Interaction of Monomolecular G4-DNA Nanowires with TMPyP: Evidence for Intercalation†

Irit Lubitz,‡ Natalia Borovok,‡ and Alexander Kotlyar*,‡,§

Department of Biochemistry, George S. Wise Faculty of Life Sciences, and Nanotechnology Center, Tel Aviv University, Ramat Aviv, 69978 Israel

Received July 2, 2007; Revised Manuscript Received September 5, 2007

ABSTRACT: Interaction of *meso*-tetrakis(4-*N*-methylpyridyl)porphyrin (TMPyP) with G4-wires composed of ∼1000 stacked tetrads (Kotlyar, A. B., Borovok, N., Molotsky, T., Cohen, H., Shapir, E., and Porath, D. (2005) Long monomolecular G4-DNA nanowires, *Adv. Mater. 17*, 1901−1905) was studied. These wires exist in either K (Na)-free or K forms in contrast to short telomeric G-quadruplexes, which are stable only in the presence of monovalent cations. We showed that a stable complex between K-free G4-wires and the porphyrin is formed at a TMPyP to tetrad molar ratio of 0.5. A 19 nm shift and a hypochromicity of 58% in the absorption spectrum, the induced CD of the porphyrin, and efficient energy transfer between TMPyP and K-free G4-wires suggest an intercalative mechanism of TMPyP binding. The K form interacts with TMPyP much weaker than the K-free form of the wires. Binding of TMPyP to the K form is characterized by a small (3 nm) shift of the Soret band, a weak positive induced CD in the Soret region, and the absence of energy transfer between the G-bases and the porphyrin. These parameters reflect a nonintercalative binding of TMPyP to the K form of the wires. We suggest that K ions positioned in the center space between the adjacent tetrads limit the access of TMPyP and other organic molecules to this region, thus enabling only nonintercalative modes of ligand binding to G-quadruplex DNAs.

5,10,15,20-Tetrakis(*N*-methyl-4-pyridyl)porphyrin (TMPyP),[1] a commercially available planar cationic porphyrin, is known to bind and stabilize different types of G-quadruplex structures (*2*−*7*). These structures are formed as a result of self-assembly of various G-rich telomeric oligonucleotides. Depending on the sequence and the amount of G-bases in the oligonucleotide, G-quadruplexes may be intermolecular, consisting of four oligonucleotide strands or two hairpin strands, or intramolecular, consisting of a folded single strand. Two main models have been proposed for binding of TMPyP to different types of G-quadruplexes, namely, intercalative binding between adjacent G-tetrads (*6*−*8*) and end-stacking on either or both of the terminal G-tetrads (*2*−*4*, *8*). The porphyrin ring of TMPyP has a size similar to that of the G-tetrad and thus is a good fit for stacking with G-tetrads. However, intercalation of the porphyrin moiety between adjacent tetrads is considered to be less favored than external binding (*2*−*4*). Recently, an X-ray structure of a TMPyP−G4-DNA complex was presented (*9*). These X-ray data are consistent with the nonintercalative mode of TMPyP binding to G-quadruplexes; in the crystal structure TMPyP molecules are not directly interacting with G-tetrads in the G-quadruplex (*9*).

We have recently reported the synthesis of novel continuous monomolecular G-quadruplex DNA nanostructures composed of single self-folded poly(dG) strands and containing hundreds of stacked tetrads (*1*). These structures are spontaneously formed in aqueous solutions at relatively low concentrations of long (kilobases) G-strands. The intramolecular fashion of the G-strand folding as well as the G-quadruple nature of the structures was proposed on the basis of optical absorption, CD, electrophoresis, and AFM morphology imaging data (*1*). These wires, in contrast to short telomeric G-quadruplex DNA, are stable in the absence of "stabilizing" cations. The latter structures are stable only in the presence of stabilizing cations (K^+ or Na^+) (*10*−*14*); removal of the cations results in a complete unfolding of G-quadruplexs. The K-free G-wires can be transferred to a K (Na) form by addition of the cation (*1*). The ability to obtain G4 wires in either K-free or K (Na) forms makes it possible to study the influence of the center ion on the mode of TMPyP interaction with G-quadruplexes.

