Balancing flexibility and stress in DNA nanostructures
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Supplementary Information

Experimental Methods

Oligonucleotides. DNA sequences have been designed by a computer program “SEQUIN” (Seeman, N. C. J. Biomol. Struct. Dyn. 1990, 8, 573-581). All oligonucleotides were purchased from IDT, Inc. and purified by 20% denaturing PAGE.

Formation of DNA complexes/2D arrays. Strands 1 (0.6 μM), 2 (1.8 μM) and 3 or 3A (1.8 μM) were combined in Tris-Acetic-EDTA-Mg\(^{2+}\) (TAE-Mg\(^{2+}\)) buffer. Individual DNA complexes were formed by cooling mixture solutions as following: 95˚C /1 min., 65˚C/5 min., 50˚C/5 min., 37˚C/5 min., and 22˚C/5 min. DNA 2D arrays were formed by slowing cooling DNA solution from 95˚C from room temperature over 48 hours.

Denaturing Polyacrylamide Gel Electrophoresis. Gels contained 20% polyacrylamide (19:1 acrylamide/bisacrylamide) and 8.3 M urea; they were run at 55˚C. The running buffer was Tris-Borate-EDTA buffer (TBE), which consisted of 89 mM Tris buffer (pH 8.0), 89 mM boric acid, and 2 mM EDTA. Gels were run at Hoefer SE 600 electrophoresis unit at 600V (constant voltage). After electrophoresis, the gels were stained with Stains-all dye (Sigma) and scanned.

Native Polyacrylamide Gel Electrophoresis. Gels contained 6% polyacrylamide (19:1 acrylamide/bisacrylamide) and were run on a FB-VE10-1 electrophoresis unit (FisherBiotech) at 4˚C (100V, constant voltage). After electrophoresis, the gels were stained with Stains-all dye (Sigma) and scanned. TAE-Mg\(^{2+}\) buffer was used for electrophoresis at pH 8.0.

AFM Imaging. A drop of 2 μL DNA sample solution was spotted onto freshly cleaved mica surface, and sat there for 10 seconds to allow for strong adsorption. The sample drop was then washed off by 30 μL 10 mM Mg(Ac)\(_2\) solution, and dried by compressed air. DNA samples and their metal replicas were imaged by tapping-mode AFM on Nanoscope IIIa (Digital Instruments) with NSC15 tips (silicon cantilever, MikroMasch). The tip-surface interaction was minimized by optimizing the scan set-point.

DNA sequences. Strand 1-T4: 5′–Agg CAC CAT CgT Agg TTT TCT TgC CAg gCA CCA Ctc gTg TTC Tgg CCA gGC ACC ATC gTa ggT TTT TCT gCC –3′; Strand 1-T3: 5′–Agg CAC CAT CgT Agg TTT CTT gCC Agg CAC CAT CgT CAg gTT CTT CAg Agg CCA CCA TgC CAg gTa gTg TTT CTT gCC –3′; Strand 1-T2: 5′–Agg CAC CAT CgT Agg TTC TTg CCA gGC ACC ATC gTa ggT TCT gCg gCA CCA TgC TAg gTT CTT gCC –3′; Strand 1-T1: 5′–Agg CAC CAT CgT Agg TTT CTT gCC –3′; Strand 1-T10: 5′–Agg CAC CAT CgT Agg CTg gTg CCA gGC ACC ATC gTa ggT CTT gCC –3′; strand 2: 5′–ACT aTg CAA Cct gCC Tgg CCA gCC TAC gAT ggA CAC ggT AAC g –3′; strand 3: 5′–CgT aCt CgT gTg gTg CCA gGC ACC ATC gTa ggT CTT gCC –3′; strand 3A: 5′–CgC gCg TTA CCg TgT ggT TgC ATa gTC ATg–3′.
Blunt-ended 3-point-star:

Sticky-ended 3-point-star: