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Communication

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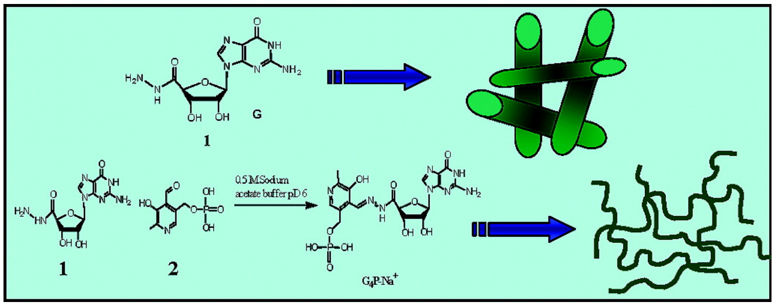

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Modulation of the Supramolecular Structure of G-Quartet Assemblies by Dynamic Covalent Decoration

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Hydrogel materials have received increasing attention because of their potential impact on the development of bioengineering and biomedical applications.[1−3] There have been numerous developments on cross-linked polymers and hydrogels due to their potential applicability in drug delivery.[3] Our efforts in this area have been driven by our recent results on guanosine-5′-hydrazide **1** that forms stable hydrogels in presence of metal cations.[4] We have investigated the sol−gel phase transition as a function of various physical and chemical parameters.[4]

The G-quartet is formed by four Hoogsten type hydrogen bonds of four G nucleobases in the presence of metal ions.[5−7] This supramolecular macrocyclic structure has four carbonyl oxygens pointing toward the central cavity (Figure 1). The cavity is well-organized for the binding of cations, particularly K^+, which increases the stability of the columnar aggregates.

In our previous study,[4] it was shown that the pH-sensitive guanosine hydrazide **1** is a powerful hydrogelator capable of undergoing dynamic decoration through the formation of reversible acylhydrazone bonds with various aldehydes. The macroscopic properties of the gels formed, such as the sol−gel transition temperature (T_{gel}) or the transparency, were found to strongly depend on the nature of the cation and/or of the decorative entities. Thus, the cation-templated self-assembly of quartets of guanosine hydrazide **1** decorated with pyridoxal monophosphate **2** gave stable gels, whereas other aldehydes gave either low viscosity solutions or precipitates depending on the nature of the aldehyde.[4]

In order to gain insight into the structure of the supramolecular hydrogels formed, we performed a comparative study by small-angle neutron scattering (SANS) of the gels formed from both guanine hydrazide **1** and its acylhydrazone derivative formed by condensation with pyridoxal-5-phosphate **2** (Figure 1). Three dispersions in deuterated water at a concentration of 15 mM were investigated. The synthesis of the guanosine hydrazide **1** and of the acylhydrazone and their gelation properties were described elsewhere.[4] SANS experiments were performed on the spectrometer PACE at Saclay, France. In the available range of wave-vector q, the scattered intensity is proportional to the form factor of the strand of the network.[8] Figures 2 and 3 represent the variations of the scattered intensity as a function of q for the [G_4-Na^+] and [G_4-K^+] systems at $T = 20$ and 55 °C.

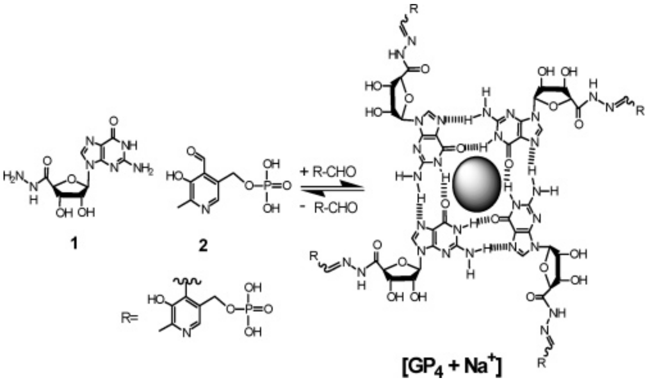

Figure 1. Reversible decoration of G-hydrazide **1** quartet with pyridoxal monophosphate **2**.

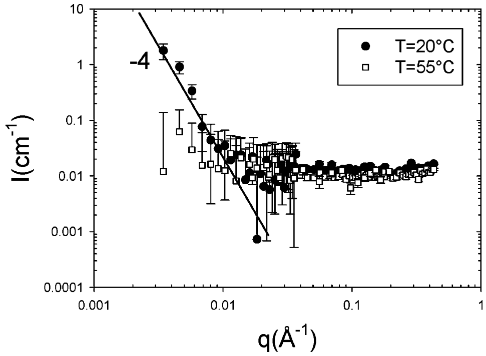

Figure 2. Scattering curves for the [G_4-Na^+] system at $T = 20$ °C ($T_{gel} - T = 20$ °C) and 55 °C ($T_{gel} - T = -15$ °C).

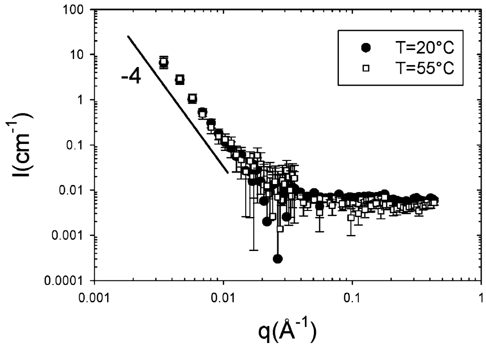

Figure 3. Scattering curves for the [G_4-K^+] system at $T = 20$ °C ($T_{gel} - T = 41$ °C) and 55 °C ($T_{gel} - T = 6$ °C).

The scattering curve obtained for the [G_4-Na^+] sample at $T = 20$ °C, that is below T_{gel}, exhibits an initial decay that can be described by a q^{-4} variation followed by a constant background. The latter can be ascribed to the incoherent scattering arising in particular from the presence of hydrogen atoms in the deuterated water resulting from an interchange with the particles, which depresses significantly the contrast between the particles and the solvent. The q^{-4} decrease is characteristic of particles with sharp interfaces and is observed for scattering wave-vectors larger than the characteristic particle size. Assuming that the gel is formed from cylindrical compact fibers, as suggested by the electron microscopy

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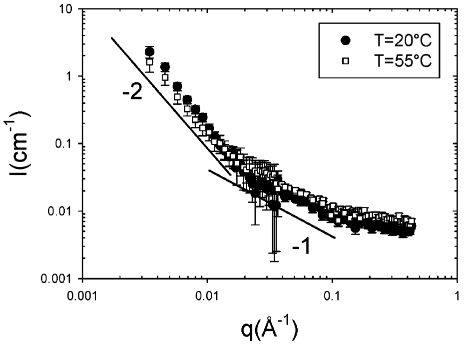

Figure 4. Variation of the scattered intensity, I, versus q for the [GP$_4$-Na$^+$] decorated system at $T = 20$ and 55 °C.

results,[4] the scattering intensity in the range $q > 1/d$, where d represents the average diameter of the cylinder, is given by the classical Porod law: $q^4I(q)/\phi(\Delta\rho)^2 = 8\pi/d$, where ϕ is the volume fraction and $(\Delta\rho)^2$ the contrast per unit volume between the polymer and the solvent.[8] The analysis of the scattering curves at $T = 20$ °C yields a value for the average diameter $d = 778$ nm, which explains the turbidity of the gel. As the temperature is increased above the sol−gel transition, the low q variation disappears and the gel becomes transparent, suggesting the breaking of the supramolecular structure.

The [G$_4$-K$^+$] sample shows the same behavior (Figure 3) as that observed for the [G$_4$-Na$^+$] system at 20 °C, but the signal remains unchanged up to 55 °C. One notes that the structure of the fibers remains remarkably stable in the temperature range investigated. The analysis of the data for this sample leads to $d = 194$ nm. Figure 4 shows the variations of I versus q obtained for the pyridoxal phosphate **2** decorated gel [GP$_4$-Na$^+$] at temperatures of 20 and 55 °C. It can be observed that the two scattering curves are superimposed, showing no effect of temperature. The shape of the scattering curve differs significantly from that observed for the nondecorated gels. One can distinguish three domains in the curves of Figure 4. At low q, $I(q)$ follows a q^{-2} decrease, followed by a q^{-1} domain, then by a background associated with the incoherent scattering. The q^{-1} behavior is the signature of the presence of locally unidirectional aggregates. In the high q limit, a contribution from the form factor of the cross section should be observed. Such a component might be buried in the incoherent background, but in any case, it would be in a range of $q > 0.1$ Å^{-1}, which points to a fiber diameter smaller than ∼20 Å. This conclusion is corroborated by the q^{-2} behavior of I in the low q regime that suggests that the fibers are semi-flexible. From the crossover position, q^*, between the q^{-2} and q^{-1} behaviors, one can deduce the persistence length of the column: $lp = 1.91/q^*$.[8] One obtains $lp = 130$ Å. Further information on the molecular level assembly process is provided by the analysis of the intermediate q^{-1} rod scattering range described by[8] $I(q) \sim (\Delta\rho)^2 \times M_L \times \pi/q$. For the system investigated here, the contrast associated with the decorated aldehyde is 100 times larger than that due to the quartets. In a first approximation, one can then assume that the scattering arises from the decorating units. Within this assumption, the analysis of the data of Figure 4, using the above relationship, leads to an average value of the mass per unit length M_L of the decorating units equal to 182 g/mol/Å. This corresponds to an average distance between quartets of ∼5 Å. The above value is significantly larger than the admitted stacking value of 3.4 Å. This suggests a structure looser than that of a compact stacking. In this respect, a model has been proposed for the K$^+$ macromolecular structure of guanosine-5′-monophosphate in solution consisting of a pseudo-four-stranded helix with guanine−guanine hydrogen bonding forming a continuous helical strand rather than the usual planar G-tetrad structure.[9] This model would account for the experimental value of the distance between quartets.

The results reported above show the strong effect of the decoration of the core entity **1** on the supramolecular structure of the gels. The guanosine hydrazide quartets self-assemble into thick compact fibers able to form cross-links between themselves. The lateral aggregation of columns could result from the formation of hydrogen bonds between the NH$_2$ group and hydroxyl groups. Note that, at acidic pH, the gels were disrupted, likely because of a protonation of NH$_2$. Furthermore, the structure of the gel fibers is sensitive to the nature of the cation. The thickness of these fibers is much larger for Na$^+$ than for K$^+$ quartets. This may be at the origin of the different sol−gel transition temperatures, the Na$^+$ hydrogels being less stable than the K$^+$ ones. The different structures of [G$_4$-Na$^+$] and [G$_4$-K$^+$] gels likely result from the different sizes of the cations. The larger radius of K$^+$ fits best the stacking distance between quartets leading to a better organization of the fibers and to the formation of sandwich structures of K$^+$ between two G$_4$ units. The hydrogel obtained by reacting pyridoxal monophosphate **2** with hydrazide **1** exhibits a quite different structure, consisting likely of single columns of loosely stacked quartets. At pD = 6, the phosphate groups are negatively charged. The resulting repulsive electrostatic interactions and also possible steric interactions prevent further lateral aggregation between the columns. As a result of this single columns self-assembling, one expects the gel to be a viscoelastic solution of very long entangled chains rather than a cross-linked network. This would explain why this system recovers in a reasonable lapse of time its gel-like aspect after shear thinning, contrary to the guanosine hydrazide gels that remain fluid solutions after being disrupted by shear.

In conclusion, the hydrogels formed from the G-quartets of **1** are strongly dependent on the nature of both the binding cation and the decorating aldehyde. The latter is of particular interest as it points to the possibility to modulate the supramolecular structure of the assemblies by dynamic covalent decoration. It would be of interest to try to correlate the gel structure with the previously reported amplification of a given constituent of a constitutional dynamic library under the pressure of the self-organization of the gel supramolecular structure.

References

(1) Drury, J. L.; Mooney, D. J. *Biomaterials* **2003**, *24*, 4337.
(2) Lee, K. Y.; Mooney, D. J. *Chem. Rev.* **2001**, *101*, 1869.
(3) (a) Hoffman, A. S. *Adv. Drug Delivery Rev.* **2002**, *54*, 3. (b) Qiu, Y.; Park, K. *Adv. Drug Delivery Rev.* **2001**, *53*, 321. (c) Kwon, I. C.; Bae, Y. H.; Kim, S. W. *Nature* **1991**, *354*, 291. (d) LaVan, D. A.; McGuire, T.; Langer, R. *Nat. Biotechnol.* **2003**, *21*, 1184. (e) Gupta, P.; Veramani, K.; Garg, S. *Drug Discovery Today* **2002**, *7*, 569. (f) Friggeri, A.; Feringa, B. L.; Esch, J. V. *J. Controlled Release* **2004**, *97*, 241.
(4) Sreenivasachary, N.; Lehn, J. M. *Proc. Natl. Acad. Sci. U.S.A.* **2005**, *102*, 5938.
(5) Guschlbauer, W.; Chantot, J. F.; Thiele, D. *J. Biomol. Struct. Dyn.* **1990**, *8*, 491.
(6) Gottarelli, G.; Masiero, S.; Spada, G. P. *J. Chem. Soc., Chem. Commun.* **1995**, 2555.
(7) Gottarelli, G.; Spada, G. P.; Garbesi, A. In *Comprehensive Supramolecular Chemistry*; Sauvage, J. P., Hosseini, M. W., Eds.; Pergamon: Elmsford, NJ, 1996; Vol. 9; pp 483−506.
(8) Buhler, E.; Boué, F. *Macromolecules* **2004**, *37*, 1600.
(9) Walmsley, J. A.; Burnett, J. F. *Biochemistry* **1999**, *38*, 14063.

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