Self-Assembly in Organic Solvents of a Deoxyguanosine Derivative Induced by Alkali Metal Picrates

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3′,5′-Didecanoyl-2′-deoxyguanosine in chlorinated organic solvents undergoes self-assembly mediated by alkali metal picrates which leads to octameric or polymeric species according to the amount of ions present.

A few classes of synthetic or natural macrocycles such as crown-ethers, calixarenes, cryptands and some cyclic peptides are able to transfer cations from the aqueous to organic phases. These molecules possess preorganized structures which selectively complex the appropriate cation.[1] Here we report a case of ion transport mediated by the assembly of eight (or more) molecular units.

It is known that guanosine derivatives, in water, form columnar aggregates characterized by the presence of tetramers (G-quartets).[2]

This self-assembly process is favoured by selected metal ions and eventually leads to the formation of well-characterized columnar liquid crystalline phases.[3] We have synthesized† 3′,5′-didecanoyl-2′-deoxyguanosine **1** which is soluble in chlorinated organic solvents.

This compound is able to transfer alkaline picrates (M^+Pic^-) from the aqueous to the organic phase. The CD spectra before and after potassium picrate extraction‡ are shown in Fig. 2.

In the presence of K^+ the exciton shape of the spectrum is quasi-specular to that of the four-stranded helix formed by polyguanylic acid and typical of an assembled species composed of at least two G-quartets.[3,4]

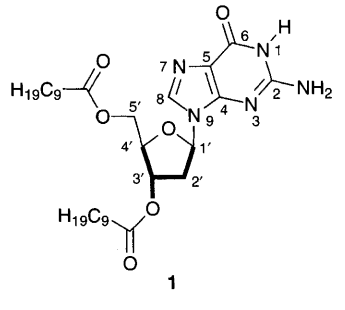
The ^1H NMR spectra between δ 13 and 6 (a region useful for characterizing the association of guanosine derivatives giving G-quartets[5,6]), obtained after extraction of different amounts of potassium picrate,§ are shown in Fig. 3 (the signals marked with * are before picrate extraction).

The imino proton signal was a singlet at δ 12.23 and, when potassium picrate was extracted, slowly transformed into a doublet at a slightly higher field (δ 12.05). The picrate signal was at δ 8.89 and allowed the determination of the quantity of K^+ present in the organic phase. The signal of H-8 was at δ 7.69 (*a*) and in the presence of K^+, two new signals of equal intensity appeared at 8.00 (b_1) and 7.41 ppm (b_2). These signals became more and more intense when more K^+ was extracted, while the original central signal decreased. When the amount of K^+ present in the organic phase was further increased and the ratio between the molar concentration of **1** and potassium ion ([**1**]/[K^+]) was smaller than 12, the two lateral bands began to decrease and a new complex band appeared at δ 7.5–7.8 (*c*). At [**1**]/[K^+] ≤ 4 the signals became broad, indicating the presence of high molecular weight species. The percentage of **1** corresponding to the three different types of signals in the H-8 region (*a*, *b* and *c*) is shown in Fig. 4(*a*).

The stoichiometry of the complex can be deduced from the picrate and the H-8 signal intensities. In the high [**1**]/[K^+] range (40–16) where the only complex is that characterized by the *b* signals, there were 8 guanosines per K^+ [see Fig. 4(*b*)]. At lower [**1**]/[K^+] ratios (≤12) the overall stoichiometry was reduced approaching an estimated value of 4 guanosines per K^+.

The two H-8 signals (b_1 and b_2), in the concentration range in which the 8 : 1 species is present, indicated two assembled G-quartets with a central C_4 symmetry axis and no C_2 axes perpendicular to it, *i.e.* a head-to-tail arrangement.[5] The K^+ ion should occupy the central position, connecting the two tetrameric units *via* coordination with the eight oxygens present in the cavity. A similar situation has been reported for the self-assembly of deoxyguanosine-5′-phosphate in water.[5]

The spectrum transformation and the stoichiometry variation beginning when the [**1**]/[K^+] ratio was *ca.* 8 can be interpreted in terms of the addition of more G-quartets to the octameric structure to give a longer columnar aggregate. If the columnar length increases, the intensity of the H-8 corresponding to the two external quartets becomes relatively weaker and new signals corresponding to the internal H-8 should appear; these signals are likely to have chemical shift values not widely



