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Cation-Complexation Behavior of Template-Assembled Synthetic G-Quartets

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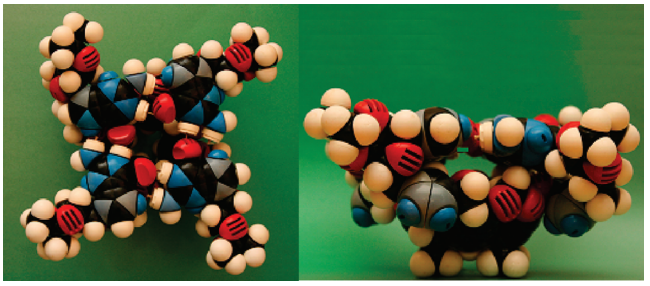

We report the preparation and solution study of a set of template-assembled synthetic G-quartets (TASQs) bound to different cations. These G-quartet baskets effectively extract cations of different sizes and valencies. They form isolated G-quartets with small cations such as Na^+ and Sr^{2+}, and dimeric assemblies with larger cations such as Cs^+. Their structures were determined by using ^1H NMR spectroscopy, and their sizes were evaluated by using a series of pulsed-field gradient NMR experiments. The effect of anion has been studied, and the cation selectivities have been investigated by a series of competition experiments.

Introduction

G-quartet chemistry is rapidly expanding as it becomes multidisciplinary.[1,2] While biological research is pointing to the important role these compounds play in cancer,[3,4] aging,[5] and genetic diseases,[6] supramolecular chemistry is exploring their functions in building new materials and devices. Lipophilic G-quartets have been developed for the latter purpose and show potential in making structures such as ion channels,[7] polymeric films,[8] liquid-crystalline phases,[9] and electronic devices.[10]

G-quartet is the common name given to the cyclic tetramers obtained from self-association of guanine (Figure 1a).[11] In the presence of metal cations, the G-quartet layers stack and form a columnar structure known as a G-quadruplex (Figure 1b).[12,13] The presence of cations is crucial for the stability and integrity of G-quadruplex assemblies.[14,15] Therefore, numerous studies have been conducted on

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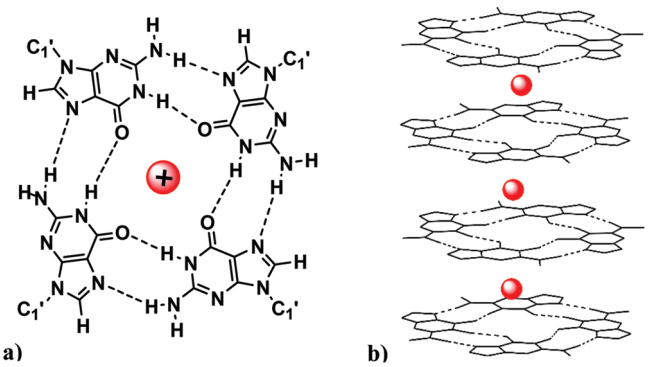

FIGURE 1. A schematic representation of (a) G-quartet and (b) G-quadruplex with bound cations. The G-quartet units are rotated about 30° with respect to each other.[2]

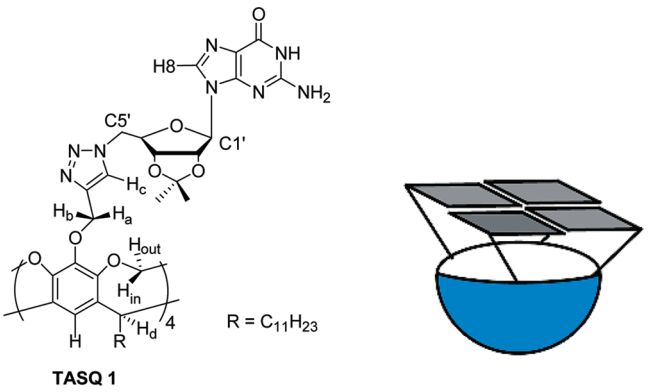

FIGURE 2. TASQ **1** and the corresponding basket-like structure.

various aspects of cation-binding such as selectivity,[16−18] location,[19−21] exchange,[22−24] and cation-dependent polymorphism.[25,26]

Recently, we reported the design, synthesis, and solution behavior of a set of lipophilic template-assembled synthetic G-quartets (TASQs, e.g., **1**)[27] where four guanines are covalently linked to a cavitand template such that the cavitand helps direct the guanines into a quartet. These TASQs are among a small number of systems reported to form cation-free G-quartets.[28−30] In addition to the contributions to stability offered by the cavitand template, TASQs can potentially control the morphology of G-quadruplex assemblies (e.g., parallel vs. antiparallel). This might be even more advantageous in hydrophilic systems in which preferential formation of one topology over other assemblies is desired. Many synthetic systems suffer from the competition of different topologies and the ensuing mixtures. We showed that preorganization of guanines via a cavitand template promotes Hoogsteen-type H-bonding and yields TASQs that manifest basket-like structures (Figure 2).

In the present study, we describe the morphologies and stoichiometries of the TASQ-cation assemblies as characterized largely by NMR spectroscopy. We found that prototypical lipophilic TASQs are capable of interacting with a wide range of cations, and are energetically and structurally sensitive to the nature of the cation used. Our present work also provides more insight into the formation and structural modification of isolated G-quartets. Isolated G-quartets are rare and consequently little is known concerning their behavior.[31]

Results and Discussion

Four different cations, Na^+ (1.02 Å), K^+ (1.38 Å), Sr^{2+} (1.18 Å), and Cs^+ (1.70 Å), were chosen for recognition analysis by TASQ **1** based on their sizes, valencies, and G-quartet binding properties.[32] The first two cations are commonly found in G-quadruplex assemblies, and the Sr^{2+} cation is also known to bind effectively to G-quadruplexes.[16] With respect to G-quadruplexes, the cesium cation is generally considered to be a nonconstructing ion: the large size of the Cs^+ cation prevents proper stacking of the bases, and results in weakening of the G-quadruplex.[14] Thus, it usually serves as a capping ion.[33] A series of solid−liquid extraction experiments were carried out on TASQ **1** in $CHCl_3$ with the solid picrate salt of the sample cations (see the Experimental Section). The products were subjected to ^1H NMR spectroscopic analysis in $CDCl_3$ (Figure 3).

In all cases, the cation-complexation was complete, resulting in the formation of a new set of signals and the complete loss of the signals due to the cation-free TASQ.[34] A comparison of the spectra revealed that each assembly has its own unique spectrum that depends on the identity of the corresponding cation. Na^+ and Sr^{2+} gave rise to spectra characterized by only one set of signals. They display all the common and specific features of the G-quartet formation that we have previously reported for the cation-free TASQs, including (a) H-bonding of the amino group, which is confirmed by the downfield shift of the H-bonded amino signal (NH_{2b}) and observed NOEs between NH/NH_{2b} and NH/NH_{2a} (Figure 4a),[35,36] (b) NOEs between H8 and NH_{2b}[37] (Figure 4a), and (c) changes in the chemical shifts of the nonexchangeable protons with respect to the spectra in DMSO-d_6.[27] 2D NOESY analysis revealed that the guanine bases in the assemblies of Na^+ and Sr^{2+} have been

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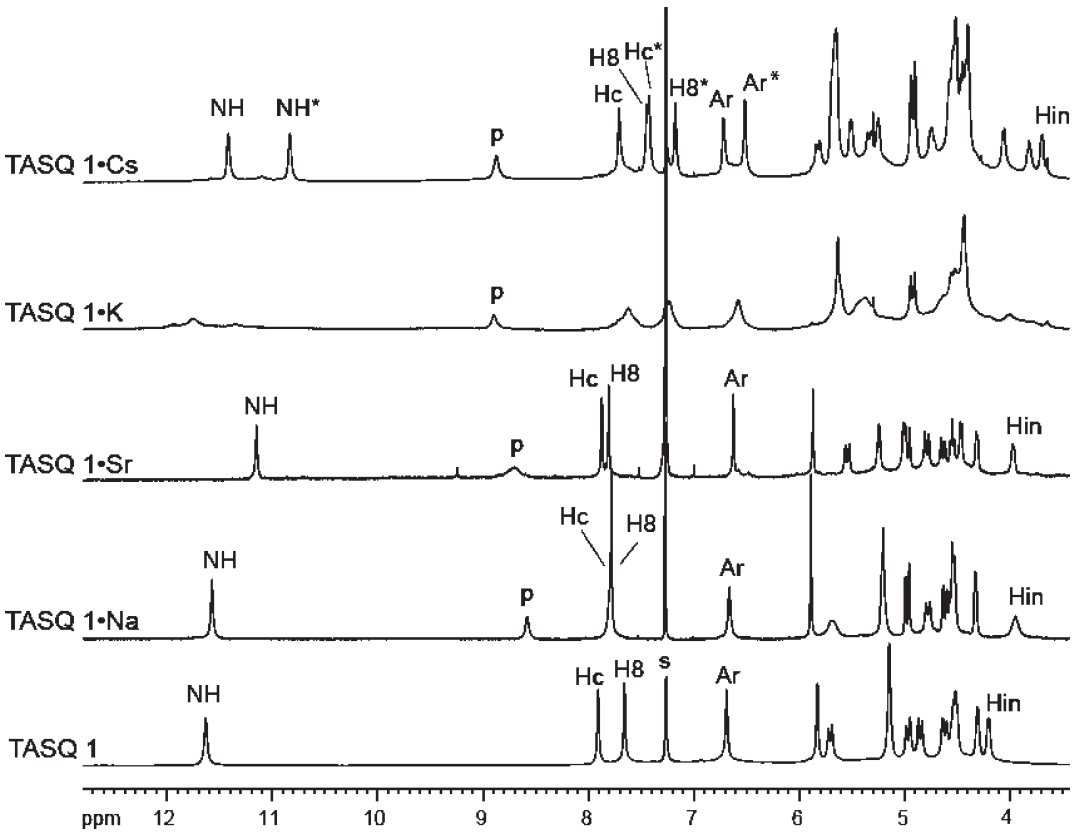

FIGURE 3. ^{1}H NMR spectra of TASQ·cation assemblies in $CDCl_3$ at 400 MHz at 25 °C. The signals labeled "p" and "s" indicate picrate and solvent, respectively. TASQ **1**·Cs^+ gave two sets of signals, one of which has been marked with asterisks.

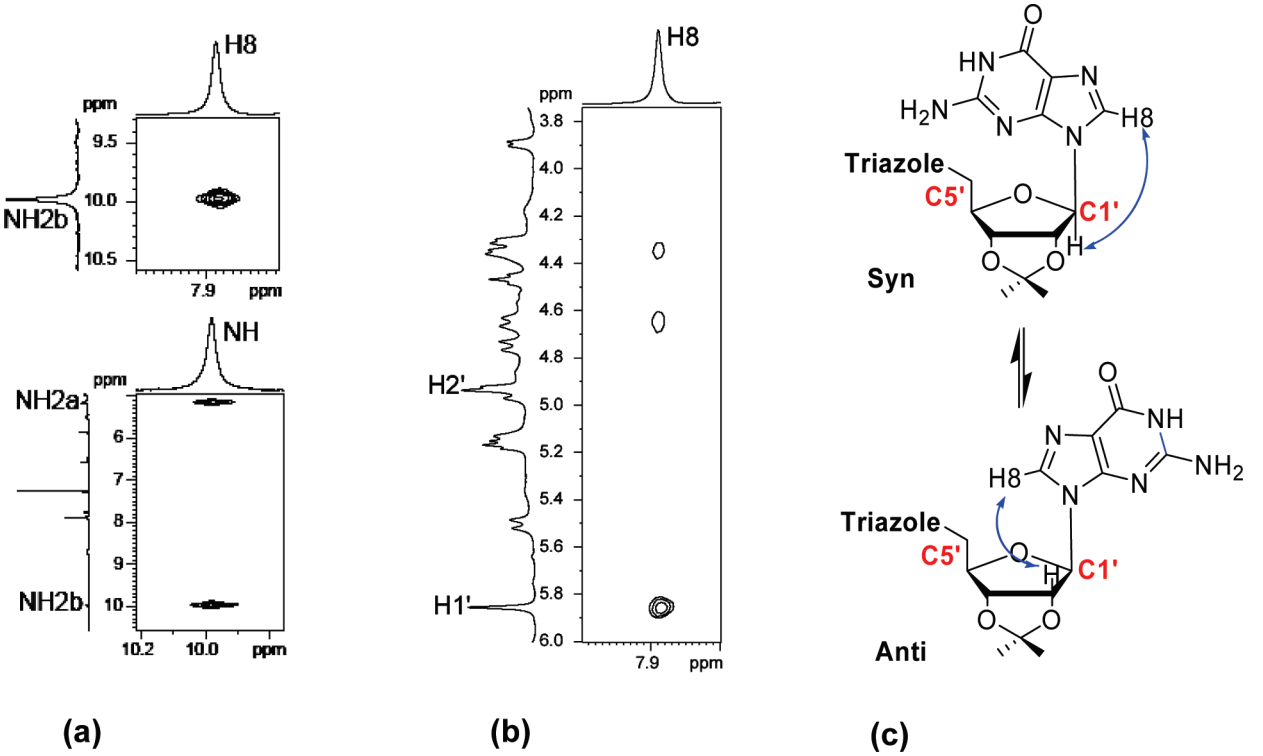

FIGURE 4. (a) Portions of a 100 ms NOESY spectrum of TASQ **1**·Sr^{2+} acquired at 400 MHz at −40 °C in $CDCl_3$ indicative of the formation of G-quartet, (b) NOEs indicative of the *syn* conformation, and (c) intrabase NOE correlations expected in *syn* (strong H1′/H8 and weak H2′/H8) and *anti* (medium H1′/H8 and strong H2′/H8) conformers.[35] The NH_2 signals which are too broad to observe at rt are visible at low temperature. Similar NOEs were observed for TASQ **1**·Na^+.[27]

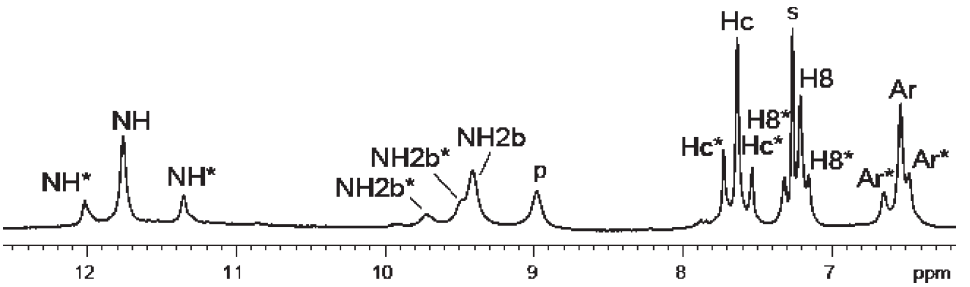

FIGURE 5. Partial ^{1}H NMR spectrum of TASQ **1**·K$^+$ at 400 MHz at −40 °C in $CDCl_3$ displaying three sets of signals. The two sets of small signals related to a second "new" species have been marked with asterisks. The signals labeled "p" and "s" correspond to the picrate and the solvent, respectively.

