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Supporting Information

DNA Aptamer Release from DNA-SWNT Hybrid by Protein Recognition

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Supporting information 1. Methods

1.1 Preparation of aptamer-SWNT hybrids

The SWNTs synthesized with the high-pressure carbon monoxide conversion (HiPco2714 process (Carbon Nanotechnologies Inc., TX USA) were used as a template for the wrapping of single-strand DNA aptamer. 0.1 μg of SWNT was introduced to 100 μM aptamer solution of 1 ml. The sequence of aptamer was 5'-GGTTGGTGTGGTTGG-3'. The formation of ssDNA-SWNT hybrids was carried out under the sonication (SONICS, SONICS&MATERIALS, INC.). Then the solution was centrifuged at 15,000 rpm for 60 min and the precipitated SWNTs which might not react with the aptamer were removed.

The centrifugation for the purification was repeated twice. The SWNTs which were completely wrapped by the aptamer were obtained through these steps, which was confirmed by UV absorbance measurement.

1.2 Reaction of aptamer-SWNT hybrids with protein

DNA aptamer can bind to specific target protein with high affinity and specificity. Mostly aptamer reacts with protein under condition of free aptamer. But in aptamer-SWNT hybrids, the aptamer is wrapped onto SWNT and not free condition. The aptamer used in this experiment is thrombin binding aptamer. When we supplied the thrombin in hybrids solution, aggregates of SWNT occurred. This means aptamer is released onto SWNT and aggregates of SWNT is shown due to hydrophobicity of SWNT. In other words, the binding between thrombin and aptamer is stronger than that between SWNT and aptamer. Aptamer on hybrid is released in order to react with thrombin. Therefore, we can see aggregates of SWNT.

1.3 Characterization of DNA-SWNT hybrids before and after reaction with protein

UV/Vis/NiR measurements were done using Lambda 19 Spectrometer (PerkinElmer). Circular dichroic spectra were measured with Chirascan plus (Applied Photophysics). For Raman measurement, a micro-Raman system (Jobin-Yvon, LabRam HR) was used with laser lines at a wavelength of 514.5 nm from an Ar-ion laser for excitation.

1.4 Computational details

We carried out molecular dynamics (MD) simulations using the Large-scale Atomic/Molecular Massively Parallel Simulator (LAMMPS) program which is a classical MD code. The single-stranded DNA aptamer with the sequence:5’-GGTTGGTGTGGTTGG-3’ and thrombin molecule were described by the CHARMM27-all

atom force field. The (6,6) metallic SWNT containing 1200 carbon atoms (12 nm long and 0.8 nm in diameter)was used in our simulation. Carbon atoms in the SWNT were described by the Lennard-Jones potential with the parameters: σ_{cc}= 3.4Å and ε_{cc} = 0.086kcal·mol^{-1}.Carbon-carbon bonds and bond angles were modeled by simple harmonic potentials with the equilibrium distance of 1.4Å and the spring constant of 938 kcal·mol^{-1}·Å^{-1} and the equilibrium angle of 120° and the spring constant of 126 kcal·mol^{-1}·radian^{-2}, respectively. In each MD simulation, energy minimization atomic configurations were obtained by potential energy minimization steps and the time step was 1.0 fs. To mimic actual experimental environments, TIP3P water molecules of 1g/ml density and Na^+ counterions were introduced in a simulation box. We employed the Nose-Hoover-type non-Hamiltonian equation of motion to keep system temperature and pressure as 300 K and 1 atm (= 101,325 Pa). The particle-particle particle-mesh (PPPM) method [6] was adopted to calculate the long-range electrostatic interaction energy.