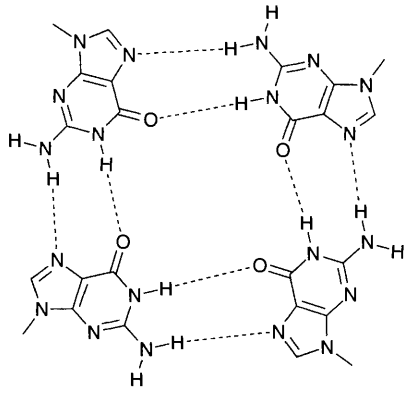


Fig. 1 The G-quartet: four guanine residues connected by the Hoogsteen pattern of hydrogen bonds

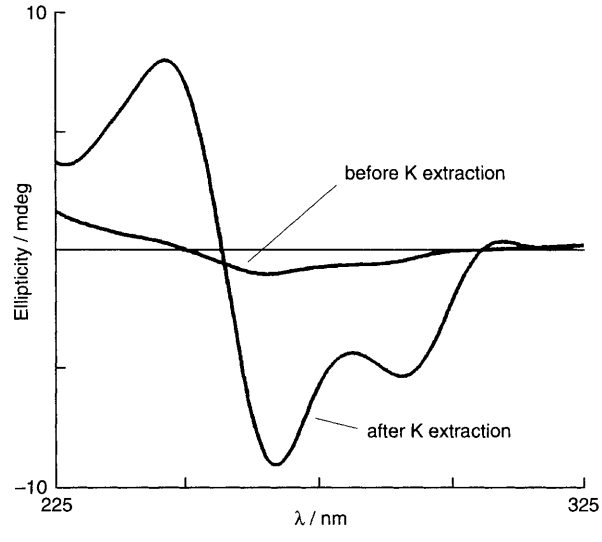


Fig. 2 The CD spectra of **1** in CH_2Cl_2 (2.5×10^{-3} mol dm^{-3}) (l = 0.01 cm, room temp.) before and after the potassium extraction‡

separated as the chemical environment is rather similar. At the same time, during polymerisation, the amount of K^+ per guanine should approach the value 4. When the aggregate further grows, the NMR signals became broad as expected when T_2 becames smaller. Recently, the high-resolution crystal structure of the tetraplex formed by $d(TG_4T)$ was reported and one sodium ion was found between each couple of G-quartets.[7] The trend shown in Fig. 4(*a*) agrees well with this interpretation: the 'free' species concentration decreased on adding picrate, while the 'octamer' increased. When more picrate was added, the 'octamer' concentration decreased and a 'polymeric' form began to appear.