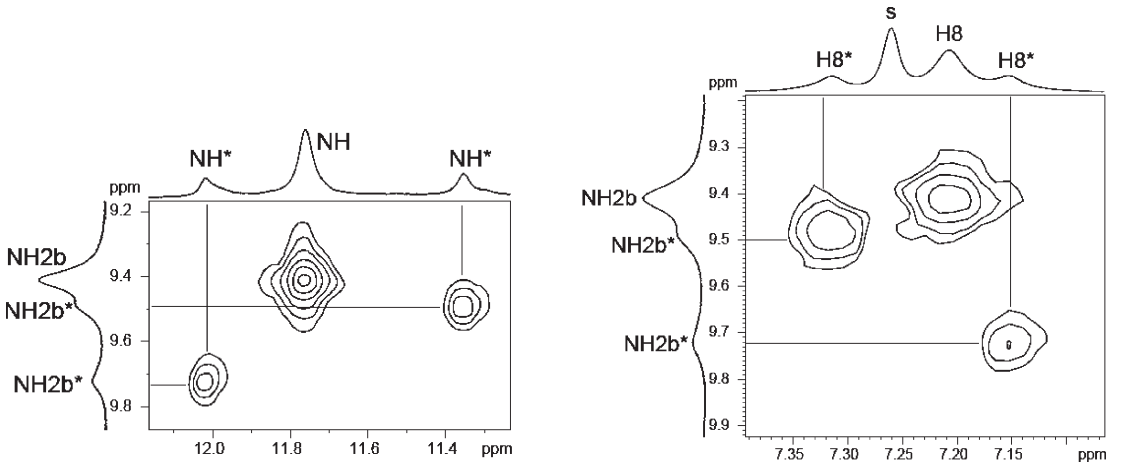

FIGURE 6. Portions of a 100 ms NOESY spectrum of TASQ **1**·K$^+$ acquired at 400 MHz at −40 °C in $CDCl_3$.

driven into *syn* conformations which provide a suitable geometry for Hoogsteen pairing and force the structure into a G-quartet (Figure 4b,c).[30]

1D NMR spectroscopy experiments also showed that the H_{in} proton of the cavitand is sensitive to the presence of cations. This sensitivity can be used as a probe for cation-binding and suggests that the coordinated cations are located near or inside the cavitand template.

The two other cations, K$^+$ and Cs$^+$, yield very different results from Na$^+$ and Sr^{2+}. TASQ **1**·K$^+$ gave a broader ^{1}H NMR spectrum at room temperature. At low temperature the spectrum of TASQ **1**·K$^+$ splits into three sets of signals indicating a dynamic mixture of structures (Figure 5).

It can be seen from Figure 6 that all the signals for TASQ **1**·K$^+$ show the characteristic NOE correlations of G-quartet assemblies (NOEs between H8/NH_{2b} and NH/NH_2). The main set of signals shows a strong H8/H1′ correlation and appears to be originating from an *all-syn*[38,39] G-quartet similar to those of Na$^+$ and Sr^{2+} (data not shown). The side signals in Figure 5 have equal intensities, and seem to be related to a second "new" species present in the solution. From signal integration, it can be approximated that this second species comprises about one-third of the population at −40 °C. In TASQ **1**·Cs$^+$, the corresponding "new" species is dominant, as the spectrum manifests only the two "new" signals. 2D NOESY experiments confirm that TASQ **1**·Cs$^+$ retains the G-quartet structure (see the Supporting Information for the observed NOEs between H8/NH_{2b} and NH/NH_2).

Although conventional NMR methods have provided valuable insight into the TASQ·cation structures, and confirm the presence of G-quartet motifs, they were not sufficient for the determination of stoichiometries. The ratio of cation per quartet was found to be 1:1 for all three assemblies of Na$^+$, Sr^{2+}, and Cs$^+$ based on the integration of the picrate proton resonance at rt (see the Supporting Information). However, the symmetry of the TASQs is clearly broken in cesium and potassium. Only one "new" set of signals is observed in the presence of the larger Cs$^+$. This reveals the presence of two different modes of binding for TASQs that depend on the size of the cations. Potassium is apparently in the intermediate size range, resulting in a mixture in which two structures have comparable stability. Conclusive evidence for the identification of these structures came from a combination of signal integration and diffusion NMR analysis.

Diffusion NMR Spectroscopy. Pulse field-gradient NMR spectroscopy has gained increasing importance in the field of supramolecular chemistry in recent years.[40,41] This technique

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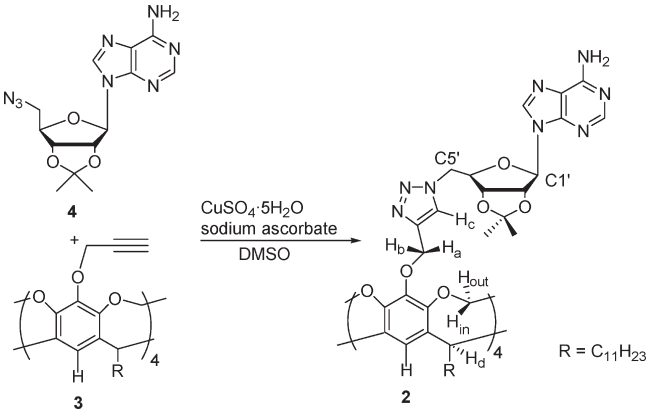
TABLE 1. Diffusion Coefficients (D) of 2, 1, and 1·Cation Assemblies at 295 K

solvent	D_2 [$\times 10^{-10}$ m^2/s]	D_1 [$\times 10^{-10}$ m^2/s]	$D_{1\cdot Na^+}$ [$\times 10^{-10}$ m^2/s]	$D_{1\cdot Sr^{2+}}$ [$\times 10^{-10}$ m^2/s]	$D_{1\cdot Cs^+}$ [$\times 10^{-10}$ m^2/s]	% ratio
DMSO-d_6	0.75 ± 0.02	0.74 ± 0.02				99
$CDCl_3$	3.55 ± 0.10	3.45 ± 0.10				97
$CDCl_3$	3.60 ± 0.10		3.48 ± 0.10			97
$CDCl_3$	3.56 ± 0.10			3.41 ± 0.10		96
$CDCl_3$		3.11 ± 0.10			2.50 ± 0.10	80

has been successfully applied to the size determination of a variety of self-assembled structures such as resorcinarenes,[42,43] (iso)guanosines,[44,45] and rosettes.[46]

For more accurate measurements, it is often necessary to use an internal standard with a similar structure. We chose free TASQ **1** as an internal standard for TASQ **1**·cations, but ^{1}H NMR experiments revealed that, with the exception of TASQ **1**·Cs^+, the assemblies undergo a rapid exchange with the cation-free TASQ, and give an average spectrum at room temperature. So only the diffusion of TASQ **1**·Cs^+ could be directly measured in the presence of free **1**. In the past, adenine compounds have been used as internal standards for diffusion study of guanine compounds.[29,44] Adenine is believed to show few interactions with itself or with guanine, and has a similar shape and molecular weight. Thus compound **2**, an adenine analogue of **1**, was synthesized by using a similar procedure as described previously for TASQs (Scheme 1),[27] and was utilized as a standard (see the Experimental Section).

