Stabilization of DNA Triple Helices by Crescent-Shaped Dibenzophenanthrolines

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Abstract: Mono- and disubstituted crescent-shaped dibenzophenanthrolines have been prepared and their ability to stabilize synthetic DNA triple helices has been investigated. Thermal denaturation experiments were conducted on two triplexes of different sequences in the presence of the various compounds. For both targets, the monosubstituted derivatives (**4a,b**) induced a large and specific increase of the temperature of the triplex-to-duplex transition compared with the duplex-to-single strand transition ($\Delta T_m^{3\rightarrow2} - \Delta T_m^{2\rightarrow1}$ up to $+32\,^{\circ}C$), and appeared to be more efficient than their disubstituted analogues. Therefore compounds **4a** and **4b** seem to be better triplex-specific ligands than the previously reported benzopyridoindole (BPI) derivatives.

Keywords: dibenzo[*b*,*j*]phenanthrolines · DNA triplex stabilization · DNA structures · N ligands · oligonucleotides

Introduction

Oligonucleotide-directed triple-helix formation has raised considerable interest as a result of its potential biological and therapeutic applications[1, 2] (for reviews see refs. [3, 4]). However, in many cases triplexes are thermodynamically less stable than corresponding duplexes. Therefore, the use of triplex-specific ligands should be of benefit to stabilize triplexes. In the past few years several ligands have been reported to bind to triplexes, with some ligands stabilizing and others destabilizing the triple-helical complexes.[5–15] Among them, benzo[*e*]pyridoindole (B*e*PI) and benzo[*g*]pyridoindole (B*g*PI) derivatives have been shown to significantly stabilize pyrimidine-motif triplexes by intercalation between $T \cdot A \times T$ base triplets.[7, 16–18] Here we report a new series of pentacyclic crescent-shaped compounds, dibenzophenanthroline derivatives, which exhibit better triplex-specific stabilization properties than the previously described B*e*PI and B*g*PI compounds.

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Results and Discussion

On the basis of simple geometric considerations and subsequent molecular modeling, the dibenzo[*b*,*j*]phenanthroline derivatives (i.e. quinacridines) would appear to have the appropriate size and shape to provide broad overlap with nucleobase triplets. Such pentacyclic angular compounds seem, therefore, to be potential candidates for stabilizing ordered nucleic acid structures such as triplexes through strong $\pi - \pi$ electronic orbital overlap.

Synthesis: Two series of water-soluble aminodibenzophenanthrolines have been synthesized: a monosubstituted series bearing one aminoalkyl side chain on the central ring and a disubstituted series bearing two side chains on the external benzo rings.

The disubstituted compounds **1–3** (Figure 1) were prepared by a general procedure that we reported recently.[19] These isomeric compounds exhibit various relative locations of the two ring nitrogens and of the aminoalkyl side chains that might interact differently with a triplex. The monosubstituted compounds **4a** and **4b** were obtained by a synthetic pathway derived from that used for compounds **1–3** (Scheme 1). Ullmann–Goldberg dicondensation between 2-aminobenzoic acid and 2,5-dibromotoluene followed by cyclization by $POCl_3$ (Scheme 1, steps a and b) afforded dichloroquinacridine **6** exclusively, with no trace of the linear isomer in spite of the considerable steric hindrance provided by the proximity of the two chlorine atoms. The selectivity of the ring closure, as has already been discussed,[19] is driven by electronic factors rather than steric ones. Furthermore, X-ray

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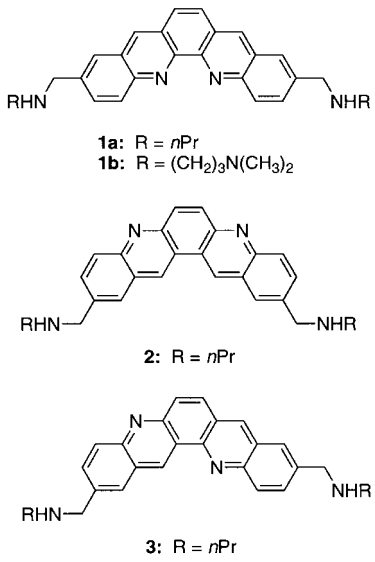
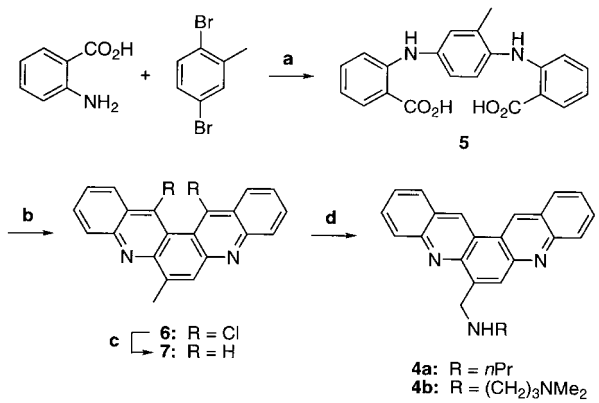


Figure 1. Chemical structure of the disubstituted dibenzo[*b*,*j*]phenanthroline derivatives (compounds **1–3**).



Scheme 1. Synthesis of the monosubstitued dibenzo[*b*,*j*][4,7]phenanthrolines **4a** and **4b**. Reagents and reaction conditions: a) Cu (0.25 equiv), CuI (0.2 equiv), K_2CO_3 (2.5 equiv), *n*-pentanol, reflux, 4 h, 65%; b) $POCl_3$, reflux, 3.5 h, 28%; c) $LiAlH_4$ (8.0 equiv), THF, reflux, 10 h, then $FeCl_3$ (4.0 equiv), ethanol/H_2O, reflux, 87%; d) i) *N*-bromosuccinimide (1.0 equiv), perbenzoic anhydride (0.06 equiv), CCl_4, reflux, 10 h, ii) RNH_2, 50 °C, 4 h, 50% (**4a**) and 33% (**4b**).

Abstract in French: *Des dibenzophénanthrolines de forme coudée, mono- et disubstituées, ont été préparées et leur capacité à stabiliser des triples hélices d'ADN synthétiques a été évaluée. Des expériences de dénaturation thermique ont été menées sur deux triplexes de séquence différente en présence de ces composés. Pour les deux cibles, les dérivés monosubstitués (**4a,b**) induisent une augmentation forte et spécifique de la température de la transition triplex–duplex par rapport à la transition duplex–simple brin ($\Delta T_m^{3\rightarrow2} - \Delta T_m^{2\rightarrow1}$ jusqu'à 32 °C), et se révélent plus efficaces que leurs analogues disubstitués. En conséquence, il semble que les composés **4a,b** soient de meilleurs ligands spécifiques des triplexes que les dérivés benzopyridoindoles (BPI) précédemment décrits.*

analysis of the disubstituted analogue of **6** (dichlorodimethylquinacridine) has shown that the aromatic system is strongly twisted to accommodate the two chlorine atoms (torsion angle between the central ring and the two adjacent heterocycles is 35.9°).[20]

The monomethyl intermediate **7** obtained by reduction of **6** was then submitted to successive bromination and nucleophilic substitution by the appropriate amine to give the amino derivatives **4a** and **4b**. This synthetic pathway (Scheme 1, step d) provided higher yields than the reaction sequence used to prepare the disubstituted analogues **1–3** (i.e. oxidation of the dimethyl precursors/Schiff base formation/reduction).[19]

Compounds **1–4** were protonated by 1N aqueous HCl to give the water-soluble salts suitable for biochemical assays. The pH dependence of the UV-visible and fluorescence spectra of **1–4** showed that the pK_a's of the two ring nitrogens lie below pH 5 and those of the side chains nitrogens above pH 7 (data not shown).[19] This indicates that all the compounds tested are exclusively protonated on the side chains at physiological pH.

Thermal denaturation experiments: Stabilization of the triple helices (Table 1, top) as well as that of the target double helices was studied by means of thermal denaturation experiments by UV absorption spectroscopy in the presence and absence of ligands as previously described.[18] In these triple helices, a 14-mer oligonucleotide binds to a 26-base-pair (bp)

Table 1. Top: A 14-mer triple helix-forming oligopyrimidine can bind to a 26-bp DNA duplex containing a 14-mer oligopyrimidine·oligopurine target sequence. Two sequences are used, **14C3** and **14C5**, which differ by the number of cytosines in the third strand (3 cytosines for **14C3** and 5 for **14C5**). Bottom: ΔT_m values (±1 °C) of triplex and duplex in the presence of compounds **1–4,** as well as B*e*PI and B*g*PI. In the absence of ligands, the $T_m^{3\rightarrow2}$ values of triplexes **14C3** and **14C5** are 18 °C and 26 °C, respectively, whereas the $T_m^{2\rightarrow1}$ values for the corresponding target duplex are 58 °C and 60 °C, respectively. The thermal denaturation experiments were carried out in 10 mM cacodylate buffer (pH 6.2) containing 0.1 M NaCl in the absence or in the presence of 15 μM ligand, for both triple and double helix at a concentration of 1.5 μM.

14C3

5' **TTCTTCTTTTTTCT** 3'
5' CGAGTT**AAGAAGAAAAAAGA**TTGAGC 3'
3' GCTCAA**TTCTTCTTTTTTCT**AACTCG 5'

14C5

5' **TTCTTCTCCTTTCT** 3'
5' CGAGTT**AAGAAGAGGAAAGA**TTGAGC 3'
3' GCTCAA**TTCTTCTCCTTTCT**AACTCG 5'

	14C3		14C5	
	$\Delta T_m^{3\rightarrow2}$ [°C]	$\Delta T_m^{2\rightarrow1}$ [°C]	$\Delta T_m^{3\rightarrow2}$ [°C]	$\Delta T_m^{2\rightarrow1}$ [°C]
1a	+7	+2	+2	+2
1b	+16	+10	+5	+10
2	+18	+6	+12	+6
3	+20	+10	+11	+10
4a	+37	+5	+24	+5
4b	+44	+12	+19	+12
BePI	+20	+12	+3	+10
BgPI	+28	+16	+12	+13

DNA fragment containing a 14-bp oligopyrimidine·oligopurine target sequence. One of the triplexes contains three positively charged $C \cdot G \times C^+$ triplets (triplex **14C3**), and the other has five (triplex **14C5**). Results are summarized in Table 1 where $\Delta T_m^{3\rightarrow2}$ and $\Delta T_m^{2\rightarrow1}$ are the difference in melting temperatures (T_m) for the triplex-to-duplex and the duplex-to-single strand transitions, respectively, in the presence and absence of ligands. It should be noted that the melting curves of triplex **14C3** in the presence of compounds **4a** and **4b** exhibit a single transition at high temperature (data not shown). Therefore a 36-bp DNA fragment, which differs from the 26-bp duplex by five additional base pairs at both ends, was used to assess the triplex stabilization in the presence of **4a** and **4b**, since a better separation between the triplex-to-duplex and the duplex-to-single strand transitions could be obtained. Figure 2 shows the triplex stabilization by these

