Supplementary Data

6-Aminoacridizinium Bromide: A fluorescence probe which lights up in AT-rich regions of DNA‡

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Materials. The 6.aminoacridizinium bromide (**1a**) was synthesized according to literature.[1] Calf thymus DNA (ct DNA) and salmon testes DNA (st DNA) sodium salt, were purchased from Sigma; (poly[dG-dC]-poly[dG-dC]) and (poly[dA-dT]-poly[dA-dT]) were purchased from Pharmacia Amersham and used as received. The concentrations (in nucleotides) were determined by UV spectroscopy (ct DNA, st DNA and (poly[dA-dT]-poly[dA-dT]): ε_{260} = 6600 $M^{-1}cm^{-1}$, (poly[dG-dC]-poly[dG-dC]): ε_{254} = 8400 $M^{-1}cm^{-1}$.

DNA binding studies. Nucleic-acid concentrations in bases, were determined spectrophotometrically using the reported data for the molar absorption coefficient at the indicated wavelength. UV/vis absorption spectra were recorded on a Hitachi U3200, and emission spectra were recorded on a Perkin-Elmer LS-50B luminescence spectrophotometer. Experiments were carried out in phosphate buffer (10 mM, = 7.0) or ETN buffer (10 mM TRIS, 1 mM EDTA, 10 mM NaCl, pH = 7.0). Binding affinities of **1a** to DNA were determined by spectrophotometric titrations. From these data, the concentration of bound and free ligand (C_b and m) was determined from the absorbance at a fixed wavelength (λ = 374 nm) according to the equations 1 and 2.

$$C_b = (A_f\text{-}A/A_f\text{-}A_b)C_0 \qquad \text{(eq. 1)}$$

$$m = C_0 - C_b \qquad \text{(eq. 2)}$$

A_f is the absorbance of the free ligand, A_b is the absorbance of the bound ligand, A is the absorbance of a mixture of the free and bound compounds, and C_0 is the total concentration of the compound. From these data, the values r (molecules of dye bound per nucleotide) and

c (dye free in solution) were calculated. To avoid large systematic inaccuracies due to experimental errors in the extintion coefficients, only data from the saturation fraction of 0.85-0.15 were used. Binding isotherms were represented as Scatchard plots, i.e. r/c *vs* r, and evaluated according to the model of McGhee and von Hippel[2] to obtain the binding constant (K) and the binding site size (n). Note: To maintain a constant concentration of the dye during the titration, the titrated DNA solution also contained acridizinium salt **1a** at the same concentration as in the couvette. Photometric titration experiments were performed in 10 mM phosphate buffer (pH 7.0).

The DNA addition to the acridizinium solutions was monitored by emission spectroscopy with an excitation wavelength at the isosbestic point, which was determined by the spectrophotometric titrations. Fluorimetric titration experiments were performed in 10 mM phosphate buffer (pH 7.0) at [**1a**] = 10^{-5} M.

Flow linear dichroism. Linear-dichroism (LD) spectra were recorded in a "flow cell" on a Jasco J500A spectropolarimeter equipped with an IBM PC and a Jasco J interface. The determination and interpretation of the data was performed as previously described.[3] DNA concentration was 2.27 mM and the measurements were performed at [DNA]/[**1a**] = 0, 0.04, 0.08, and 0.20.

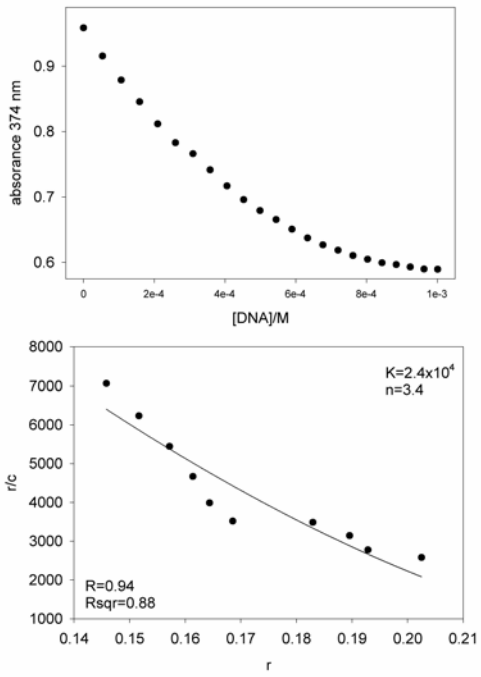

Figure S1. Plots ΔA vs [DNA] and the corresponding Scatchard plot derived from spectrophotometric titration of ct DNA to **1a**; c(**1a**) = 10^{-4} M.