SCHEME 1. Synthesis of 2



The following mixtures were prepared and subjected to analysis: **2**-**1**, **2**-**1**·Na^+, **2**-**1**·Sr^{2+}, and **1**-**1**·Cs^+ in $CDCl_3$, and **2**-**1** in DMSO-d_6 (Table 1). All the mixtures were in equimolar ratios and kept in the same NMR tube under identical conditions of concentration and temperature. In all cases, no interactions were observed between the standards and the analyzed samples. The experiments were performed at 295 K with a BPLED gradient pulse sequence, and the diffusion coefficients (D) were calculated by plotting the natural logarithm of the signal intensity versus the b-value for a representative signal. The slope of this line equals $-D$ (Figure 7). The b value is the diffusion weighting factor, which is defined as $b = (2\pi\gamma g\delta)^2(\Delta - \delta/3)$ s/m^2 (in which γ is the gyromagnetic ratio, δ is the gradient pulse duration, and g and Δ are the gradient strength and the time between the start of the first and the second gradient pulses). The results are summarized in Table 1 and Figure 7. (See the Supporting Information for data on **1**·Na^+ and **1**·Sr^{2+} in $CDCl_3$, and **1** in DMSO-d_6.)

From Table 1, it can be clearly seen that **1** remains monomeric in both DMSO-d_6 ($\varepsilon_r = 47$) and $CDCl_3$ ($\varepsilon_r = 4.8$). The observed stoichiometry in $CDCl_3$ is consistent with the previously determined basket-like geometry of TASQs, and rules out the presence of higher order aggregates. Moreover, the experimental ratios of diffusion coefficients are in very good agreement with the theoretical values obtained for spherical molecules (Table 1 and eq 1).[47]

$$\frac{D_1}{D_2} = \sqrt[3]{\frac{M_2}{M_1}} \qquad (1)$$

The data listed in Table 1 also suggest that both TASQ **1**·Na^+ and TASQ **1**·Sr^{2+} form monomeric assemblies (see the Supporting Information). These results agree well with the stoichiometries obtained from picrate peak integration. To the best of our knowledge these are the first isolated cation-bound unimolecular G-quartets (as opposed to G-quadruplexes) reported in solution phase: It is known that the coordination of cations to the carbonyl oxygens of the guanines is the dominant force promoting the stacking of the bases in lipophilic systems.[48] Therefore, lipophilic G-quartets tend to stack such that the self-assembly process is very unlikely to stop at the quartet level in the presence of cations.[49] Indeed, the presence of cation-templated isolated G-quartets has only been reported in the gas phase.[50]

The diffusion NMR experiments show that in contrast to the behavior of small cations, cesium promotes formation of a dimer. The experimental ratio of 80% obtained for $D_{1\cdot Cs^+}/D_1$ is in close agreement with the theoretical value of 79% calculated for a dimeric system from eq 1 (Figure 7b). Considering the asymmetry found in the ^{1}H NMR spectra, in addition to the 1:1 ratio of cation per quartet and symmetry of the system, this dimer has to be an asymmetric

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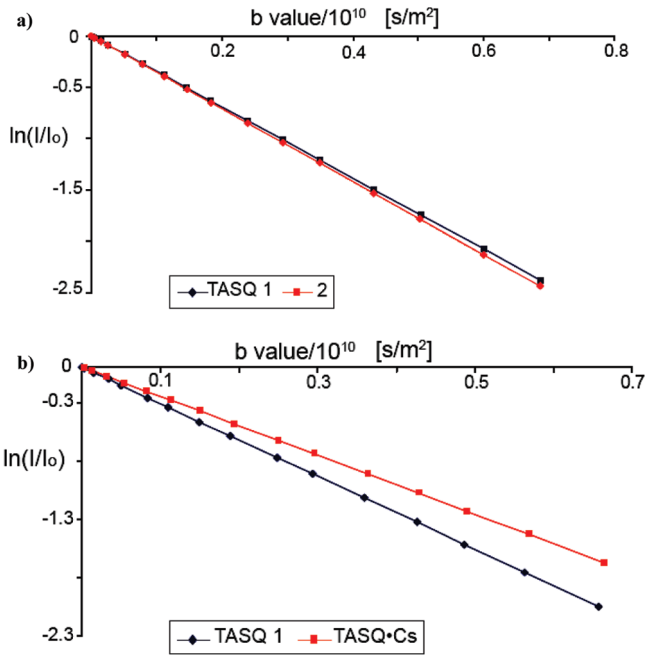

FIGURE 7. Normalized signal decay as a function of the *b* value at 295 K for (a) **1**−**2** in $CDCl_3$ and (b) **1**−**1**·Cs^+ in $CDCl_3$. See the Supporting Information for the diffusion plots of the other assemblies. The representative signals are H1′ for a and NH for b. [*b* value = $(2\pi\gamma g\delta)^2(\Delta - \delta/3)$ s/m^2].