14C3

5′ **TTCTTCTTTTTTCT**

5′ GATACGAGTT**AAGAAGAAAAAAGA**TTGAGCTGATAG

3′ CTATGCTCAA**TTCTTCTTTTTTCT**AACTCGACTATC

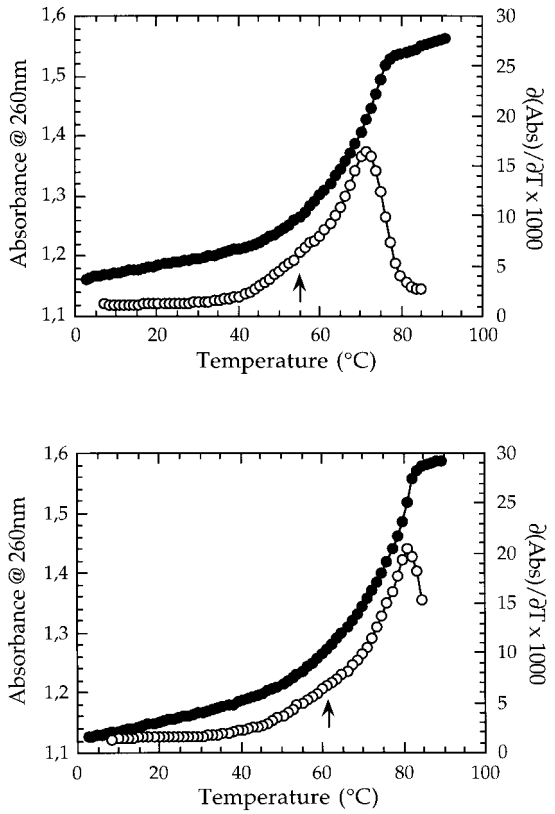


Figure 2. Melting curves (filled circles) and their first derivatives (open circles) of triplex **14C3** with a 36-bp target duplex in the presence of **4a** (top) and **4b** (bottom). The arrows point to the center of the shoulder as an approximate estimate of $T_m^{3\rightarrow2}$ value.

compounds with the 36-bp duplex. The melting curves reveal two overlapped transitions, which were confirmed by the presence of a shoulder near the main peak on the first derivatives of the melting curves. The control experiment with the 36-duplex in presence of **4a** and **4b** indicates that the main peak is related to the duplex-to-single strand transition (data not shown). Therefore the observed shoulder is ascribed to the triplex-to-duplex transition, and the $T_m^{3\rightarrow2}$ value is estimated at the center of the shoulder.

The disubstituted compounds **1a**, **2**, and **3**, which differ from each other by the relative location of two ring nitrogen atoms (Figure 1), exhibit different triplex/duplex stabilization properties. The $\Delta T_m^{3\rightarrow2}$ values of compounds **2** and **3** are higher than those of **1a** by about 10 °C in both triplexes, whereas the $\Delta T_m^{2\rightarrow1}$ values of **2** and **3** are higher than those of **1a** by 4 °C and 8 °C, respectively, in both duplexes. To determine the role played by the aminoalkyl side chains, compound **4a** was compared with **2**. In **4a**, one aminopropyl side chain is attached to the central ring, whereas two aminopropyl groups are attached to the external benzene rings of **2**. Compound **4a** exhibits significantly higher triplex stabilization properties for both triplexes than compound **2**, but only moderate duplex stabilization. It should be noted that the presence of additional charges in compounds **1b** and **4b** does increase the $\Delta T_m^{3\rightarrow2}$ values for the triplex **14C3**, but only marginally stabilizes (**1b**) or even destabilizes (**4b**) the triplex **14C5**. This might be explained by electrostatic repulsion between the ligand and $C \cdot G \times C^+$ base triplets; compounds **1b** and **4b** bear two positive charges per side chain and, therefore, interact less favorably with the triplex **14C5** than with the **14C3**.

The comparison of the triplex/duplex stabilization of the newly synthesized compounds with the previously reported benzo[*e*]- and benzo[*g*]-pyridoindoles (B*e*PI and B*g*PI) reveals that compound **4b** is the most triplex-specific ligand for triplex **14C3**, whereas compound **4a** is best for triplex **14C5**. Again, this difference can be explained by a greater extent of electrostatic repulsion between the five $C \cdot G \times C^+$ triplets and the biprotonated aminoalkyl side chain of compound **4b** than that between the five triplets and the monoprotonated side chain of compound **4a**. Recently we described the properties of pentacyclic benzoquinoloquinoxaline (BQQ) and benzoindoloquinoline (BIQ) that also produce a high stabilization of triplex structures.[21, 22] In particular BQQ exhibited a high selectivity for the triplex compared with the corresponding duplex.[21]

Linear dichroism experiments have provided evidence that benzopyridoindole derivatives intercalate between the base triplets of the triple helix.[23] ΔT_m measurements reported in the present work do not address the question of the structure of the complexes formed by dibenzophenanthroline derivatives. Further investigations are required to characterize their binding mode and affinity.

