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Supporting Information

Supramolecular nanofibrils inhibit cancer progression *in vitro* and *in vivo*

*Yi Kuang, Xuewen Du, Jie Zhou, and Bing Xu**

Materials and methods

Negative stained TEM: Carbon coated grids were glow discharged to increase their hydrophilicity before use. The sample solution (3 μL) was placed on the grid to cover the grid surface. After being rinsed for 10 s, the grid was tilted, allowing the sample-loaded surface to touch a drop of ddH_2O. The edge of the grid was immediately leaned to touch a filter paper for three times to remove water from the grid. The grid was stained by letting the grid touch a drop of 2.0 % (w/v) uranyl acetate with the sample-loaded surface. Excessive stain solution was removed by gently touching the edge of the grid on a filter paper for 3 times. The grid was air dried for a few minutes and was then examined immediately.

Statistical analysis of ITC data: The statistical significance of the ΔH_{dil} at [**1**] = 192 μg/mL to ΔH_{dil} at [**1**] = 144 μg/mL and ΔH_{dil} at [**1**] = 96 μg/mL was examined using Student's t-test. First, the Cohen's d was calculated as follow:

$$d = (\mu 1 - \mu 2) / \sqrt{(\sigma 1^2 + \sigma 2^2)/2}$$

where μ1 and μ2 are the mean values of the two groups and σ12 and σ22 are the SD values of the two groups.

Then, the power was calculated using G*Power 3.1.7:

Input: Tail(s): Two; Effect size: the Cohen's d calculated from above; α err prob: 0.05; Sample size group ΔH_{dil} at [**1**] = 192 μg/mL: 16; Sample size group ΔH_{dil} at [**1**] = 144 μg/mL: 19; Sample size group ΔH_{dil} at [**1**] = 96 μg/mL: 11.

The results are as follow:

	ΔH_{dil} at [**1**] = 192 μg/mL to ΔH_{dil} at [**1**] = 144 μg/mL	ΔH_{dil} at [**1**] = 192 μg/mL to ΔH_{dil} at [**1**] = 96 μg/mL
Noncentrality parameter δ	15.44	3.58
Critical t	2.03	2.06
df	33	25
Power (1-β err prob)	1.00	0.93

The statistical analysis shows that the difference of ΔH_{dil} at [**1**] = 192 μg/mL to ΔH_{dil} at [**1**] = 144 μg/mL and ΔH_{dil} at [**1**] = 96 μg/mL are both significant.

*Intracellular concentration and degradation of **1***: HeLa cells in exponential growth phase were seeded in 35 mm Petri dish at a concentration of 2.0×10^5 cell/dish in MEM medium supplemented with 10% FBS and 1% antibiotic. The cells were allowed to attach to the wells for 24 h at 37 ˚C, 5% CO_2, then the culture medium was removed, and 1 mL new culture medium dissolved with **1** at 144 or 192 μg/mL was placed into each well. After culturing at 37 ˚C, 5% CO_2 for 24 h, the cells were washed by PBS buffer for 3 times then collected using cell scraper in 100μL ddH_2O and 100μL MeOH. The cells were disrupted by freeze-and-thaw for 3 times and centrifuged at 12,000 rpm for 5 min at 4 ˚C. The suspension was analyzed by HPLC to get the molar concentration of **1** and the proteolytic remnant of **1** by standard solutions with known concentration. *R* was calculated as follow: [proteolytic remnant]/[**1**]; n=1. The amount of **1** (M.W. 480 Da) and proteolytic remnant of **1** (M.W. 330 Da) were calculated and presented in μg/mL.

Cell culture and MTT cell viability test: HeLa cells were purchased from the American Type Culture Collection (ATCC, Manassas, VA, USA). The HeLa cells were propagated in Eagle's minimal essential medium (MEM) supplemented with 10% fetal bovine serum (FBS) and antibiotics in a fully humidified incubator containing 5% CO_2 at 37°C. HeLa cells in exponential growth phase were seeded in a 96 well plate at a concentration of 2.0×10^4 cell/well in MEM medium supplemented with 10% FBS and 1% antibiotic. The cells were allowed to attach to the wells for 24 h at 37 ˚C, 5% CO_2, then the culture medium was removed, and 100 μL new culture medium dissolved with **1** at gradient concentrations was placed into each well. After culturing at 37 ˚C, 5% CO_2 for desired time, each well was added by 10 μL of 5 mg/mL MTT ((3-(4,5-Dimethylthiazol-2-yl)-2,5-diphenyltetrazolium bromide), and then incubate in dark for 4 h. 100 μL 10% SDS with 0.01 M HCl was added to each well to stop the reduction reaction. After incubation of the cells at 37 ˚C for overnight, the cell viability is measured. Data represented the mean ± standard deviation of three independent experiments.

HeLa xenograft: The animal care facilities and programs of the Foster Animal Research Facility in Brandeis University meet the requirements of the law and NIH regulations. The animals are subjected to regular veterinary care on a routine basis. The animal experiments described in this study were performed according to the *Guide for the Care and Use of Laboratory Animals* published by the NIH. The nu/nu nude mice were purchased from Charles River Laboratories (Wilmington, MA). HeLa cells were collected in exponential growth phase. 100 μL of HeLa cell suspension in complete culture medium at 5×10^7 cell/mL was subcutaneously injected into the right flank of nu/nu mice. After 30 days of inoculation, mice bearing HeLa tumor at average volume of 30 mm^3 were randomly divided into three groups. Subcutaneous peritumoral injections were performed every three days for six doses. One group was treated with 100 μL of **1** at 2.4 mg/mL in PBS buffer; the other group was treated with 100 μL of **1** at 0.24 mg/mL in PBS buffer; control group was treated with 100 μL PBS

buffer. Based on the average body weight of the mice (24 g) and the molar mass of **1** (480 g/mol), the dosages in gram per kilogram of body weight of 1 on the mice were calculated as follow: 100 μL × 2.4 mg/mL × 480 g/mol / 24 g = 10 mg/kg; 100 μL × 0.24 mg/mL × 480 g/mol / 24 g = 1 mg/kg. The experiments were performed twice. The first test includes: 4 mice for the 10 mg/kg treated group; 4 mice for the 1 mg/kg treated group; 3 mice for the control group. The second test includes: 5 mice for the 10 mg/kg treated group; 3 mice for the control group. The statistic significance in relative tumor size between the control group and the group treated by 10 mg/kg of **1** was examined by Student's t test.

Table S1. DichroWeb analysis data sheet of CD of PBS buffers containing [**1**] at 192 μg/mL.

Batch	*Ordered α-helix*	*Random α-helix*	*Ordered β-sheet*	*Random β-sheet*	*Turns*	*Unordered*	***Total***
1	0.000	0.000	1.000	0.000	0.000	0.000	**1**
2	0.000	0.000	0.988	0.000	0.012	0.000	**1**

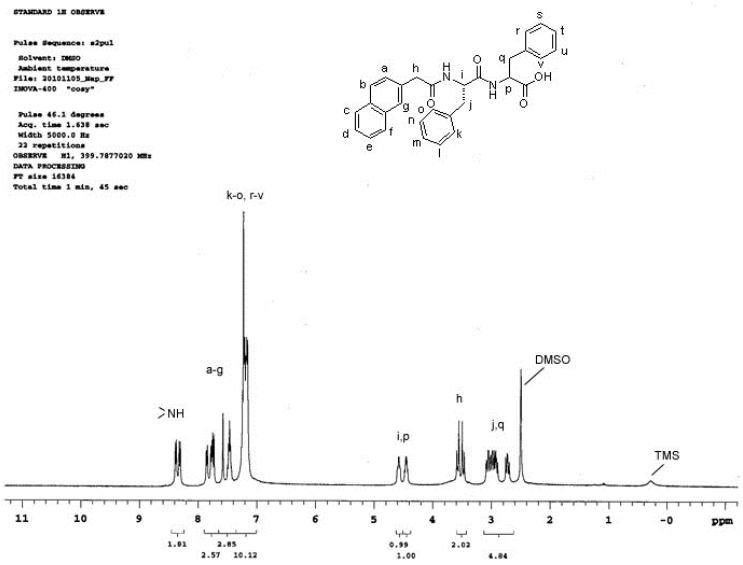

Fig. S1. ^{1}H-NMR spectrum of **1**.