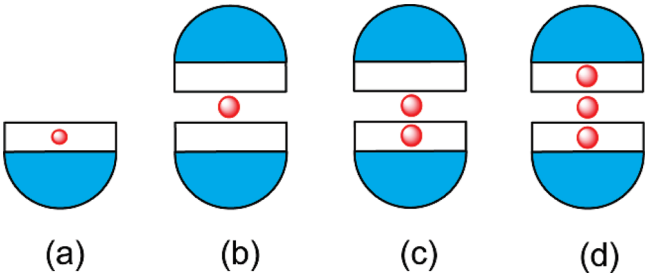

FIGURE 8. Proposed G-quartet-based structures of TASQ·cation assemblies: (a) isolated G-quartet, (b, d) symmetric dimers, and (c) asymmetric dimer. Only structures consistent with structures a and c were observed.

dimer such as structure c in Figure 8. The coordination of a third cation is probably prevented due to repulsive forces between cations. The results obtained for **1**·Cs^+ also shed light on the identity of TASQ **1**·K^+ and suggest that it is most likely a mixture of a monomer and a dimer such as in structures a and c, respectively, of Figure 8, which exchange rapidly at room temperature.

The Anion Effect. Picrate salts have extensive applications in extraction experiments, and their interactions with various ionophores such as crown ethers have been investigated.[51,52] They are only slightly soluble in chloroform ($\varepsilon_r = 4.8$), and exist mostly as ion pair complexes in this media.[53] The picrate anion is a mono- or bidentate ligand that can coordinate to alkali or alkaline earth metal cations through its phenolic and *o*-nitro groups.[52] It can be seen from Figure 3 that the picrate anion interacts with the TASQ·cation assemblies. The upfield shift of the picrate signal in the monomeric assemblies suggests that the picrate anion is associated with the cations and is positioned near or above the aromatic guanines.[54] Similar observations have been reported by Davis et al., who demonstrated that a capping picrate anion can stabilize a sodium-templated octamer and prevent it from converting to a hexadecamer.[44] On the basis of the chemical shifts of the picrate anion one can conclude that the capping effect of the anion in TASQ **1** is controlled by the size of the cation, and is more pronounced for the smaller cations. Larger cations such as cesium (and potassium somewhat) prefer formation of dimeric systems in which ion pairs are completely dissociated. The information obtained from diffusion NMR is also complementary to this finding. The diffusion coefficients of the picrate anion for TASQ·Na^+ ($D_{anion} = 3.39 \pm 0.10$) and TASQ·$Sr^{2+}$ ($D_{anion} = 3.40 \pm 0.10$) are the same as those of their corresponding complexes within experimental error. Considering the molecular weight of the anion, this suggests a strong association between the anion and the complex. The diffusion coefficient of the picrate anion in TASQ·Cs^+ ($D_{anion} = 3.70 \pm 0.10$) is approximately 1.5 times that of the diffusion coefficient of the complex ($D_{complex} = 2.50 \pm 0.10$), which suggests a weaker association compared to those of Na^+ and Sr^{2+}.

A set of extraction experiments in the presence of noncoordinating anions (I^-, BPh_4^-) verified the above interpretation. Under these new conditions, TASQ **1**·Cs^+ gave a spectrum similar to the one recorded in the presence of picrate anion. This suggests that the formation of asymmetric dimer is not affected by the anion. TASQ **1**·$Na^+$and TASQ **1**·Sr^{2+}, in contrast, yielded very broad spectra in the absence of picrate indicating the loss of the monomer as the singular species (see the Supporting Information).

Selectivities of TASQs. Cation selectivity of TASQ **1** was also evaluated in a set of competition experiments. Addition of solid sodium picrate to a solution of TASQ **1**·Cs^+ in chloroform resulted in complete replacement of cesium ions with sodium. This process is also accompanied by a major structural change in which the dimeric system converts to a monomer. In contrast, addition of solid cesium picrate to a solution of TASQ **1**·Na^+ in chloroform resulted in no change. These results indicate that TASQ **1** has a high Na^+/Cs^+ selectivity. In a similar set of experiments performed on the complexes of Sr^{2+} and Na^+, the divalent cation completely replaced the sodium ion. The selectivity of TASQ **1** for potassium was also evaluated to be between that of sodium and cesium.[55] The results are summarized in Table 2.

The high affinity of TASQ **1** for strontium may be of industrial significance. $^{90}Sr^{2+}$ and $^{137}Cs^+$ are both products

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(55) G-quartets bind K^+ selectively over Na^+ in aqueous solutions. This selectivity benefits from the lower dehydration energy of K^+ with respect to Na^+. Under the anhydrous conditions reported here, the selectivity observed reflects the intrinsic selectivity. See ref 1 and references cited therein.