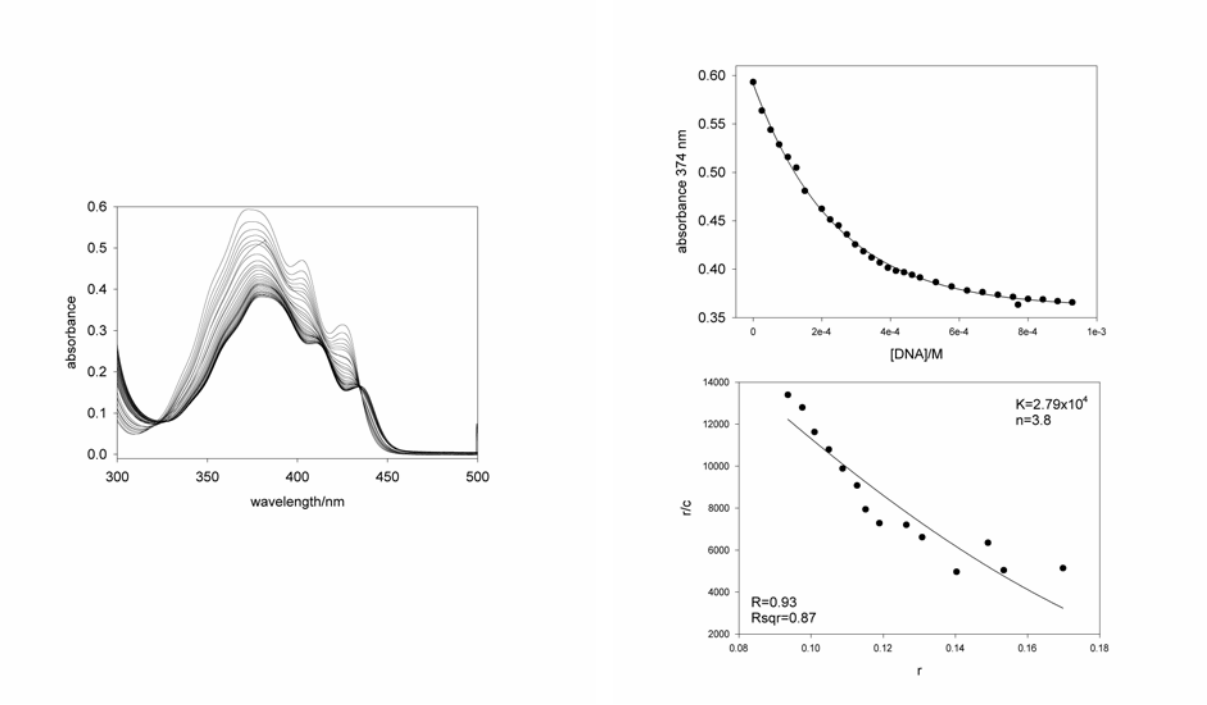

Figure S2. Plots ΔA vs [DNA] and the corresponding Scatchard plot derived from spectrophotometric titration of poly[dA-dT]-poly[dA-dT] to **1a**; c(**1a**) = 5×10^{-5} M.

