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 μ M), **2** (1.8 μ M) and **3** or **3A** (1.8 μ M) were combined in Tris-Acetic-EDTA-Mg²⁺ (TAE-Mg²⁺) 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-Mg²⁺ buffer was used for electrophoresis at pH 8.0.

AFM Imaging. A drop of 2 μ L 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 μ L 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 TCg TAg gTT TTC TTg CCA ggC ACC ATC gTA ggT TTT CTT gCC–3'; Strand **1-T3**: 5'–Agg CAC CAT CgT Agg TTT CTT gCC–3'; Strand **1-T2**: 5'–Agg CAC CAT CgT Agg TTT CTT gCC–3'; Strand **1-T2**: 5'–Agg CAC CAT CgT Agg TTC TTg CCA ggC ACC ATC gTA ggT TCT TgC CAg gCA CCA TCg TAg gTT CTT gCC–3'; Strand **1-T1**: 5'–Agg CAC CAT CgT Agg TCT TgC CAg gCA CCA TCg TAg gTC TTg CCA ggC ACC ATC gTA ggT CTT gCC–3'; Strand **1-T0**: 5'–Agg CAC CAT CgT Agg CAA gCC TAC gAT ggA CAC ggT AAC g–3'; strand **3**: 5'–CgT TAC CgT gTg gTT gCA TAg T–3'; strand **3**A: 5'–CgC gCg TTA CCg TgT ggT TgC ATA gTC ATg–3'.

