Supplementary Material (ESI) for Chemical Communications

Supporting Information

Supramolecular Aptamer-Thrombin Linear or Branched Nanostructures

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Experimental Section:

Materials. Oligonucleotides (**1**), (**2**), and **(3)** were purchased from Genosys (Sigma, USA), and **(4)** was purchased from Integrate DNA Technologies, Inc. (USA). Chemicals were purchased from Sigma unless otherwise noted. Thrombin from human plasma was purchased from Sigma. Ultrapure water from a NANOpure Diamond (Barnstead, USA) source was used throughout all the experiments.

General Methods. All atomic force microscopy (AFM) imaging were performed at room temperature using a Multimode scanning probe microscope with a Nanoscope 3A controller (Digital Instruments / Veeco Probes / USA). Confocal fluorescence microscopy (CFM) were carried out on an Olympus FluoView FV300 Confocal Laser Scanning Microscpe with a UIS PLAPO 60× / 1.4 oil lens using a 488 nm Argon laser and a 543 nm He-Ne Laser for excitation. Alexa 488 fluorescence was observed at 505 − 530 nm. AFM topographical images were taken on samples deposited on freshly cleaved mica surfaces (Structure Probe, Inc., USA) that were first passivated with a 5 mM $MgCl_2$ solution for 1 min followed by the casting of a drop of the analyzed solution. Images recorded using Ultrasharp SiN AFM tips (Mikromasch, Germany) in tapping mode at

their resonant frequency, and the images were analyzed with WsXM SPIP software (Nanotec, Inc., Spain)[16].

Self-assembly of supramolecular aptamer-thrombin linear nanostructures. Linear α,β-bis-aptamer (**1**), 50×10^{-9} M, was reacted with thrombin, 50×10^{-9} M, in an aptamer binding buffer solution consisting of 20 mM Tris-HCl (pH 7.4), 140 mM NaCl, 5 mM KCl, 5 mM $CaCl_2$ and 1 mM $MgCl_2$ to form supramolecular aptamer-thrombin linear nanostructures. The reaction mixture was carried out at room temperature for 2 hrs. The supramolecular nanostructures were examined with no further purification.

Self-assembly of supramolecular aptamer-thrombin branched nanostructures. The tri-dentate aptamer (**2**) was mixed with the linear α,β-bis-aptamer, (**1**), and thrombin at a ratio of 1:5:5 that corresponded to 5 nM, 50 nM and 50 nM respectively. The branched supramolecular thrombin-aptamers nanostructure formed in an aptamer binding buffer solution consisting of 20 mM Tris-HCl (pH 7.4), 140 mM NaCl, 5 mM KCl, 5 mM $CaCl_2$ and 1 mM $MgCl_2$. The reaction mixture was carried out at room temperature for 4 hrs. The supramolecular nanostructures were examined with no further purification.

Self-assembly of supramolecular aptamer-thrombin linear fluorescent nanostructures. The linear α,β-bis-aptamer (**3)**, 50×10^{-9} M, was reacted with thrombin, 50×10^{-9} M and Alexa 488-labelled (**4)**, 100 nM, in an aptamer binding buffer solution consisting of 20 mM Tris-HCl (pH 7.4), 140 mM NaCl, 5 mM KCl, 5 mM $CaCl_2$ and 1 mM $MgCl_2$ to form supramolecular aptamer-thrombin linear nanostructures. The reaction mixture was carried out at room temperature for 2 hrs. The supramolecular nanostructures were examined with no further purification. The fluorescence emission was measured at $\lambda_{ex} = 488$ nm, $\lambda_{em} = 505 - 530$ nm.

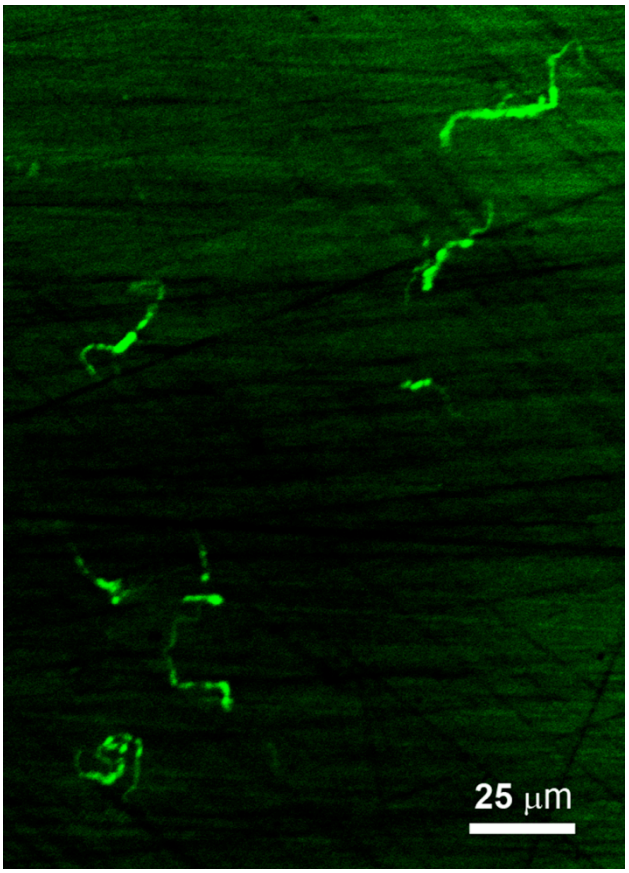

Figure S1. Confocal microscope image of the bundles bis-aptamer-bridged nanostructures.