Supporting information 2. UNrwapping efficiency of DNA aptamer

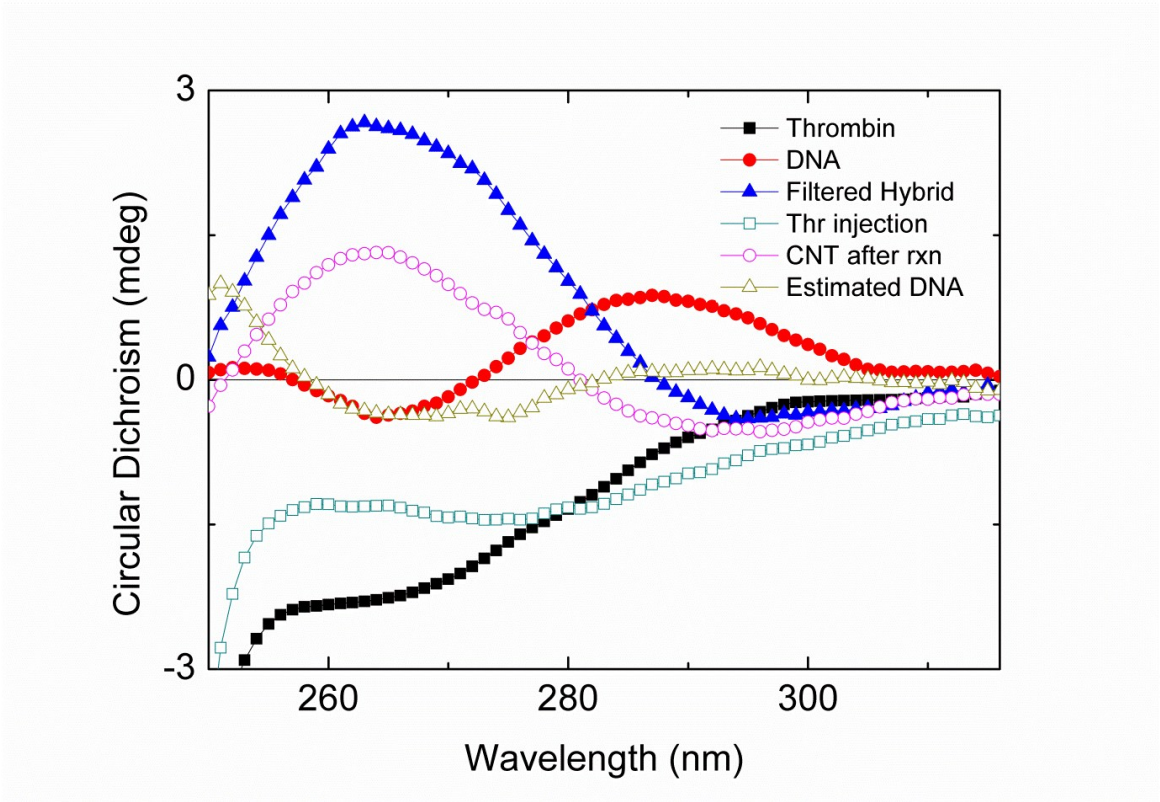

Figure S1 CD spectra of thrombin and same amount of aptamer used in wrapping, hybrid, mixture of hybrid and thrombin, and SWNT harvested by centrifugation after reaction of hybrid and thrombin.

Circular dichroism (CD) spectroscopy is a tool used widely for structural studies of small organic molecules, proteins, and DNAs. Also, value amplitude of CD is proportional to the sample concentration. So, we used CD for detection of wrapping and unwrapping of DNA onto SWNT. We measured the CD spectra of thrombin and same amount of aptamer used in wrapping, hybrid, mixture of hybrid and thrombin, and SWNT harvested by centrifugation after reaction of hybrid and thrombin. The signal value of estimated aptamer released from hybrid by reaction with thrombin is calculated by subtracting the signal value of thrombin, SWNT after reaction of hybrid and thrombin from that of mixture of hybrid and thrombin. The signal value of estimated aptamer is less than that of initial signal value of aptamer that

used in wrapping. It means that unwrapping efficiency is low but there could be the way to improve the unwrapping efficiency by external energy source.

Supporting information 3. Quantitative analysis of the reaction of the hybrids and protein

