Supplementary Material

Cationic side-chains control DNA/RNA binding properties and antiproliferative activity of dicationic dibenzotetraaza[14]annulene derivatives.

Marijana Radić Stojković,[a] Marko Marjanović,[c] Dariusz Pawlica,[b] Lukasz Dudek,[b] Julita Eilmes,[b] Marijeta Kralj,[c] Ivo Piantanida,[a*]

[a] *Division of Organic Chemistry and Biochemistry, Ruđer Bošković Institute, Bijenička cesta 54, PO Box 180, HR-10002 Zagreb, Croatia*

[b] *Department of Chemistry, Jagiellonian University, Ingardena 3 ,30-060 Kraków, Poland*

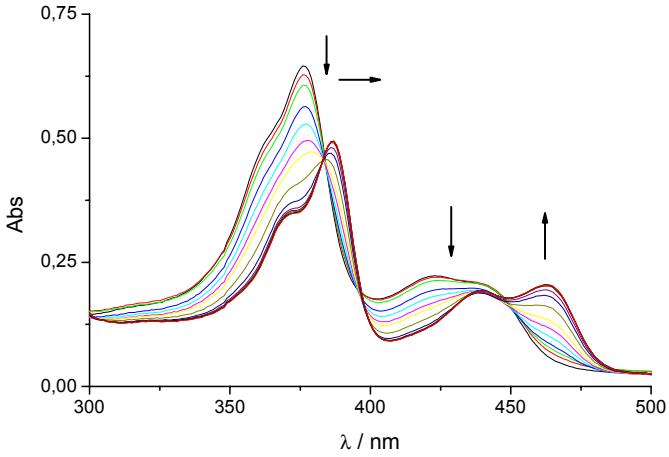
[c] *Division of Molecular Medicine, Ruđer Bošković Institute, Bijenička cesta 54, P. O. Box 180, HR-10002 Zagreb, Croatia*

Solubility

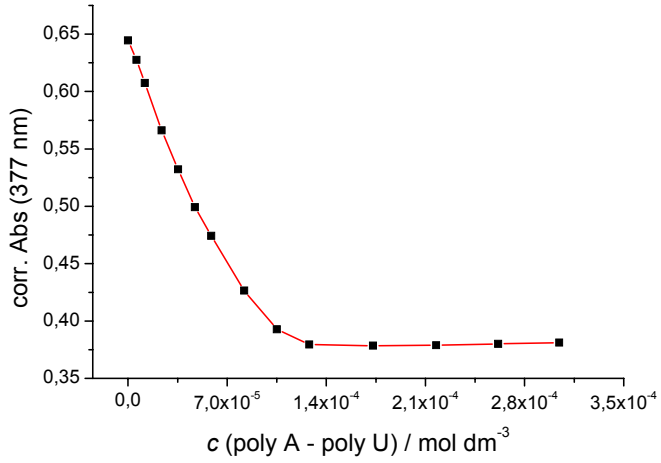
Studied compounds were dissolved in sodium cacodylate buffer, I = 0.05 mol dm^{-3} at pH = 7.0 (**4**, c=1.54 × 10^{-3} mol dm^{-3}; **5**, c=1.44 × 10^{-3} mol dm^{-3}; **6**, c=1.55 × 10^{-3} mol dm^{-3}; **7**, c=1.67 × 10^{-3} mol dm^{-3}).

The absorbencies of buffered aqueous solutions of studied compounds are proportional to their concentrations up to c=3-4 × 10^{-5} mol dm^{-3}. Buffered aqueous solutions of studied compounds were stable for at least one week, when stored in a dark and cold place (+ 8^{0}C).

Spectrophotometric titrations

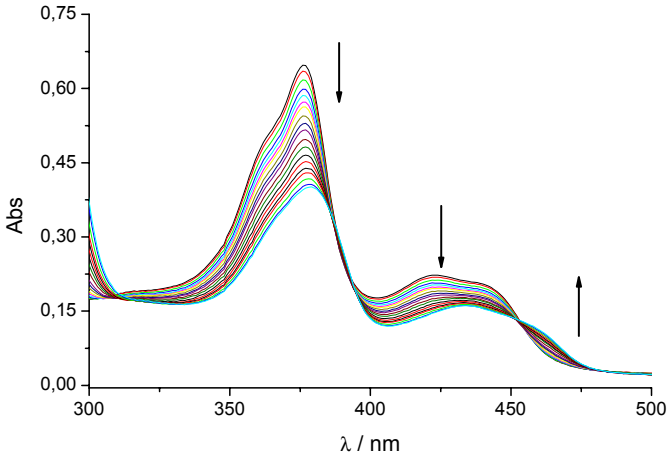


a)

