View metadata, citation and similar papers at core.ac.uk

brought to you by CORE

provided by Caltech Authors - Main

Supporting Information

© Wiley-VCH 2007

69451 Weinheim, Germany

Recognition of “Mirror-Image” DNA

by Small Molecules

Christian Dose[†], Dominik Ho[‡], Hermann E. Gaub[‡],

Peter B. Dervan[†*] & Christian H. Albrecht[‡*]

[†]Division of Chemistry and Chemical Engineering California Institute of Technology Pasadena, California 91125, USA, Fax: (+1) 626-683-8753, E-mail: dervan@caltech.edu

[‡]Lehrstuhl für Angewandte Physik and Center for Nanoscience Ludwig-Maximilians-Universität Amalienstrasse 54, 80799 Munich, Germany, Fax: (+49) 89-2180-2050, E-mail: christian.albrecht@physik.uni-muenchen.de

General Procedures and Materials:

Polyamide conjugates were synthesized on solid-phase using published Boc-based protocols and purified by reverse-phase high pressure liquid chromatography (HPLC).[3] Analytical HPLC was performed on a Beckman Gold system equipped with a diode array detector using a Phenomenex Gemini column (5 µm particle size, C18 110A, 250 x 4.6 mm, 5 micron). Preparative HPLC was performed on a Beckman Gold system equipped with a single-wavelength detector monitoring at 310 nm using a Phenomenex Gemini column (5 µm particle size, C18 110A, 250 x 21.2 mm, 5 micron). For both analytical and preparative HPLC, solvent A was 0.1% (v/v) aqueous trifluoroacetic acid (TFA) and solvent B was acetonitrile. Solvent gradients were adjusted as needed. UV-Vis spectra were recorded in water on a Hewlett-Packard Model 8452 A diode array spectrophotometer. All polyamide concentrations were determined using an extinction coefficient of 69.200 $M^{-1}\bullet cm^{-1}$ at λ_{max} near 310 nm. Matrix-assisted, LASER desorption/ionization time-of-flight mass spectrometry (MALDI-TOF MS) was performed using an Applied Biosystems Voyager DR Pro spectrometer. Polyamide **5**: MALDI-TOF $[M+H]^+$ calcd for $C_{57}H_{71}N_{22}O_{10}^+$ = 1223.6, observed = 1223.4; polyamide **(*R*)-6**: MALDI-TOF $[M+H]^+$ calcd for $C_{57}H_{72}N_{23}O_{10}^+$ = 1238.6, observed = 1238.6; polyamide **(*S*)-6**: MALDI-TOF $[M+H]^+$ calcd for $C_{57}H_{72}N_{23}O_{10}^+$ = 1238.6, observed = 1238.5; polyamide **(*R*)-7**: MALDI-TOF $[M+H]^+$ calcd for $C_{58}H_{73}N_{22}O_{10}^+$ = 1237.6, observed = 1237.3; polyamide **(*S*)-7**: MALDI-TOF $[M+H]^+$ calcd for $C_{58}H_{73}N_{22}O_{10}^+$ = 1237.6, observed = 1237.5.

D-DNA oligomers **1** and **2** were purchased HPLC purified from Integrated DNA Technologies (Coralville, USA). L-DNA oligomers **3**, **4**, and DNA-conjugates used for the molecular force balance 5′-H_2N-TTTTTTTTTTCAGTCGCTGACCAACCTCGT-3′, 3′-GTCAGCGACTGGTTGGAGCACTTTTT-T-5′-5′-*T*(Cy3)*TTTTTCACGAGGTTGGTCAGCGACTG*-3′, 3′-*TGCTCCAACCAGTCGCTGACTTTTTT-TTTT*-5′-biotin (italic letters represent L-DNA monomers) were purchased HPLC purified from IBA GmbH (Goettingen, Germany). The DNA-oligomers for the molecular force balance containing the polyamide-binding motifs (gray) were aligned as shown in Figure S1.

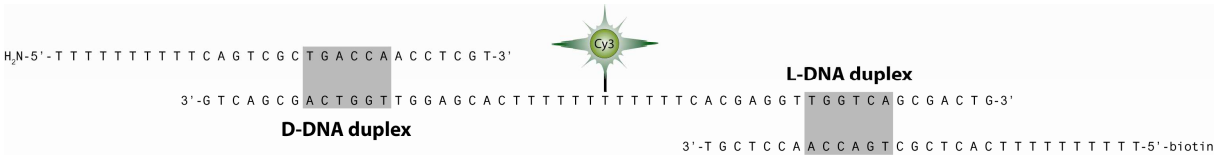

Figure S1. DNA sequences and polyamide binding motifs of the molecular force balance.

The molecular force balance setup was performed according to literature [12]. A polymethylmethacrylat (PMMA) fluid cell placed on top of the glass slide was used for polyamide addition. Polydimethylsiloxane (PDMS) stamps were PEG-biotin functionalized using epoxy-trimethoxysilane (ABCR, Karlsruhe, Germany) and amino-PEG-biotin (mw: 3400 g/mol, Rapp Polymere, Goettingen, Germany). The PDMS stamps were incubated in 1x PBS (phosphate buffer saline) containing 15 nM streptavidin (Invitrogen, Karlsruhe, Germany) and 0.4% (w/w) bovine serum albumin (BSA) for at least one hour before use. Polyamides in 1x PBS buffer were added to the fluid cell 30 min prior to the experiment. Streptavidin coated PDMS stamps were approached to the force balances using high precision stepper motors (OWIS, Staufen, Germany) and a piezo actuator, monitored by reflection interference contrast microscopy (RICM). The biotinylated force balances and the streptavidin coated PDMS stamp were allowed to couple for 10 min, followed by retraction of the PDMS stamp at a velocity of 5 µm/s. Images of the molecular force balance glass slide were recorded by a confocal fluorescence scanner at 4 µm resolution (Tecan, Austria). Melting temperature analysis was monitored on a Beckman UV-Vis spectrometer at 260 nm within 25-90 °C by applying a heating rate of 0.5 °C/min. Measurements were performed in a degassed buffer containing 2 µM DNA duplex/polyamide (1:1), 10 mM NaCl, and 100 mM NaH_2PO_4 at pH 7.0. T_m-values were defined as the maximum of the first derivative of the melting curve.

Polyamide ball-and-stick representation legend: Black and white circles represent imidazole and pyrrol rings, respectively, half-circles represent γ-aminobutyric acid, half-circles containing a cross represent positive charged amines, and white diamonds represent β-alanine moieties. All polyamides contain 3-(dimethylamino)-1-propylamine (Dp) as tail.