Here we studied the interaction of TMPyP with K-free and K forms of continuous monomolecular G4-wires composed of more than 1000 stacked tetrads. UV/vis absorption, CD, and fluorescent spectroscopy were used to determine the stoichiometry and the mode of TMPyP binding to the wires. We showed that TMPyP binds strongly to the K-free form of the wires by an intercalative mechanism and binds weakly to the K form of the wires via outside, nonintercalative modes. We suggested that the presence of K^+ in the space between the guanine planes prevents intercalation of aromatic molecules into G-guadruplexes.

† This work is supported by a European Grants for Future & Emerging Technologies (IST-2001-38951−"DNA-Based Nanowires" and FP6-291292−"DNA-Based Nanodevices").

* To whom correspondence should be addressed. Phone: +972-3-6407138. Fax: +972-3-6406834. E-mail: s2shak@post.tau.ac.il.

‡ George S. Wise Faculty of Life Sciences.

§ Nanotechnology Center.

[1] Abbreviations: TMPyP, *meso*-tetrakis(4-*N*-methylpyridyl)porphyrin; AFM, atomic force microscopy; CD, circular dichroism.

10.1021/bi701301u CCC: $37.00
Published on Web 10/23/2007

MATERIALS AND METHODS

Materials. Unless otherwise stated, reagents were obtained from Sigma-Aldrich and were used without further purification. TMPyP was dissolved in doubly distilled water. The stock solution was passed through a 0.22 μm filter, and the concentration of the porphyrin was determined using the extinction coefficient (226 mM^{-1} cm^{-1}) at 422 nm (*15*).

DNA Samples. The long monomolecular G4-wires were prepared essentially as described in our recent paper (*1*); the procedure includes the following steps: enzymatic synthesis of 5000 bp poly(dG)−poly(dC) molecules (*16*), separation of the poly(dG) and the poly(dC) strands in 0.1 M LiOH using HPLC, and folding of the purified poly(dG) strands by lowering the pH of the solution. The wires were prepared in the absence of stabilizing ions (K^+ and Na^+) and thus will be termed the "K-free" G4-wires throughout this paper. The K-free G4-wires were converted to the K form by a 30 min incubation in 10 mM KCl, 2 mM Tris/HCl buffer (pH 7.5) at 25 °C.

The TMPyP−DNA complex was prepared by incubation of the K-free G4-wires with an amount of TMPyP equal to that of G-tetrads in the wire in 100 mM Tris/HCl buffer (pH 7.5) for 1 h at 25 °C. The complex was subsequently passed through a Sephadex G-25 column (1 × 5 cm) equilibrated with 100 mM Tris/HCl buffer (pH 7.5). The amount of TMPyP eluted in the void volume of the column in complex with DNA was measured by absorption spectroscopy (see below); a ratio of 0.5 was estimated for the complex between the porphyrin and G-tetrad. A complex of TMPyP with K^+-containing G4-wires was made by incubation of TMPyP with half the amount of G-tetrads in the wire in 100 mM Tris/HCl buffer (pH 7.5) for 1 h at 25 °C.

Absorption, CD, and Fluorescent Spectroscopy. Absorption spectra of G4-wires were recorded with a U2000 Hitachi (Japan) spectrophotometer. The G-tetrad concentration was estimated using the extinction coefficient (36 mM^{-1} cm^{-1}) at 260 nm. The coefficient is smaller than that of the sum of four independent G-bases (46.8 mM^{-1} cm^{-1}), due to the hypochromic effect caused by the stacking interaction between G-bases in the wire. Addition of 0.1 M LiOH results in complete unfolding of the G4-wire and as a result in an increase of absorption. A hyperchromicity value of 1.3 (30%) was measured by comparing the G-wire absorption at alkaline and neutral pH (a correction was made to the spectrum to account for the changes induced by deprotonation of G-bases in LiOH). The extinction coefficient of the G-tetrad was thus estimated as the sum of the coefficients of four independent G-bases divided by 1.3 (the hyperchromicity value) and is equal to 36 mM^{-1} cm^{-1}. CD spectra were recorded at 25 °C with an Aviv model 202 series (Aviv Instrument Inc.) circular dichroism spectrometer. Each spectrum was recorded from 220 to 500 nm and was an average of four measurements. Recording specifications were wavelength step 1 nm, settling time 0.333 s, average time 1.0 s, bandwidth 1.0 nm, and path length 1 cm. Fluorescence spectra were recorded with a FluoroMax (Jobin Yvon) spectrofluorometer using a 150 W arc xenon lamp as a light source. The spectra were recorded in a 0.4 cm × 1 cm quartz cell with a band-pass of 5 nm at the excitation side and 5 nm at the emission side. The contribution of buffer to the measured fluorescence was subtracted. In all cases a Schott UV305 filter was used on the excitation side. All measurements were conducted in 100 mM Tris/HCl (pH 7.5); the absorption of all samples was 0.3 or less.