Conclusion

The newly synthesized dibenzophenanthroline derivatives (compounds **4a** and **4b**) are the most efficient triplex-stabilizing ligands for both triplex sequences studied in the present work. Therefore, compounds **4** could represent promising new lead molecules for further development of triplex-stabilizing ligands.

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Experimental Section

Thermal denaturation experiments: All thermal denaturation studies were carried out on a Uvikon 940 spectrophotometer, interfaced to an IBM-AT personal computer for data collection and analysis. Temperature control of the cell holder was achieved by a Haake D8 circulating water bath, the temperature of which was decreased from 90 to 0 °C and then increased back to 90 °C at a rate of 0.1 °C min^{-1} with a Haake PG 20 thermoprogrammer. The absorbance at 260 nm was recorded every 10 minutes. The maxima of the first derivatives of the melting curves gave a good approximation of the half-dissociation temperature (T_m) and allowed us to characterize the stability of the complexes in an easy and reproducible way. The DNA melting experiments were carried out in a 10 mM cacodylate buffer (pH 6.2) containing 0.1 M NaCl in the absence and in the presence of 15 μM ligands for both triple and double helices at the concentration of 1.5 μM.

Synthesis: All commercially available chemicals were reagent grade and used without further purification. ^{1}H and ^{13}C NMR spectra were recorded on a Bruker AC 200 spectrometer. UV/Vis experiments were monitored on a Beckmann DU 640 spectrophotometer. Fluorescence measurements were performed on a Spex Fluoromax spectrophotometer equipped with a Hamamatsu R928 photomultiplier (PM) and a thermostated cell holder. Melting points were determined on an Electrothermal 9100 apparatus. IR spectra were obtained on a Bruker Vector 22 FT-IR spectrometer. The microanalyses were performed at the Service Régional de Microanalyse de l'Université Pierre et Marie Curie (Paris).

2,2′-(2-Methyl-1,4-phenylenediamino)bisbenzoic acid (**5**): A mixture of 2,5-dibromotoluene (5.0 g, 20.0 mmol), 2-aminobenzoic acid (13.72 g, 100.0 mmol), copper bronze (318 mg, 5.0 mmol), CuI (762 mg, 4.0 mmol), potassium carbonate (6.91 g, 50.0 mmol), and *n*-pentanol (75 mL) was stirred under reflux for 4 h. After removing the solvent in vacuo, methanol (250 mL) was added and the solution was filtered through Celite. The filtrate was concentrated under vacuum and concd HCl was added. The precipitate was washed with hot methanol and recrystallized from THF/DMSO/water to yield 4.7 g (65 %) of a pale yellow powder. M.p. 230–240 °C; ^{1}H NMR (200 MHz, [D$_6$]DMSO): $\delta = 2.17$ (s, 3H), 6.65–6.80 (m, 3H), 7.10–7.42 (m, 6H), 7.88 (2d, $J = 7.0$ Hz, 2H), 9.40 (s, 1H), 9.60 (s, 1H), 12.97 (br s, 2H); IR (nujol): $\tilde{\nu} = 3326, 3200–3000, 1657$ cm^{-1}; $C_{21}H_{18}N_2O_4 \cdot 0.7\,H_2O$ (374.99): calcd C 67.26, H 5.21, N 7.47; found C 67.22, H 4.99, N 7.03.