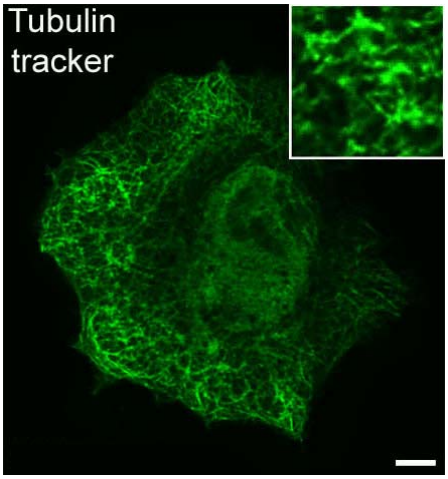

Fig. S2. Tubulin tracker stained HeLa cell treated by **2** for 24 h. Inset is 3X enlarged image. Scale bar = 10 μm.

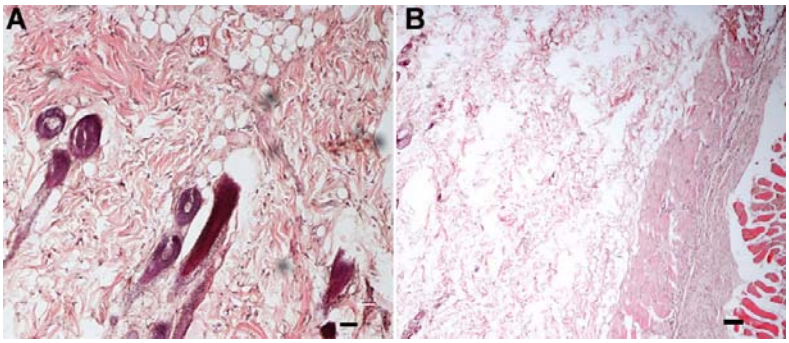

Fig. S3. Hematoxylin and eosin stain stained tissue of mice injected with **1**. (A) Staining of tissue at injection site on mice treated by subcutaneous injection of 1 mL PBS (1 injection; tissue sample taken after 10 days). (B) Staining of tissue at injection site on mice treated by subcutaneous injection of **1** (4.8 mg/mL in PBS; 1 mL; 1 injection; tissue sample taken after 10 days of injection). Scale bar = 50 μm.

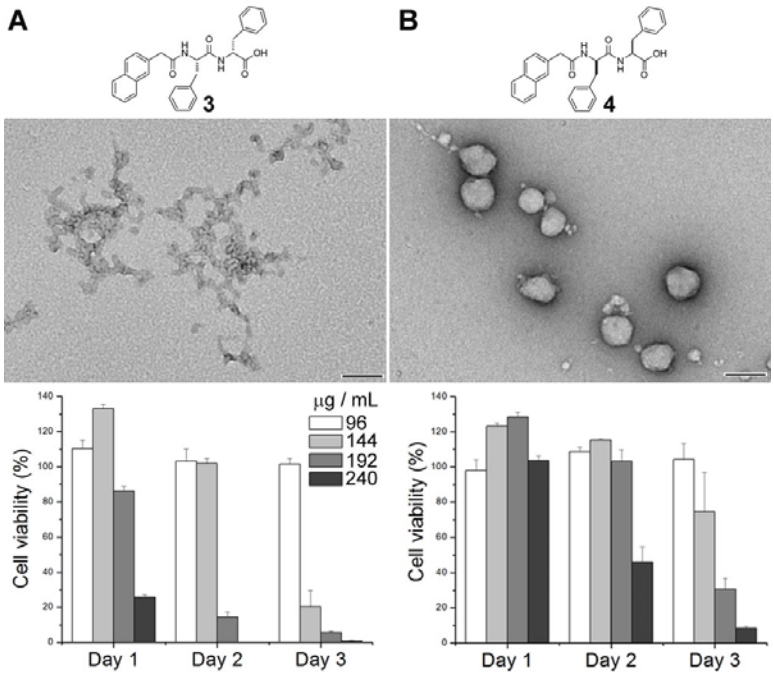

Fig S4. Chemical structures and cytotoxicities of stereochemical isomers of **1**. (A) Structure of **3**, in which the second Phe is replaced by D-Phe, the TEM image of **3** in PBS (240 μg/mL), and the cell viability of HeLa cells treated by **3**. (B) Structure of **4**, in which the first Phe is replaced by D-Phe, the TEM image of **4** in PBS (240 μg/mL), and the cell viability of HeLa cells treated by **4**. Scale bar = 100 nm.