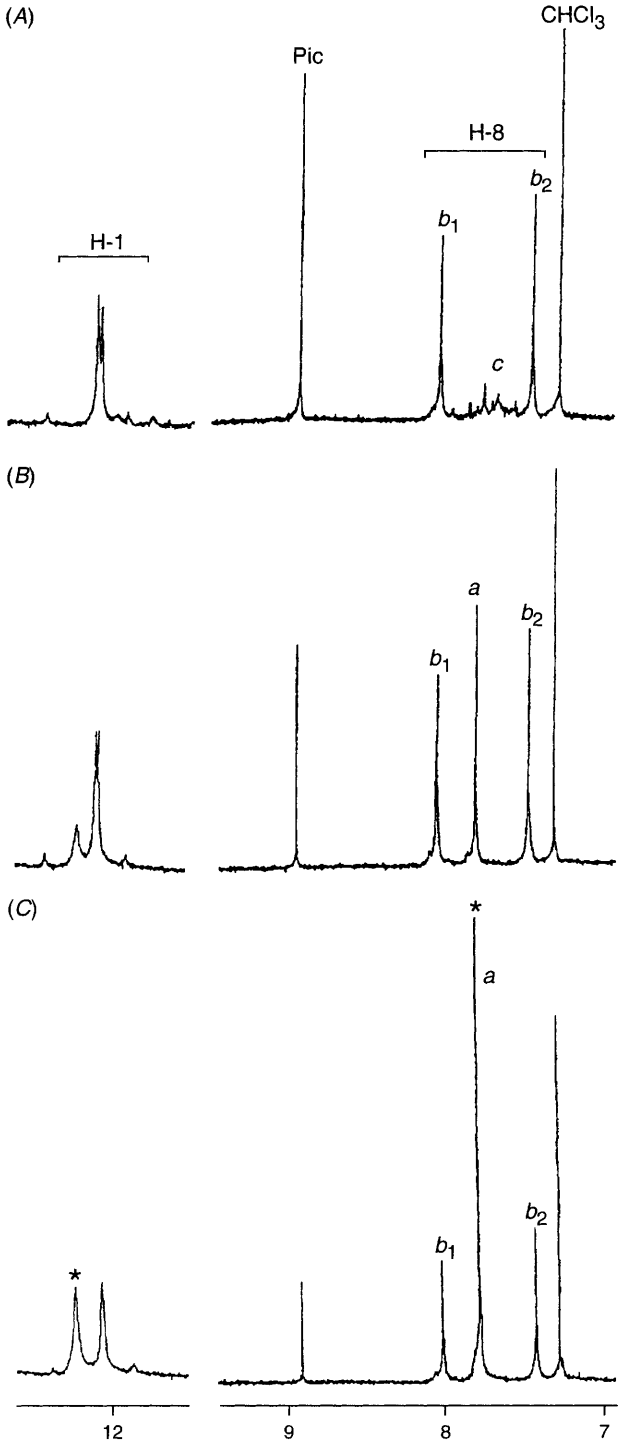


Fig. 3 The ^1H NMR spectra of **1** after extraction of different amount of K^+Pic^-.§ The ratio [**1**]/[K^+] is 40 (*C*), 20 (*B*), and 8 (*A*). Starred signals refer to the situation before extraction.

The results are also surprising in relation to the stacking interactions mechanisms between the bases:[8] stacking forces are usually considered important only in water; if so in the present case, the cation coordination must play a dominant role.

There is extraction selectivity; K^+ is extracted better than Na^+ and Cs^+: the extraction percentage of alkali picrate by **1**, from water to $CHCl_3$ at 20 °C, are 67 ± 4, 56 ± 4 and 59 ± 4 for K^+, Na^+ and Cs^+, respectively.¶

A final remark: from NMR spectroscopy, derivative **1** in $CHCl_3$ without added cations is likely to be dimeric[10] with H-bonds between the two CONH moieties. The imino proton is in fact at *ca.* δ 12 even before cation addition, while the NH_2 group resonates at δ 6.4 and should not be H-bonded;[11] when K^+ was extracted, this signal slowly became weaker until it eventually disappeared.

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Note added in proof: after this paper was submitted, the self-assembly of a lipophylic isoguanosine derivative with similar cation dependence was reported (J. T. Davis, S. Tirumala, J. R.