13,14-Dichloro-6-methyldibenzo[*b*,*j*][4,7]phenanthroline (**6**): Compound **5** (17.5 g, 48.3 mmol) and phosphorous oxychloride (135 mL, 1.45 mol) were refluxed under nitrogen for 3.5 h. $POCl_3$ was removed under vacuum, CH_2Cl_2(250 mL) was added, and the solution was slowly poured into cold 15 % NH_4OH (200 mL). The organic layer was decanted and the aqueous layer was extracted with CH_2Cl_2 (3 × 100 mL). The mixed organic layers were dried over Na_2SO_4 and evaporated. The crude product was purified by chromatography on alumina eluted with CH_2Cl_2/MeOH (99:1), followed by recrystallization from $CHCl_3$, yielding 5.01 g (28 %) of a white powder. $R_f = 0.92$ (alumina, CH_2Cl_2 99 %, MeOH 1 %); m.p. 188 °C; ^{1}H NMR (200 MHz, $CDCl_3$): $\delta = 2.76$ (d, $J = 1.3$ Hz, 3H), 7.64–7.75 (2td and 1br s, $J_1 = 1.6$ Hz, $J_2 = 5.4$ Hz, 3H), 7.80–7.89 (2td, $J_1 = 1.6$ Hz, $J_2 = 6.9$ Hz, 2H), 8.18–8.29 (2d, $J = 13.5$ Hz, 2H), 8.43–8.49 (2d, $J = 8.4$ Hz, 2H); ^{13}C NMR (200 MHz, $CDCl_3$): $\delta = 18.4, 120.3, 120.4, 124.6, 124.7, 124.8, 127.2, 127.6, 129.0, 129.7, 130.5, 130.7, 131.5, 141.0, 141.3, 141.4, 147.3, 147.9, 150.5, 150.7$; $C_{21}H_{12}N_2Cl_2 \cdot 0.12\,CHCl_3$ (377.57): calcd C 67.19, H 3.24, N 7.42; found C 67.27, H 3.17, N 7.49.

6-Methyldibenzo[*b*,*j*][4,7]phenanthroline (**7**): Compound **6** (200 mg, 0.55 mmol) in dry THF (5 mL) was added dropwise under nitrogen to a refluxing solution of $LiAlH_4$ (167 mg, 4.4 mmol) in dry THF (5 mL) for 15 min. The reaction mixture was refluxed for 10 h and the deep blue solution was allowed to cool to room temperature. The excess of hydride was slowly hydrolyzed with THF/H_2O (1:1). THF was removed in vacuo and the solution was extracted with CH_2Cl_2. After drying over Na_2SO_4, an orange-red solid was obtained that contained a mixture of **6** and partially reduced derivatives. The solid was refluxed in ethanol (25 mL), and $FeCl_3$ (600 mg, 2.2 mmol) dissolved in water (10 mL) was slowly added. The solution was refluxed for 15 h then allowed to cool and 15 % NH_4OH was added. The black precipitate was filtered and washed with methanol. The alcoholic solvents were removed under vacuum from the filtrate, and water was then extracted with CH_2Cl_2. The organic layer was dried over Na_2SO_4 and evaporated, giving a brown product that was recrystallized from $CHCl_3$ to yield 142 mg (87 %) of yellow needles. M.p. 174 °C; ^{1}H NMR (200 MHz, $CDCl_3/CD_3OD$ 2:1): $\delta = 2.83$ (d, $J = 1.1$ Hz, 3H), 7.55–7.65 (2td, 2H), 7.74–7.83 (2td, 3H), 7.88 (d, J = 1.0 Hz, 1H), 8.06 (2d, 2H), 8.10 (d, 1H), 8.23 (d, 1H), 9.33 (s, 1H), 9.34 (s, 1H); $C_{21}H_{14}N_2 \cdot 0.035\,CHCl_3$ (298.53): calcd C 84.63, H 4.74, N 9.38; found: C 84.66, H 4.79, N 9.41.