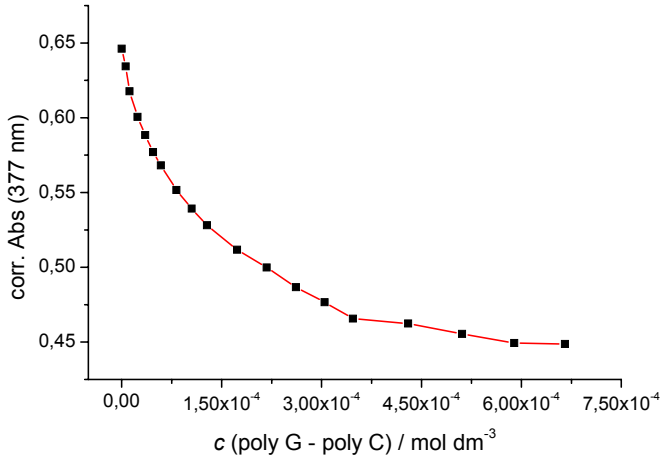


b)

Figure S1. a) Changes in UV/Vis spectrum of **4** (c= 1.53 × 10^{-5} mol dm^{-3}) upon titration with poly A – poly U; b) Dependence of **4** absorbance at λ_{max} = 377 nm on c(poly A – poly U), at pH=7, sodium cacodylate buffer, I = 0.05 mol dm^{-3}.



a)



b)

Figure S2. a) Changes in UV/Vis spectrum of **4** (c= 1.53 × 10^{-5} mol dm^{-3}) upon titration with poly G – poly C; b) Dependence of **4** absorbance at λ_{max} = 377 nm on c(poly G – poly C), at pH=7, sodium cacodylate buffer, I = 0.05 mol dm^{-3}.

Viscometry measurements

Viscometry measurements were conducted with an Ubbelohde viscometer system AVS 350 (Schott). The temperature was maintained at 25 +/- 0.1 °C. Aliquots of drug stock solutions were added to 5.5 ml of 5 × 10^{-4} mol dm^{-3} ct-DNA solution in sodium cacodylate buffer, I = 0.05 mol dm^{-3}, pH=7, with a compound to DNA phosphate ratio r less than 0.2. Dilution never exceeded 4% and was corrected for in the calculations. The flow times were measured at least five times optically with a deviation of +/- 0.2 s. The viscosity index α was obtained from the flow times at varying r according to the following equation:[1]

$$L/L_0 = [\,(t_r - t_0)\,/\,(t_{DNA} - t_0)\,]^{1/3} = 1 + \alpha * r$$

Where t_0, t_{DNA} and t_r denote the flow times of buffer, free DNA and DNA complex at ratio $r_{[compound]\,/\,[ct\text{-}DNA]}$, respectively; L/L_0 is the relative DNA lengthening. The L/L_0 to $r_{[compound]\,/\,[ct\text{-}DNA]}$-plot was fitted to a straight line that gave slope α. The error in α is ≤ 0.1.

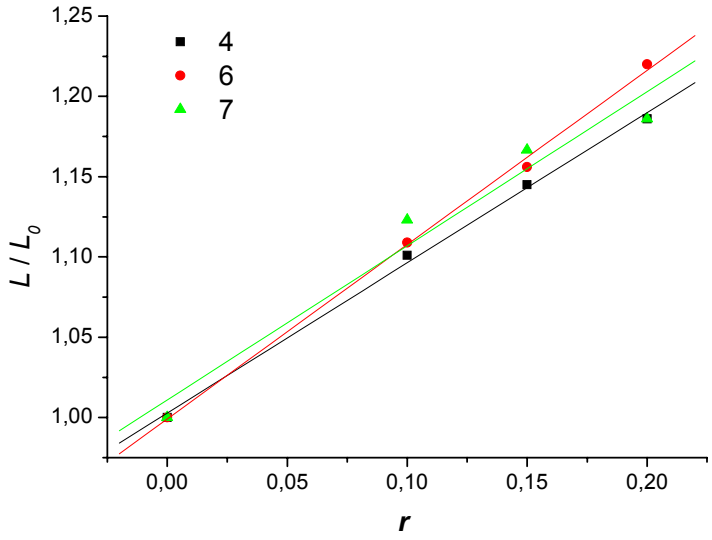


Figure S3. Relative DNA helix length extension (L/L_0) *vs*. ratio $r_{[compound]\,/\,[ct\text{-}DNA]}$ plot for **4**, **6** and **7** at pH=7, sodium cacodylate buffer, I = 0.05 mol dm^{-3}. Results: $\alpha = 0.93 \pm 0.03$ (for **4**); 1.08 ± 0.04 (for **6**); 0.96 ± 0.1 (for **7**). It was not possible to determine α for compound **5** because of a precipitation.

---

[1] G. Cohen and H. Eisenberg, *Biopolymers* 1969, **8,** 45; M. Wirth, O. Buchardt, T. Koch, P. E. Nielsen nad B. Nordén, *J. Am. Chem. Soc*. 1988, **110,** 932.