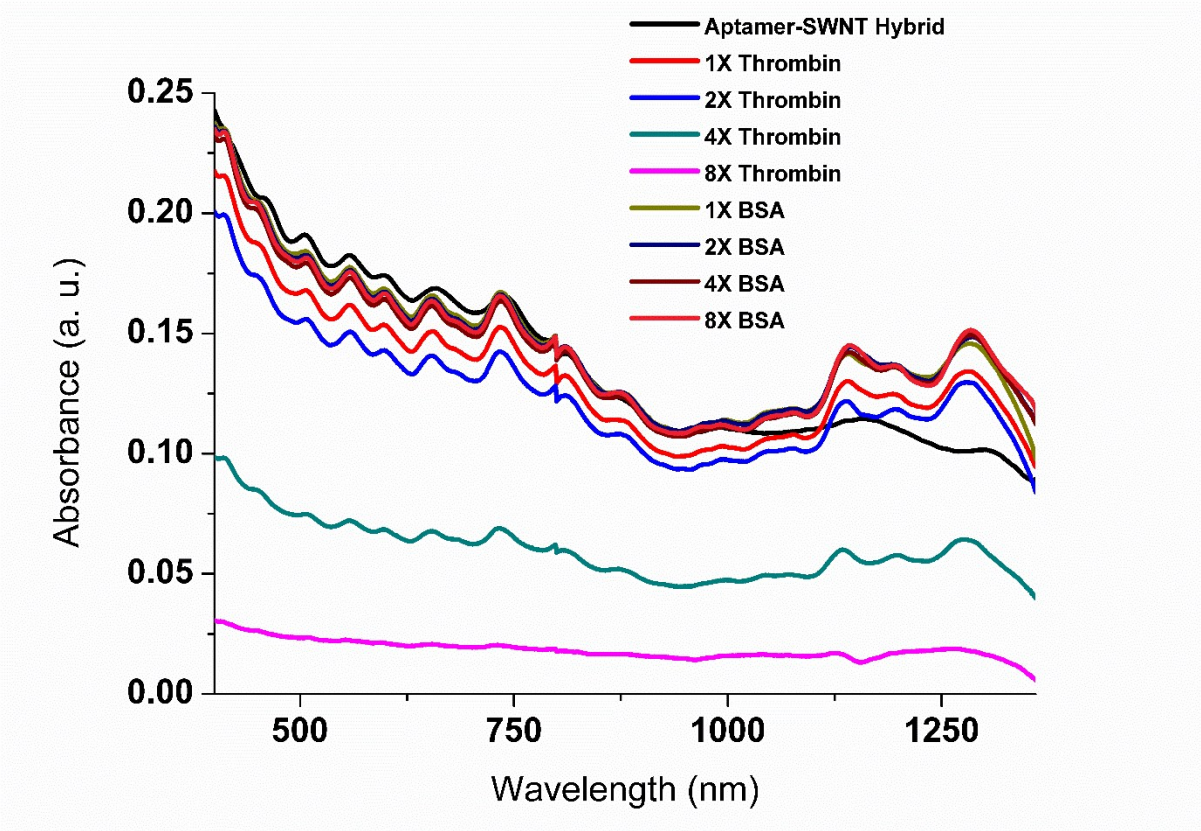

Figure S2 UV/vis/NiR spectra of the aptamer-SWNT hybrids and protein inserted sample. As increasing the concentration of thrombin inserted, the absorbance of the supernatants became lower. However, BSA has no difference according to inserted concentration.

UV/vis/NiR spectrophotometer is a useful tool to characterize quantitative property of the SWNT as shown in Figure S2. After each reaction with different concentrations of thrombin, the supernatants were gathered and measured. The aptamer-SWNT hybrids themselves show the highest spectrum among absorbance spectra. As increasing the concentration of thrombin inserted into the aptamer-SWNT hybrid solution, the spectra of the supernatants become lower, which means that larger amount of the SWNT was deposited at the bottom due to unwrapping by the reaction with thrombin. At higher range than 1000 nm, the hybrid

only shows different peak from the samples in presence of the protein, thrombin or BSA, and its reason is unclear. However, the spectra are almost same regardless of the amount of BSA introduced. Based on this observation, unwrapping specifically occurs and its efficiency depends on the concentration of target protein inserted into the hybrids solution.

Supporting information 4. Raman measurement of the SWNT in the absence with protein