TABLE 2. The Selectivities of TASQs

starting species	added M·picrate	resulting species
TASQ **1**·Na^+	Cs·Pic	TASQ **1**·Na^+
TASQ **1**·Cs^+	Na·Pic	TASQ **1**·Na^+
TASQ **1**·Na^+	Sr·Pic	TASQ **1**·Sr^{2+}
TASQ **1**·Sr^{2+}	Na·Pic	TASQ **1**·Sr^{2+}
TASQ **1**·Na^+	K·Pic	TASQ **1**·Na^+
TASQ **1**·K^+	Na·Pic	TASQ **1**·Na^+
TASQ **1**·Cs^+	K·Pic	TASQ **1**·K^+
TASQ **1**·K^+	Cs·Pic	TASQ **1**·K^+

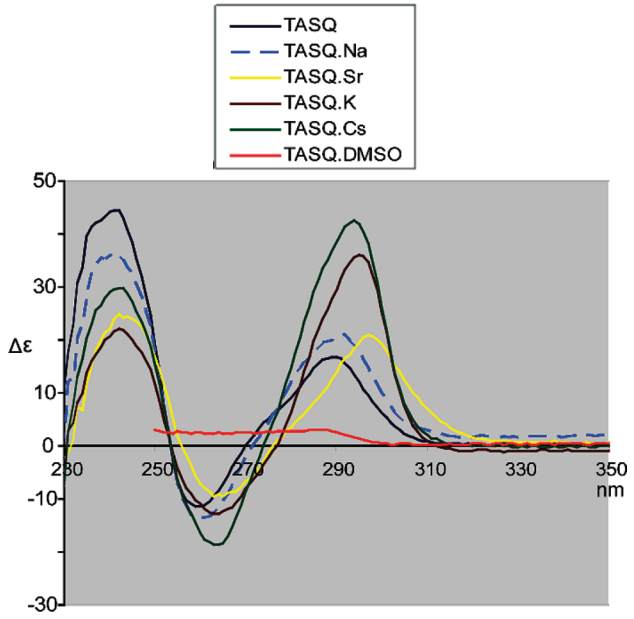

FIGURE 9. CD spectra of a 0.2 mM solution of TASQ **1** in chloroform and DMSO, and TASQ **1**·cation assemblies in chloroform.

of nuclear fission and their selective stripping from salted radioactive waste solutions has been the subject of considerable research in the past.[56,57] It should also be noted that these results display only the selectivities in the presence of picrate anion. As noted above, the anion changes the structure of the assemblies and may modulate the overall selectivities. A series of control experiments in which **2** was mixed with either sodium or cesium picrates showed no changes either in the color or in the ^{1}H NMR spectra. Thus, guanines are essential for the cation-affinity of TASQs. The overall selectivity displayed by TASQ **1** is $Sr^{2+} \gg Na^+ \gg K^+ \gg Cs^+$. The magnitude of these selectivities is estimated as more than a factor of 20 between each pair of cations.

CD Spectroscopy. Furanose sugars are chiral and induce a small CD band in the electronic transition of a single nucleotide.[58] When guanines are arranged in a G-quadruplex this CD band is intense and gives valuable information about the structures in solution.[59] The CD spectra of TASQ·cation assemblies exhibit a negative band at ~260 nm and positive bands at ~240 and ~290 nm for all of the complexes in chloroform (Figure 9). However, these CD bands cannot be attributed only to the H-bonding of guanines. The cavitand and triazole are two other chromophores present that absorb in the same region and contribute to the observed CD bands.[60] This poses an obstacle for the detailed interpretation of the CD bands. Nevertheless, the following features can be pointed out: (a) the general shape and polarity of the spectra are independent of the type and number of the cations bound and (b) there is some variation in the wavelength and intensities of the CD bands, and (c) the CD bands that TASQ **1** manifests in chloroform are absent in DMSO, which is consistent with the ^{1}H NMR data that suggest the loss of the G-quartet structure in the polar DMSO solvent (Figure 9).

Conclusions

We have developed the concept of template-assembled synthetic G-quartets (TASQs) as an approach for controlling the morphology of G-quadruplex assemblies.[27] In the present study, we demonstrated the affinity of TASQs for cations, and explored the structural consequences of cation-complexation. Four cations (Na^+, Sr^{2+}, K^+, and Cs^+) were selected and NMR spectroscopic methods were utilized to investigate the resulting structures. ^{1}H NMR and ^{1}H−^{1}H NOESY analysis were performed on each assembly and confirmed the presence of G-quartet units. Diffusion NMR experiments were undertaken and the sizes of the complexes were determined. The combined NMR analyses suggest the presence of an isolated unimolecular G-quartet in the absence of cations and in the presence of Na^+ and Sr^{2+}. In the presence of Cs^+, we found an asymmetric dimer in solution. In the presence of K^+, both an asymmetric dimer and a unimolecular G-quartet were found. A deeper analysis revealed the role of the anions in formation and stabilization of these structures. Interconversion of structures was investigated. Switching from Cs^+ to Na^+, for example, induced a conformational change from a dimer to a monomer. Selectivities were evaluated and Sr^{2+} was found to be coordinated more strongly than the other cations.

Currently we are working on larger, water-soluble TASQs. We hope that such compounds will find application, for example, as screens for drugs that recognize and stabilize natural G-quadruplexes; such drugs have potential as anticancer agents.[1]

Experimental Section

Compound 2. To a solution of cavitand **3**[61] (111 mg, 0.081 mmol) and 5′-azido-2′,3′-*O*-isopropylidene adenosine **4**[62] (120 mg, 0.360 mmol) in 10 mL of degassed DMSO was added $CuSO_4$·$5H_2O$ (0.202 mL, 0.040 M aqueous solution, 0.008 mmol) and sodium ascorbate (0.202 mL of freshly prepared 0.400 M aqueous

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solution, 0.081 mmol). The reaction mixture was stirred at 60 °C overnight under a positive pressure of N_2. The solvent was removed in vacuo, and the crude products were washed thoroughly with hot deionized water. A few drops of ammonium hydroxide were used to remove the copper catalyst. The crude reaction mixture was subjected to column chromatography on silica gel with use of a 1:3 mixture of EtOH/EtOAc. The solvent was evaporated and the obtained precipitate dissolved in DMSO. Precipitation with deionized water, filtration, and subsequent washing of the precipitate with hot deionized water yielded the pure product (Scheme 1).