6-(*n*-Propylaminomethyl)dibenzo[*b*,*j*][4,7]phenanthroline (**4a**): Compound **7** (1.0 g, 3.4 mmol), *N*-bromosuccinimide (605 mg, 3.4 mmol), and perbenzoic anhydride (50 mg, 0.21 mmol) were refluxed in CCl_4 (75 mL) for 10 h. After cooling, the precipitate was filtered, washed with CCl_4, and dissolved in CH_2Cl_2. The solution was extracted with a 1 % Na_2CO_3 aqueous solution and dried over Na_2SO_4. The solvent was evaporated to yield 982 mg of the 6-bromomethyl derivative (white powder) of sufficient purity for further reactions. M.p. > 250 °C (decomp); ^{1}H NMR (200 MHz, $CDCl_3/CD_3OD$ 2:1): $\delta = 5.31$ (s, 2H), 7.66 (m, 2H), 7.85 (m, 2H), 8.12–8.27 (m, 5H), 9.49 (s, 1H), 9.50 (s, 1H). The above crude bromomethyl compound (100 mg, 0.27 mmol) was heated to 50 °C in *n*-propylamine (5 mL) for 4 h. The excess amine was removed under vacuum, CH_2Cl_2 was added, and the solution was extracted with a 5 % Na_2CO_3 aqueous solution. The organic layer was dried over Na_2SO_4 and evaporated. The crude product was recrystallized from 1 N HCl/THF, yielding 74 mg (50 % from **7**) of a yellow powder. M.p. > 240 °C (decomp); ^{1}H NMR (200 MHz, CD_3OD/D_2O 2:1): $\delta = 1.07$ (t, 3H), 1.86 (m, 2H), 3.20 (t, 2H), 4.60 (s, 2H), 7.45–8.03 (m, 9H), 9.15 (s, 1H), 9.18 (s, 1H); UV/Vis (H_2O, pH = 7.1): λ_{max} (ε) = 246 (56 400), 306 (31 700), 326 nm (35 000); fluorescence (H_2O, pH = 7.1, $\lambda_{exc} = 326$ nm): $\lambda_{em} = 413$, 436, 463 nm; $C_{24}H_{21}N_3 \cdot 2\,HCl$ (424.37): calcd C 67.93, H 5.46, N 9.90; found C 67.77, H 5.60, N 9.79.

6-Di[(3-dimethylaminopropyl)aminomethyl]dibenzo[*b*,*j*][4,7]phenanthroline (**4b**): Compound **4b** was synthesized in the same manner as described above for **4a** from the crude 6-bromomethyl derivative (114 mg, 0.27 mmol) and 3-(dimethylamino)propylamine (7.7 mL). Compound **4b** · HCl was obtained by adding 1 N HCl to the free amine and subsequent precipitation with THF to yield 66 mg (33 % from **7**) of a yellow powder. M.p. > 240 °C (decomp); ^{1}H NMR (200 MHz, D_2O, ref DSS): $\delta = 2.43$ (m, 2H), 3.05 (s, 6H), 3.43 (m, 4H), 4.78 (s, 2H), 7.35–7.60 (m, 7H), 7.80 (d, 1H), 7.83 (s, 1H), 8.41 (s, 1H), 8.84 (s, 1H); ^{13}C NMR (200 MHz, D_2O, ref DSS): $\delta = 21.5, 43.2, 45.1, 48.8, 54.6, 120.5, 121.5, 122.6, 126.2, 127.1, 128.1, 128.8, 129.5, 132.1, 132.6, 135.5, 137.0, 137.6, 139.7, 140.6, 143.6, 146.4$; $C_{26}H_{26}N_4 \cdot 3\,HCl \cdot 0.3\,H_2O$ (509.31): calcd C 61.32, H 5.86, N 11.00; found C 61.26, H 5.99, N 11.06.