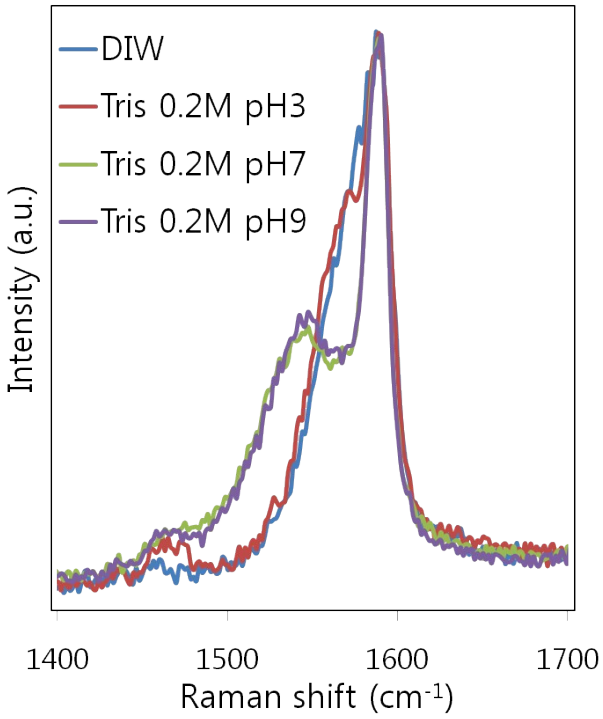

Figure S3 Raman spectrum of aptamer-SWNT hybrid in different buffer condition

Raman spectra of aptamer-SWNT hybrid were observed in different buffer condition (Figure S3). Raman spectra of aptamer-SWNT hybrid show the different lineshape according to the pH. In the acidic condition, hybrid shows the semiconducting Lorentzian line. We can assume that Raman spectra is affected by the pH condition. So, we did the following experiment. We put the thrombin(4mg/ml) or BSA(8mg/ml) in aptamer-SWNT hybrid and after that we changed the buffer condition using diluted HCl from pH 3 to pH 7 and measured the acidic condition using litmus paper and observed the Raman spectra of each of them (Figure S4). In case of hybrid in thrombin, Raman spectra show the Lorentzian lineshape in any condition. The aptamer on hybrid is released by thrombin protein and even

if the buffer condition is change into acidic condition, Raman spectra show the Breit-Wigner-Fano (BWF) lineshape due to the SWNT released from aptamer-SWNT hybrid. But in case of hybrid in BSA, we can observed Raman spectra is changed into semiconducting Lorentzian lineshape from Breit-Wigner-Fano (BWF) lineshape according to the lowering of pH. Because hybrid does not react with BSA, hybrid still remains in solution. So, Raman spectra show the Lorentzian lineshape at acidic condition. In other words, proteins make the solution more basic. And in basic condition, we cannot detect correct Raman spectra by wrapping confirmation between aptamer and SWNT. In acidic condition, we can detect the correct Raman spectra of aptamer-SWNT hybrid even if there is protein in hybrid sample.

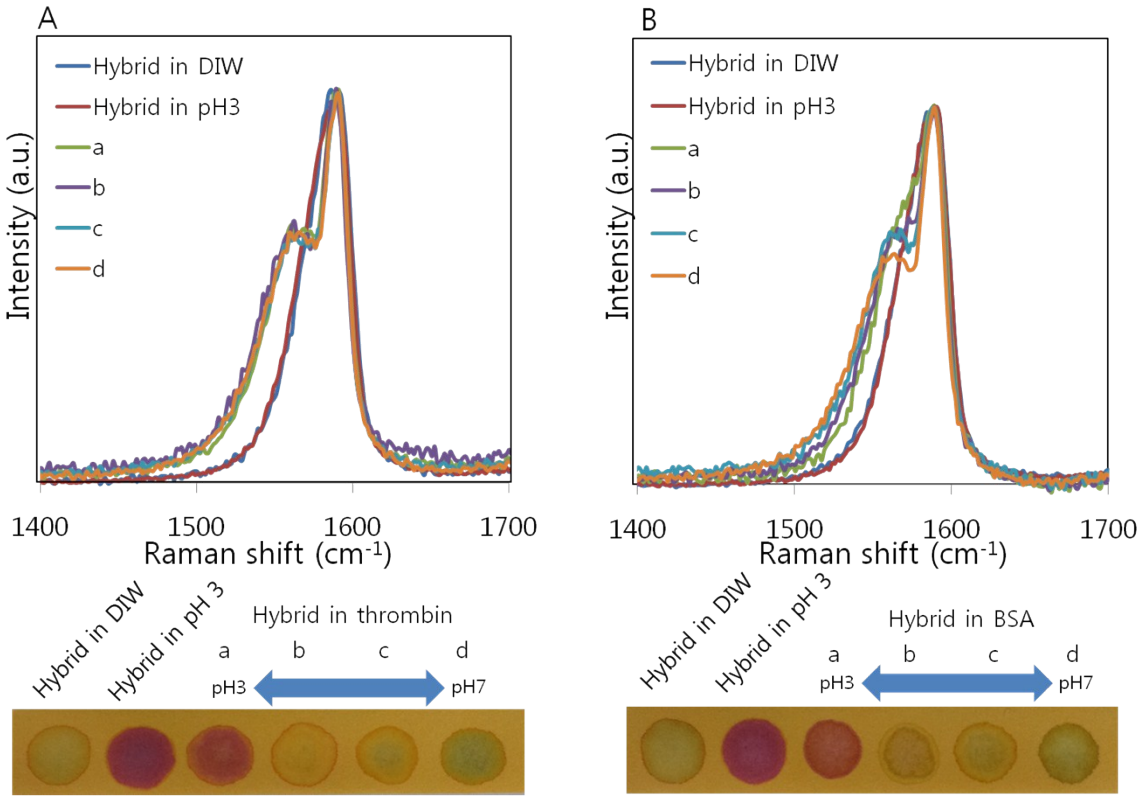

Figure S4 Raman spectra of hybrid in thrombin or BSA in different buffer conditions.