2: yield 68%; ^{1}H NMR (400 MHz, DMSO-d_6, 298 K) δ 8.26 (s, 4 H, H8), 8.18 (s, 4 H, H2), 8.06 (s, 4 H, H_c), 7.33 (r bs, 8H, NH_2), 7.26 (s, 4 H, ArH), 6.22 (d, 4H, H1′), 5.88 (d, 4 H, H_{out}), 5.46 (dd, 4 H, H2′), 5.14 (dd, 4 H, H3′), 4.90 (m, 8 H, H_a, H_b), 4.76 (dd, 4 H, H5′b), 4.65 (dd, 4 H, H5′a), 4.56 (t, 4 H, H_d), 4.55 (m, 4 H, H4′), 4.25 (d, 4 H, H_{in}), 2.30 (m, 8 H, CH_2 cavitand feet), 1.49 (s, 12 H, CH_3 sugar), 1.38 (m, 8 H, CH_2 feet), 1.29 (s, 12 H, CH_3 sugar), 1.20−1.28 (m, 64 H, CH_2 feet), 0.84 (t, 12 H, CH_3 feet); ^{13}C NMR (400 MHz, $CDCl_3$, 298 K) δ 156.2, 153.2, 148.9, 148.2, 148.1, 145.1, 144.0, 140.0, 139.0, 124.3, 120.0, 114.9, 114.7, 99.6, 90.4, 84.9, 83.7, 81.7, 67.4, 51.3, 37.0, 32.0, 30.0, 29.8 (multiple peaks), 29.5, 28.1, 27.2, 25.5, 22.8, 14.2; MALDI-TOF-MS calcd for $C_{140}H_{184}N_{32}O_{24}\cdot H^+$ 2699.42, found [M + H^+] 2700.1.

1·Na$^+$: ^{1}H NMR (400 MHz, $CDCl_3$, 298 K) δ 11.55 (s, 4 H, NH), 8.69 (s, 2 H, picrate), 7.79 (s, 4 H, H_c), 7.78 (s, 4 H, H8), 6.65 (s, 4 H, ArH), 5.88 (d, 4H, H1′), 5.67 (d, 4 H, H_b), 5.19 (dd, 4 H, H2′), 5.18 (dd, 4 H, H3′), 4.96 (dd, 4 H, H5′b), 4.76 (d, 4 H, H_a), 4.61 (dd, 4 H, H5′a), 4.53 (t, 4 H, H_d), 4.50 (d, 4 H, H_{out}), 4.31 (m, 4 H, H4′), 3.93 (d, 4 H, H_{in}), 2.30 (m, 8 H, CH_2 cavitand feet), 1.48 (s, 12 H, CH_3 sugar), 1.39 (m, 8 H, CH_2 feet), 1.29 (s, 12 H, CH_3 sugar), 1.23−1.30 (m, 64 H, CH_2 feet), 0.84 (t, 12 H, CH_3 feet).

1·Sr^{2+}: ^{1}H NMR (400 MHz, $CDCl_3$, 298 K) δ 11.15 (s, 4 H, NH), 8.70 (s, 2 H, picrate), 7.87 (s, 4 H, H_c), 7.80 (s, 4 H, H8), 6.62 (s, 4 H, ArH), 5.86 (d, 4H, H1′), 5.54 (d, 4 H, H_b), 5.24 (dd, 4 H, H3′), 5.00 (dd, 4 H, H2′), 4.96 (dd, 4 H, H5′b), 4.78 (d, 4 H, H_a), 4.63 (dd, 4 H, H5′a), 4.54 (t, 4 H, H_d), 4.46 (d, 4 H, H_{out}), 4.30 (m, 4 H, H4′), 3.95 (d, 4 H, H_{in}), 2.30 (m, 8 H, CH_2 cavitand feet), 1.48 (s, 12 H, CH_3 sugar), 1.39 (m, 8 H, CH_2 feet), 1.29 (s, 12 H, CH_3 sugar), 1.23−1.30 (m, 64 H, CH_2 feet), 0.84 (t, 12 H, CH_3 feet).

1·Cs$^+$: (partially assigned, region 6−12), ^{1}H NMR (400 MHz, $CDCl_3$, 298 K) δ 11.41 (s, 4 H, NH), 10.83 (s, 4 H, NH*), 8.87 (s, 4 H, picrate), 7.70 (s, 4 H, H_c), 7.42 (s, 4 H, H_c*), 7.44 (s, 4 H, H8), 7.17 (s, 4 H, H8*), 6.71 (s, 4 H, ArH), 6.51 (s, 4 H, ArH*).

Picrate Extraction Experiments. Picrate salts were synthesized following the literature method.[63] In a typical solid−liquid extraction experiment 0.01 mmol of **1** was mixed with 0.04 mmol of sodium picrate then the solution was stirred in chloroform for 6 days. The resulting mixture was centrifuged and the solution was filtered through a piece of glass wool (packed in a Pasteur pipet). The product was isolated as a yellow solid after evaporation of chloroform at room temperature. Chloroform/aqueous picrate extraction was not practical, as precipitation resulted.

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Supporting Information Available: General procedure and additional NMR spectroscopic data for TASQ·cation assemblies (Figures S1−S5). This material is available free of charge via the Internet at http://pubs.acs.org.

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