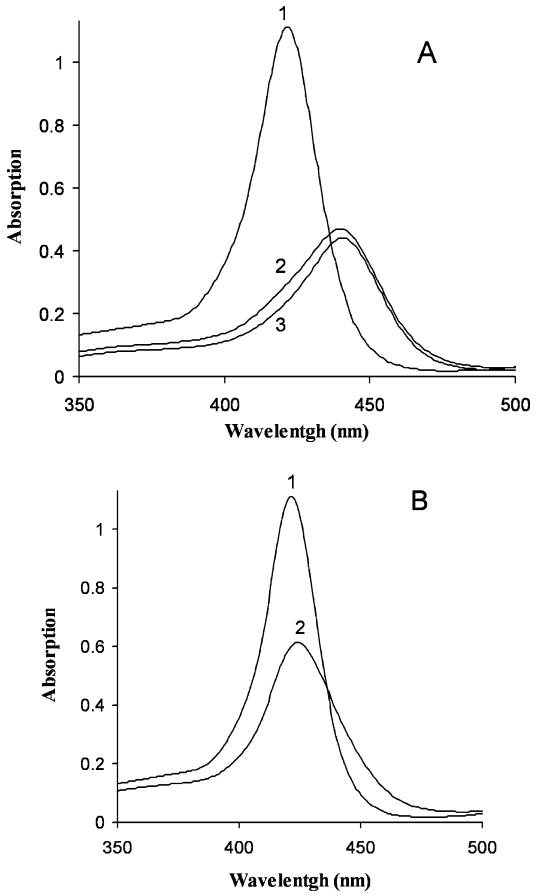

FIGURE 1: Visible absorption spectra of TMPyP in complex with K-free and K forms of G4-wires. A 5 μM concentration of TMPyP (line 1 in panels A and B) in 100 mM Tris/HCl (pH 7.5) was incubated for 30 min with 10 μM (in tetrads) K-free G4-wires (panel A, line 2); the mixture was passed through a G-25 Sephadex column as described in the Materials and Methods (line 3 in panel A). The latter spectrum is multiplied by a factor 1.6 to account for the sample dilution during chromatography. A 10 μM concentration (in tetrads) of G4-wires was preincubated for 30 min at 25 °C with 10 mM KCl prior to addition of 5 μM TMPyP (panel B, line 2). The spectra were recorded 15 min after addition of the porphyrin.

RESULTS

Figure 1A shows the UV/vis absorption spectra of TMPyP in the presence and the absence of the K-free G4-wires. The ratio between the amount of the porphyrin and the tetrads in the sample (*r*) is equal to 0.5. The concentrations of TMPyP and G-tetrads were determined from extinction coefficients of 226 mM^{-1} cm^{-1} at 422 nm (*15*) and 36 mM^{-1} cm^{-1} at 260 nm, correspondingly (see the Materials and Methods). As seen in Figure 1A (compare line 1 and line 2) a substantial shift in the Soret band peak position (from 422 to 441 nm) and a substantial hypochromicity of the Soret band (58%) take place upon binding of the porphyrin to the wires. The complex between TMPyP and G4 wires is very stable and does not dissociate during size-exclusion chromatography. The absorption spectrum of the DNA fraction that eluted from a Sephadex G-25 column is almost identical to that of the initial complex (compare line 2 and line 3, Figure 1A). The increase of *r* beyond 0.5 results in progressive reduction

Table 1: Size-Exclusion Chromatography of G4-Wire TMPyP Complexes

TMPyP to tetrad ratio	
before the column	after the column[a]
0.4	0.39 ± 0.2
0.5	0.48 ± 0.2
0.6	0.49 ± 0.2
0.8	0.50 ± 0.2

[a] The K-free G4-wires were incubated with TMPyP in different ratios in 100 mM Tris/HCl (pH 7.5) for 1 h at 25 °C. The complex was subsequently passed through a Sephadex G-25 column (1 × 5 cm) equilibrated with 100 mM Tris/HCl buffer (pH 7.5). The TMPyP to tetrad ratio in the complex eluted in the void volume of the column was measured by absorption spectroscopy as described in the Materials and Methods. All numbers represent an average of 3−5 measurements.