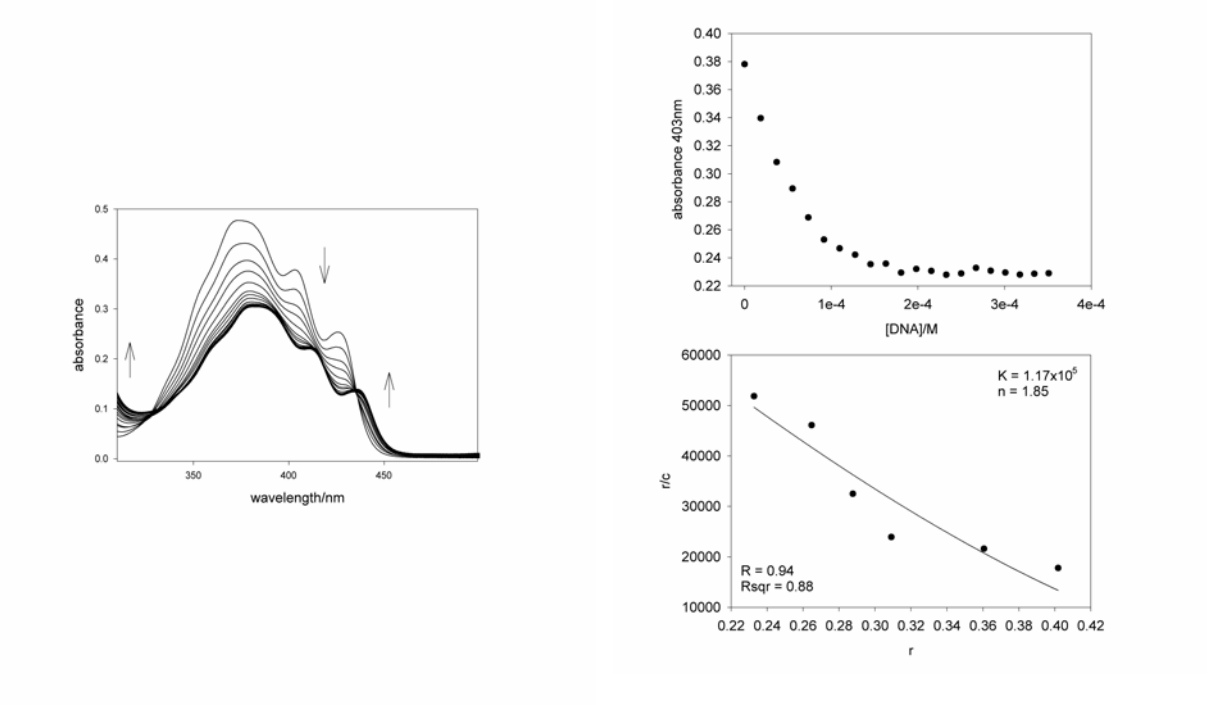

Figure S3. Plots ΔA vs [DNA] and the corresponding Scatchard plot derived from spectrophotometric titration of poly[dC-dG]-poly[dC-dG] to **1a**; c(**1a**) = 3×10^{-5} M

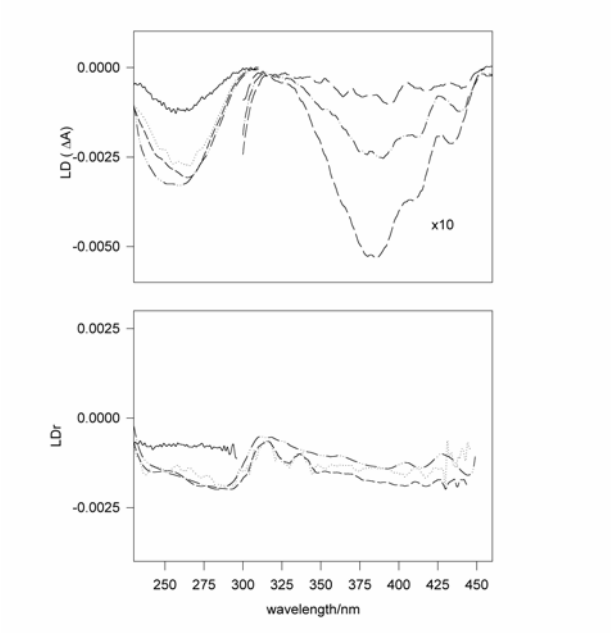

Figure S4. LD(**A**), and LD_r (**B**) spectra of mixtures of st DNA and 6-aminoacridizinium bromide (**1a**) at different [**1a**]/[DNA] ratios 0, 0.04, 0.08, and 0.20 (in order of increasing signal intensity).

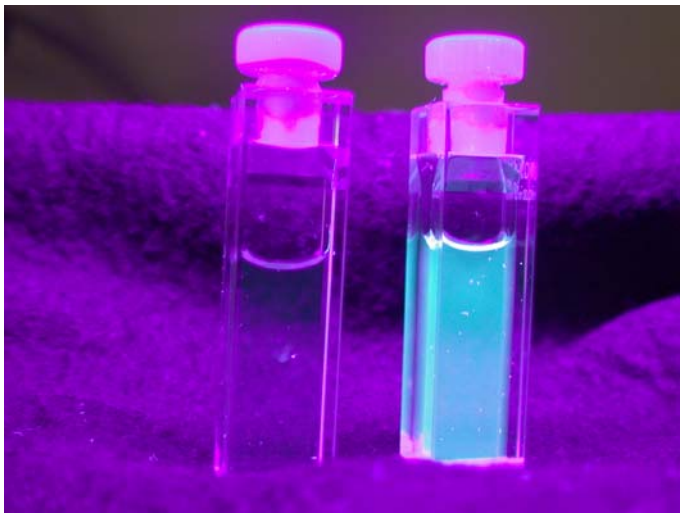

Figure S5. Samples of **1a** (c = 10^{-5} M) in the presence of ct DNA (left, c =) and poly[dA-dT]-poly[dA-dT]) (right; c =), λ_{ex} = 350 nm.

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[1] C. K. Bradsher and J. P. Sherer, *J. Org. Chem.*, 1967, **32**, 733.

[2] J. D. McGhee and P. H. von Hippel, *J. Mol. Biol.*, 1974, **86**, 469.

[3] H. Ihmels, K. Faulhaber, C. Sturm, G. Bringmann, K. Messer, N. Gabellini, D. Vedaldi and G. Viola, *Photochem. Photobiol.*, 2001, **74**, 505.