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[1] T. Le Doan, L. Perrouault, D. Praseuth, N. Habhoub, J.-L. Decout, N. T. Thuong, J. Lhomme, C. Hélène, *Nucleic Acids Res.* **1987**, *15*, 7749.
[2] H. E. Moser, P. B. Dervan, *Science* **1987**, *238*, 645.
[3] N. T. Thuong, C. Hélène, *Angew. Chem.* **1993**, *105*, 697; *Angew. Chem. Int. Ed. Engl.* **1993**, *32*, 666.
[4] J.-S. Sun, T. Garestier, C. Hélène, *Curr. Opin. Struct. Biol.* **1996**, *6*, 327.
[5] P. V. Scaria, R. H. Shafer, *J. Biol. Chem.* **1991**, *266*, 5417.
[6] J. L. Mergny, D. Collier, M. Rougée, T. Montenay-Garestier, C. Hélène, *Nucleic Acids Res.* **1991**, *19*, 1521.
[7] J. L. Mergny, G. Duval-Valentin, C. H. Nguyen, L. Perrouault, B. Faucon, M. Rougée, T. Montenay-Garestier, E. Bisagni, C. Hélène, *Science* **1992**, *256*, 1681.
[8] J. S. Lee, L. J. P. Latimer, K. J. Hampel, *Biochemistry* **1993**, *32*, 5591.
[9] W. D. Wilson, F. A. Tanious, S. Mizan, S. Yao, A. S. Kiselyov, G. Zon, L. Strekowski, *Biochemistry* **1993**, *32*, 10614.
[10] K. R. Fox, P. Polucci, T. C. Jenkins, S. Neidle, *Proc. Natl. Acad. Sci. USA* **1995**, *92*, 7887.
[11] L. J. P. Latimer, N. Payton, G. Forsyth, J. S. Lee, *Biochem. Cell. Biol.* **1995**, *73*, 11.
[12] Y. W. Park, K. J. Breslauer, *Proc. Natl. Acad. Sci. USA* **1992**, *89*, 6653.
[13] M. Durand, N. T. Thuong, J. C. Maurizot, *J. Biol. Chem.* **1992**, *267*, 24394.
[14] M. Durand, N. T. Thuong, J. C. Maurizot, *J. Biomol. Struct. Dyn.* **1994**, 1191.
[15] D. S. Pilch, K. J. Breslauer, *Proc. Natl. Acad. Sci. USA* **1994**, *91*, 9332.

[16] D. S. Pilch, M. J. Waring, J.-S. Sun, M. Rougée, C. H. Nguyen, E. Bisagni, T. Garestier, C. Hélène, *J. Mol. Biol.* **1993**, *232*, 926.
[17] D. S. Pilch, M. T. Martin, C. H. Nguyen, J.-S. Sun, E. Bisagni, T. Garestier, C. Hélène, *J. Am. Chem. Soc.* **1993**, *115*, 9942.
[18] C. Escudé, C. H. Nguyen, J. L. Mergny, J.-S. Sun, E. Bisagni, T. Garestier, C. Hélène, *J. Am. Chem. Soc.* **1995**, *117*, 10212.
[19] O. Baudoin, M.-P. Teulade-Fichou, J.-P. Vigneron, J.-M. Lehn, *J. Org. Chem.* **1997**, *62*, 5458.
[20] M. Césario, M.-P. Teulade-Fichou, O. Baudoin, J.-P. Vigneron, J.-M. Lehn, unpublished data.
[21] C. Escudé, C.-H. Nguyen, S. Kukreti, Y. Janin, J.-S. Sun, E. Bisagni, T. Garestier, C. Hélène, *Proc. Natl. Acad. Sci. USA* **1998**, *95*, 3591.
[22] C.-H. Nguyen, C. Marchand, S. Delage, J.-S. Sun, T. Garestier, C. Hélène, E. Bisagni, *J. Am. Chem. Soc.* **1998**, *120*, 2501.
[23] S. K. Kim, J.-S. Sun, T. Garestier, C. Hélène, E. Bisagni, A. Rodger, B. Nordén, *Biopolymers* **1997**, *42*, 101.

  0947-6539/98/0408-1508 $ 17.50+.50/0