of the shift in the Soret band and the hypochromicity compared to those shown in Figure 1A (data not presented). When an excess of TMPyP ($r > 0.5$) was added to the wires, the porphyrin was eluted from the Sephadex G-25 column as two well-separated peaks. The fraction that was eluted in the void volume of the Sephadex column corresponds to TMPyP bound to DNA; the second fraction that was eluted much later corresponds to free TMPyP. The G-tetrad to TMPyP ratio in the former fraction does not exceed 0.5, regardless of the amount of TMPyP that was added to the sample before chromatography (see Table 1). This chromatographic behavior is consistent with the formation of a high-affinity complex between TMPyP and K-free G4-wires in which one porphyrin residue is bound per two tetrads of the wire.

Binding of TMPyP to the G4-wires is strongly affected by K ions. All measurements with K-free and K forms of the wires were conducted under similar ionic strength conditions. Preincubation of the K-free wires for 30 min with 10 mM KCl followed by the addition of TMPyP results in a small red shift of the Soret band (from 422 to 425 nm) and a smaller reduction of the peak intensity (~45%) compared to that induced by binding to the K-free G-wires (compare parts A and B of Figure 1). The complex formed between TMPyP and G4-wires in the presence of K^+ is unstable and completely dissociates during column chromatography. The DNA fraction that eluted from the Sephadex G-25 column in 100 mM Tris/HCl buffer (pH 7.5) does not contain TMPyP (data not shown). These data indicate that the cation and TMPyP compete with one another for binding to G4-wires. In the presence of K ions no tight binding of TMPyP to the wires takes place. On the contrary the K-free form of G4-wires binds the porphyrin very tightly. The high affinity leads to the slow (hours) release of TMPyP from the K-free form of the DNA. The much higher affinity of G4-wires to TMPyP compared to K ions together with the slowness of the porphyrin dissociation results in an inability of the cation at 10 mM concentration to replace the porphyrin even during long (several hours) incubation at 25 °C. It has been shown for duplex DNA that a large red shift and hypochromicity of the Soret band are characteristic of porphyrin intercalation. On the other hand, the outside-binding mode of porphyrins to DNA is characterized by a smaller band shift and lower hypochromicity (*17*, *18*). A strong effect of K ions on the magnitude of the spectral shift (see Figure 1) suggests different modes of TMPyP interaction with K-free and K forms of G4-wires.

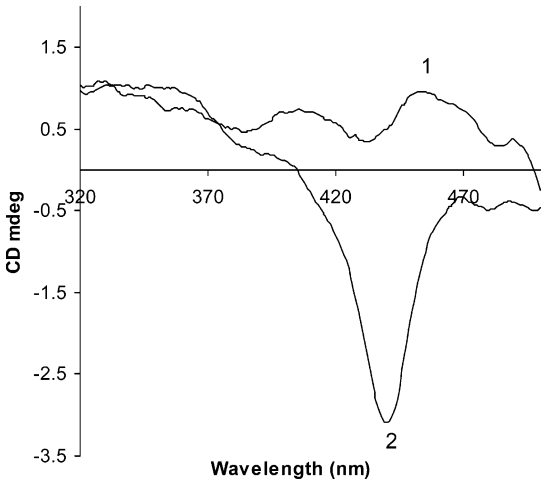

FIGURE 2: Induced CD spectra in the Soret region of TMPyP in the presence of K-free and K forms of G4-wires. An 8 μM concentration of TMPyP was incubated for 30 min in 100 mM Tris/HCl, pH 7.5, with 16 μM (in tetrads) K-free G4-wires (line 2). G4-wires (16 μM in tetrads) were preincubated for 30 min at 25 °C with 10 mM KCl prior to the addition of 8 μM TMPyP (line 1). The spectra were recorded as described in the Materials and Methods 15 min after addition of the porphyrin.