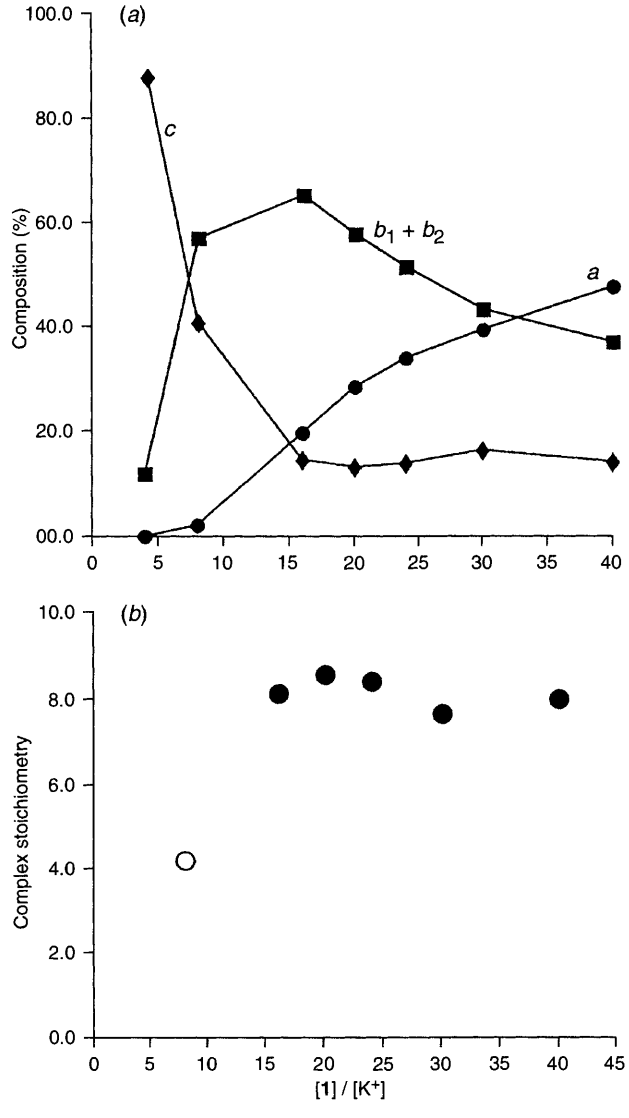

Fig. 4 (*a*) The percentage of 'G' showing the different signals in the H-8 region as a function of the [**1**]/[K^+] ratio. (*b*) The stoichiometry of the complex characterized by signals b_1 and b_2 in the H-8 region. At [**1**]/[K^+] < 12 the value is no more constant, denoting that different species became important.

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Footnotes

† The amino group of 2′-deoxyguanosine was protected with FMOC and acylation was carried out with decanoyl chloride in pyridine; deprotection performed with piperidine in CH_2Cl_2 at room temp., without significant cleavage of the two ester functions. The final compound has the following characteristics: ^{1}H NMR (300 MHz) δ($CDCl_3$) 0.86 and 0.88 (6 H, tt, 2 Me), 1.10–1.40 (24 H, m, 12 CH_2), 1.52–1.73 (4 H, m, 2 CH_2CH_2CO), 2.35 and 2.36 (4 H, tt, 2 CH_2CO), 2.42–2.93 (2 H, mm, H-2′/H-2″), 4.29–4.48 (3 H, m, H-5′/H-5″/H-4′), 5.39 (1H, m, H3′) 6.23 (1 H, m, H-1′) 6.38 (2 H, bs, NH_2), 7.69 (1 H, s, H-8) and 12.23 (1 H, bs, NH); ^{13}C NMR (75.54 MHz) δ($CDCl_3$) 14.587 (CH_3), 23.139 (CH_2), 25.291 (CH_2), 29.593 (CH_2), 29.622 (CH_2), 29.749 (CH_2), 29.767 (CH_2), 29.804 (CH_2), 29.887 (CH_2), 32.331 (CH_2), 34.543 (CH_2), 34.608 (CH_2), 37.719 (CH_2), 64.196 (CH_2), 74.933 (CH), 83.030 (CH), 84.781 (CH), 117.884 (C), 136.140 (CH), 151.751 (C), 154.341 (C), 159.493 (C), 173.650 (C) and 174.033 (C).

‡ Picrate extraction experiments were performed according to the following procedure: aqueous picrate (1 ml; 2.5×10^{-3} mol dm^{-3}) was added to **1** in CH_2Cl_2 or $CHCl_3$ (1 ml; 2.5×10^{-3} mol dm^{-3}): the resulting biphasic system was stirred for 10 min, vortexed for 8 min overall and centrifuged for 10 min.

§ Different amounts of solid K^+Pic^- were added to **1** in $CDCl_3$ (5.2×10^{-2} mol dm^{-3}). These heterogeneous systems were sonicated for 15 min and then allowed to stand at 4 °C for at least 5 d before recording the NMR spectra.

¶ Picrate extraction experiments were performed according to the following procedure: aqueous picrate (1 ml; 1.1×10^{-3} mol dm^{-3}): was added to **1** in $CHCl_3$ (1 ml; 1.75×10^{-2} mol dm^{-3}): the resulting biphasic system was stirred for 10 min, vortexed for 8 min and then centrifuged for 10 min. The concentration of the picrate ion was then determined spectrophotometrically both in the aqueous and organic phases (see ref. 9). Blank experiments showed that no significant picrate extraction occurred in the absence of **1**.

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