Supporting information 5. Radial Breathing Modes (RBMs) in Raman spectra

Figure S5 shows the Raman spectra of a pristine SWNT. The diameters d of the SWNTs were determined to be 1.1 ± 0.2 nm according to the equation $d = c_1/(\omega_{RBM} - c_2)$, where $\omega_{RBM}$is the frequency of the radial breathing mode (RBM) of the SWNTs, and c_1 = 223.5 cm^{-1} and c_2= 12.5 cm^{-1} have been predetermined for typical SWNTs. The tangential G mode also shows metallic characteristics, with a broad and asymmetric BWF line shape.

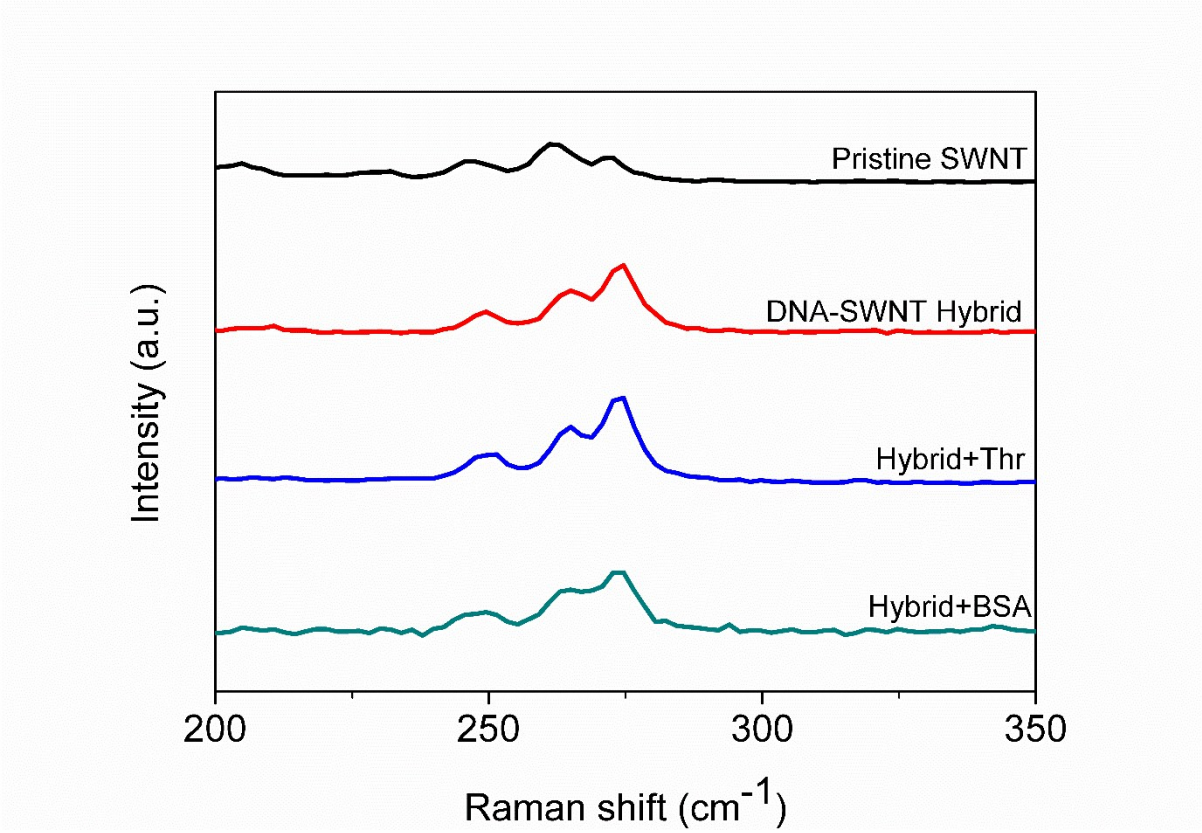

Figure S5 Radial Breathing Mode (RBM) in Raman spectra of pristine SWNT, DNA-SWNT Hybrids, thrombin inserted sample, and BSA inserted sample