To further clarify the mode of TMPyP binding to both forms of G4-wires, the induced CD spectra of TMPyP were measured. Although TMPyP is achiral, CD signals are induced once it is bound to the chiral wires. Figure 2 shows the induced CD signals for complexes of TMPyP with G4-wires in the absence and the presence of K ions (line 2 and line 1, correspondingly). The molar TMPyP to tetrad ratio in both cases is equal to 0.5. As seen in Figure 2, TMPyP shows a negative peak in the Soret region in the case of K-free G4-wires (line 2) and a broad positive ellipticity in the case of K^+-containing G4-wires (line 1). It was shown (*17*−*19*) that the sign of the induced CD spectra in the Soret region of porphyrins bound to nucleic acids provides important information about their binding modes: a positive induced CD band is indicative of outside binding, and a negative induced CD band is produced upon intercalation. Thus, the data presented in Figure 2 are consistent with intercalation of TMPyP residues between G-tetrads in the K-free wire and outside binding of the porphyrin to G4-wires in the presence of K ions (see Figure 2).

The energy transfer experiments further support an intercalative binding of TMPyP to K-free G4-wires and a nonintercalative binding of the porphyrin to G4-wires complexed with K ions. It was demonstrated earlier (*20*) that energy transfer between DNA bases and a bound fluorescent ligand is only possible if the ligand closely contacts the DNA bases. This happens when a ligand is intercalated between stacked base pairs in the DNA duplex, triads in triple DNA, and tetrads in G4-DNA (*21*−*25*). Energy transfer studies can thus distinguish between intercalation and outside binding of fluorophores to different types of DNA. As seen in Figure 3, the fluorescence emission of TMPyP solution at 660 nm is weak when excited between 240 and 320 nm, which is the DNA absorption region (Figure 3, line 1). However, the emission intensity of TMPyP is strongly enhanced in the presence of K-free G4-wires (Figure 3, line 2) as a consequence of energy transfer from tetrads to porphyrin residues. This enhancement is not observed in the presence of K^+ ions (Figure 3, line 3). These data clearly show that

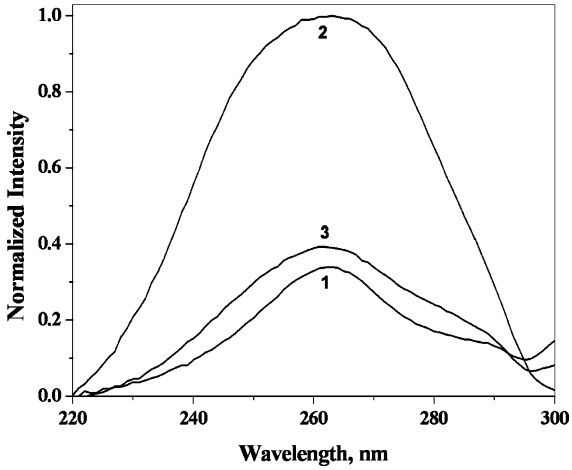

FIGURE 3: Energy transfer from G-bases to TMPyP. Excitation spectra (λ_{em} = 660 nm) of TMPyP (line 1) in the presence of K-free (line 2) and K (line 3) forms of G4-wires were recorded with a FluoroMax (Jobin Yvon) spectrofluorometer in a quartz cell (0.4 cm × 1 cm) with 5 nm excitation and 5 nm emission slits. The contribution of buffer was subtracted from the fluorescence spectra. In all cases, a Schott UV305 filter in front of the emission monochromator was used to block scattering of excitation light. All measurements were conducted in 100 mM Tris/HCl (pH 7.5); the absorption of all samples was 0.3.

significant transfer of energy occurs from excited G-bases to TMPyP, thus suggesting an intercalative mode of TMPyP binding to K-free G4-wires. The absence of energy transfer between G-bases and the porphyrins in the K form of G4-wires is a strong indication for a nonintercalative mode of TMPyP binding to these wires.

DISCUSSION

A number of studies have investigated the nature and stoichiometry of TMPyP interaction with different forms of G-quadruplexes. Various models of TMPyP binding to G-quadruplexes have been proposed on the basis of spectroscopic, CD, fluorescent, and NMR studies (*2*, *4*, *6*−*8*). The planar structure of the porphyrin, the molecular dimensions that resemble those of a G-tetrad, and hydrophobicity of the central part produce conditions favorable for intercalation of TMPyP into the guanine tetrads. However, experimental and theoretical data suggest that G-tetraplexes mainly accommodate TMPyP via external end-stacking with the outer layer of the guanine tetrad on either one end or both ends of the tetraplex, but not via intercalation (*2*−*4*). The X-ray structure of a TMPyP−G4-DNA complex (*9*) also confirmed a nonintercalative mode of TMPyP binding to G-quadruplexes. In the crystal structure, TMPyP molecules do not directly interact with G-tetrads in the G-quadruplex. The authors suggested that the absence of intercalation is due to relatively low selectivity of TMPyP binding to quadruplex DNAs with respect to double-stranded DNA. The other possibility is that univalent alkaline cations (K^+ or Na^+) positioned between adjacent tetrad planes in G-quadruplexes (*10*−*14*) prevent intercalation of TMPyP. As clearly evident from X-ray images of the TMPyP−G4-DNA complex (*9*), the center space between the adjacent tetrads is occupied by K ions.

