNPs	pSiNPs-ODS	pSiNPs(SN-38)	pSiNPs(SN-38)-ODS
DLS (PDI)	276.0 ± 128.1 nm (0.251)	314.5 ± 134.5 nm (0.237)	281.2 ± 100.8 nm (0.436)	367.1 ± 108.4 nm (0.387)
Zeta-potential (mV)	-23.3 ± 11.2	-35.7 ± 11.9	-31.6 ± 14.9	-32.1 ± 12.8

Table S2. Hydrodynamic size/PDI and zeta-potential value of nanoparticle formulations (pSiNPs, pSiNPs(SN-38)/F127, pSiNPs(SN-38)-ODS/F127). The values were measured in DI·H₂O. Each experiment was performed in quintuplicate to determine the mean and standard deviation (S.D.).

Solvent	DI·H ₂ O		
Sample	pSiNPs	pSiNPs(SN-38)/F127	pSiNPs(SN-38)-ODS/F127
DLS (PDI)	218.0 ± 82.6 nm (0.240)	442.1 ± 76.35 nm (0.270)	364.4 ± 69.54 nm (0.118)
Zeta-potential (mV)	-36.3 ± 6.17	-24.3 ± 6.01	-26.3 ± 6.08

4. References

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