We have recently produced long, continuous G4-DNA wires composed of hundreds of stacked tetrads that, in contrast to short telomeric G-quadruplexes (for reviews see, e.g., refs *26*−*29*), are stable in the absence of "stabilizing" cations (*1*) and exist in either K-free or K forms. We have demonstrated that preincubation of TMPyP with the K-free form of G4-wires results in formation of a strong complex between the wires and the porphyrin that does not dissociate during size-exclusion chromatography at relatively high (in 100 mM Tris/HCl) ionic strength. The porphyrin to tetrad ratio in this high-affinity complex is equal to 0.5. Incubation of TMPyP with G4-wires in the presence of K ions results in a much weaker complex that is completely dissociated on the Sephadex column in 100 mM Tris/HCl. We have shown that the mode of TMPyP interaction with K-free and K forms of G4-wires is entirely different. It is well-established that intercalation of porphyrins into DNA is typically indicated by large red shifts of the Soret band, a negative induced CD band in the Soret region, and fluorescent energy transfer between the bases and the dye (*17*−*19*, *23*, *24*). On the other hand, outside binding without stacking is typically characterized by small red shifts of the Soret band, weak positive induced CD in the Soret region, and the absence of energy transfer between nucleic bases and ligands (*17*−*19*, *23*, *24*). Visible absorption, circular dichroism, and fluorescent energy transfer data presented here (see Figures 1−3) correspond nicely to intercalation of TMPyP into K-free G4-wires and to a nonintercalative outside binding of the porphyrin to the K form of the wires. As seen in Figure 1, a strong complex between the porphyrin and the K-free G4-wires is characterized by a 19 nm red shift and ∼60% hypochromism in the Soret band. In the case of TMPyP interaction with the K form of the wires the hypochromism at 425 nm is ∼45% and the absorption spectrum is shifted only by about 3 nm. A negative induced CD band in the Soret region and high-efficiency energy transfer (see Figures 2 and 3, lines 2) are observed for a strong complex of TMPyP with K-free G4-wires; the complex between TMPyP and the K form of the wires is characterized by a weak and broad positive band around 420

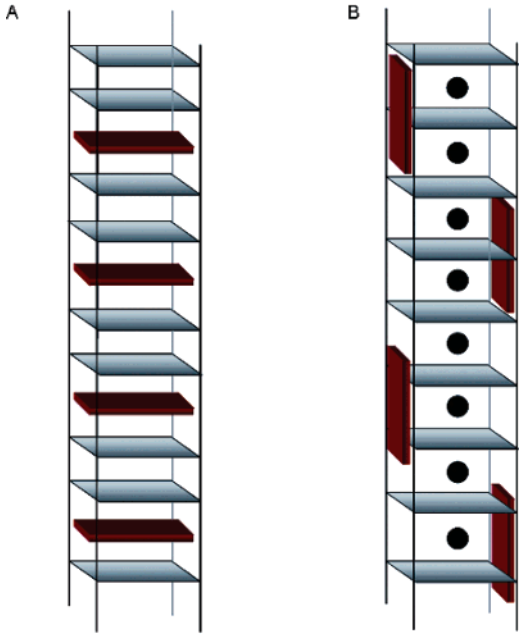

FIGURE 4: Schematic drawing of TMPyP interaction with K-free (A) and K (B) forms of the G4-wire. Porphyrins are illustrated as red boxes, tetrads as gray planes, and potassium ions as black spheres.

nm and the absence of energy transfer (see Figure 2, line 1, and Figure 3, line 3, correspondingly).

In summary, absorption, CD, and fluorescent data strongly indicate that TMPyP binds to the K-free form of G4-wires via intercalation with high affinity. K ions, if present in the center space between the adjacent tetrads, limit the access of TMPyP to this region, thus enabling only a nonintercalative mode of TMPyP binding to the K form of the wire (see Figure 4). The presence of K^+ (Na^+) ions between the stacked G-tetrads might also be the reason for the lack of intercalation of TMPyP and other organic molecules into short telomeric G-quadruplexes reported in the literature (*2*−*4*, *9*).

Guanine tetrads were proposed as building blocks of molecular nanowires for nanoelectronics (*28*, *29*). The wires that we invented (*1*) comprise a large number (hundreds) of stacked guanine tetrads providing better conditions for π-overlap compared to base pairs of the canonical double-stranded DNA. A high content of guanines, which have the lowest ionization potential among DNA bases, also makes charge migration through G4-wires highly probable. We have recently demonstrated that the wires are characterized by higher charge mobility as compared to double-stranded DNA (*30*). This observation makes G4-DNA a promising candidate for nanoelectronic applications. The intercalation of TMPyP into the wires demonstrated here might increase the π-stacking among the aromatic planes, G-tetrads, and porphyrin (see Figure 4) in the complex and as a result improve the conductivity of the wires. Metal-free and zinc porphyrins are photoactive and characterized by a long-lasting excited triplet state. The triplet state is characterized by a low redox potential and is capable of abstracting an electron at electrical potentials much lower than those of the ground state (*31*). This property of porphyrins will allow current to flow through TMPyP−G4 complexes at relatively low applied electrical potentials in the presence of light. Developing stable complexes of G4-wires with porphyrins and other intercalators which are capable of reversibly changing their electrical conductivity upon photoirradiation might be useful for electrical and electrooptical applications of the wires.

REFERENCES

1. Kotlyar, A. B., Borovok, N., Molotsky, T., Cohen, H., Shapir, E., and Porath, D. (2005) Long monomolecular G4-DNA nanowires, *Adv. Mater. 17*, 1901−1905.
2. Wheelhouse, R. T., Sun, D., Han, H., Han, F. X., and Hurley, L. H. (1998) Cationic porphyrins as telomerase inhibitors: the interaction of tetra-(*N*-methyl-4-pyridyl)porphine with quadruplex DNA, *J. Am. Chem. Soc. 120*, 3261−3262.
3. Han, F. X., Wheelhouse, R. T., and Hurley, L. H. (1999) Interactions of TMPyP4 and TMPyP2 with quadruplex DNA. Structural basis for the differential effects on telomerase inhibition, *J. Am. Chem. Soc. 121*, 3561−3570.
4. Han, H., Langley, D. R., Rangan, A., and Hurley, L. H. (2001) Selective interactions of cationic porphyrins with G-quadruplex structures, *J. Am. Chem. Soc. 123*, 8902−8913.
5. Kim, M. Y., Gleason-Guzman, M., Izbicka, E., Nishioka, D., and Hurley, L. H. (2003) The different biological effects of telomestatin and TMPyP4 can be attributed to their selectivity for interaction with intramolecular or intermolecular G-quadruplex structures, *Cancer Res. 63*, 3247−56.
6. Anantha, N.V., Azam, M,. and Sheardy, R.D. (1998) Porphyrin binding to quadrupled T4G4, *Biochemistry 37*, 2709−2714.
7. Haq, I., Trent, J. O., Chowdhry, B. Z., and Jenkins, T. C. (1999) Intercalative G-tetraplex stabilization of telomeric DNA by a cationic Porphyrin, *J. Am. Chem. Soc. 121*, 1768−1779.
8. Wei, C., Jia, G., Yuan, J., Feng, Z., and Li, C. (2006) A spectroscopic study on the interactions of porphyrin with G quadruplex DNAs, *Biochemistry 45*, 6681−6691.
9. Parkinson, G. N., Ghosh, R., and Neidle, S. (2007) Structural basis for binding of porphyrin to human telomeres, *Biochemistry 46*, 2390−2397.
10. Phillips, K., Dauter, Z., Murchie, A. I. H., Lilley, D. M. J., and Luisi, B. (1997) The crystal structure of a parallel- stranded parallel-stranded guanine tetraplex at 0.95 angstrom resolution, *J. Mol. Biol.* 273, 171−182.
11. Laughlan, G., Murchie, A. I. H., Norman, D. G., Moore, M. H., Moody, P. C .E., Lilley, D .M. J., and Luisi, B. (1994) The high-resolution crystal structure of a parallel-stranded guanine tetraplex, *Science 265*, 520−524.
12. Parkinson, G. N., Lee, M. P., and Neidle, S. (2002) Crystal structure of parallel quadruplexes from human telomeric DNA, *Nature (London)* 417, 876−880.
13. Burge, S., Parkinson, G. N., Hazel, P., Todd, A. K., and Neidle, S. (2006) Quadruplex DNA: sequence, topology and structure, *Nucleic Acids Res. 34*, 5402−5415.
14. Gu, J., and Leszczynski, J. (2002) Origin of Na^+/K^+ selectivity of the guanine tetraplexes in water: The theoretical rationale, *J. Phys. Chem. A 106*, 529−532.
15. Pasternack, R. F., Huber, P. R., Boyd, P., Engasser, G., Francesconi, L., Gibbs, E., Fasella, P., Venturo, G. C., and Hinds, L. C. (1972) Aggregation of meso-substituted water-soluble porphyrins, *J. Am. Chem. Soc. 94*, 4511−4517.
16. Kotlyar, A. B., Borovok, N., Molotsky, T., Fadeev, L., and Gozin, M. (2005) In vitro synthesis of uniform poly(dG)-poly(dC) by Klenow exo− fragment of polymerase I, *Nucleic Acids Res. 33*, 525−535.
17. Pasternack, R. F., Gibbs, E. J., Villafranca., and J. J. (1983) Interactions of porphyrins with nucleic acids, *Biochemistry 22*, 2406−2414.
18. Pasternack, R. F., Garrity, P., Ehrlich, B., Davis, C. B., Gibbs, E. J., Orloff, G., Giartosio, A., and Turano, C. (1986) The influence of ionic strength on the binding of a water soluble porphyrin to nucleic acids, *Nucleic Acids Res. 14*, 5919−5931.
19. Pasternack, R. F. (2003) Circular dichroism and the interactions of water soluble porphyrins with DNA-A, *Chirality 15*, 329−332.
20. Suh, D., and Chaires, J. B. (1995) Criteria for the mode of binding of DNA binding agents, *Bioorg. Med. Chem. 3*, 723−728.
21. Le Pecq, J. B., and Paoletti, C. (1967) A fluorescent complex between ethidium bromide and nucleic acids. Physical−chemical characterization, *J. Mol. Biol. 27*, 87−106.
22. Scaria, P. V., and Shafer, R. H. (1991) Binding of ethidium bromide to a DNA triple helix, *J. Biol. Chem. 266*, 5417- 5423.
23. Sehlstedt, U., Kim, S. K., Carter, P., Goodisman, J., Vollano, J. F., Nordon, B., and Dabrowiak, J. C. (1994) Interaction of cationic porphyrins with DNA, *Biochemistry 33*, 417−426.
24. Sari, M. A., Battioni, J. P., Dupre, D., Mansuy, D., and Le Pecq, J. B. (1990) Interaction of cationic porphyrins with DNA: Importance of the number and position of the charges and minimum structural requirements for intercalation, *Biochemistry 29*, 4205−4215.
25. Mergny, J. L., Duval-Valentin, G., Nguyen, C. H., Perrouault, L., Faucon, B., Rougeé, M., Montenay Garestier, T., Bisagni, E., and Hélène, C. (1992) Triple helix-specific ligands, *Science 256*, 1681−1684.
26. Gilbert, D. E. and Feigon, J. (1999) Multistranded DNA structures, *Curr. Opin. Struct. Biol. 9*, 305−314.
27. Kerwin, S. M. (2000) G-Quadruplex DNA as a target for drug design, *Curr. Pharm. Des. 6*, 441−471.
28. Keniry, M. A. (2001) Quadruplex structures in nucleic acids, *Biopolymers 56*, 123−146.
29. Davis, J. T. (2004) G-quartets 40 years later: from 50-GMP to molecular biology and supramolecular chemistry, *Angew. Chem., Int. Ed. 43*, 668−698.
30. Cohen, H., Sapir, T., Borovok, N., Molotsky, T., De Felice, R., Kotlyar, A. B., and Porath, D. (2007) Polarizability of G4-DNA observed by electrostatic force microscopy measurements, *Nano Lett. 7*, 981−986.
31. Chanon, M., Hawley, M. D., and Fox, M. A. (1988) in *Photoinduced Electron Transfer, Part A* (Fox, M. A., and Chanon, M., Eds.) pp 1−59, Elsevier, Amsterdam.

BI701301U