Figure 1 shows a typical spectrum of the cluster distribution generated in the source and introduced into the analyzer cell of the FTMS-2000 spectrometer following thermalization of the ions for 1 s in a background pressure of Ar at 2×10^{-6} Torr. The enhanced intensities of C_{60}^+ and C_{70}^+ are readily apparent in this spectrum. The C_n^+ ions were then permitted to react for 1 s with $Fe(CO)_5$, which was introduced into the cell using a pulsed solenoid valve.[19] The data shown in Figure 2 indicate a remarkable selectivity in which C_{60}^+ and C_{70}^+ appear to be by far the most reactive species under these experimental conditions. This conclusion was more clearly confirmed by using double-resonance ejection to isolate narrow regions of C_n^+ and C_{60}^+ in particular.[20] This was also particularly helpful in distinguishing $FeC_n(CO)_4^+$ from C_{n+14}^+.

Reactions 1 and 2 are observed in nearly equal amounts upon reacting isolated C_{60}^+ with $Fe(CO)_5$. However, reaction 2

$$C_{60}^+ + Fe(CO)_5 \rightarrow Fe(CO)_4C_{60}^+ + CO \quad (1)$$

$$\rightarrow Fe(CO)_3C_{60}^+ + 2CO \quad (2)$$

apparently results from kinetically or internally excited C_{60}^+ and is completely absent when the C_{60}^+ is subjected to the 1-s cooling period. For comparison, no reaction is observed for $Cr(CO)_6$, possibly due to steric or electronic reasons. CID[21,22] of $Fe(CO)_4C_{60}^+$ (19–92 eV, lab frame) resulted in the consecutive loss of carbonyls to eventually yield FeC_{60}^+. A further increase in CID energy (114–185 eV) gives C_{60}^+ exclusively, consistent with our earlier study.[9] These results indicate that $D°(Fe^+–C_{60}) > D°(Fe^+–CO) = 36.6 \pm 1.8$ kcal/mol,[23] which is also consistent with our previous findings.[9]

Secondary reactions are observed for $Fe(CO)_4C_{60}^+$, reaction 3, and for $Fe(CO)_3C_{60}^+$, reactions 4–7.

$$Fe(CO)_4C_{60}^+ + Fe(CO)_5 \rightarrow Fe_2(CO)_8C_{60}^+ + CO \quad (3)$$

$$Fe(CO)_3C_{60}^+ + Fe(CO)_5 \rightarrow Fe_2(CO)_6C_{60}^+ + 2CO \quad (4)$$

$$Fe_2(CO)_6C_{60}^+ + Fe(CO)_5 \rightarrow Fe_3(CO)_8C_{60}^+ + 3CO\ (78\%) \quad (5)$$

$$\rightarrow Fe_3(CO)_9C_{60}^+ + 2CO\ (22\%) \quad (6)$$

$$Fe_3(CO)_8C_{60}^+ + Fe(CO)_5 \rightarrow Fe_4(CO)_{11}C_{60}^+ + 2CO\ (100\%) \quad (7)$$

Previous studies on the clustering reactions of metal carbonyls have established empirical rules on intermediate ion reactivity and termination steps based on the 18-electron rule.[24-27] Applying these concepts indicates that in this case C_{60} acts like a two-electron donor, as opposed to the expected five- or six-electron donor. However, this is in accord with the limited examples of isolated metal–C_{60} complexes suggesting C_{60} as a two-electron donor via two neighboring carbon positions, similar to an alkene ligand.[7,8] Finally, the decreasing tendency of further clustering reactions with increasing carbonyl coordination suggests that the reactions occur on the metal center, disfavoring a more exotic reaction pattern where individual metal carbonyl groups are attached to the different parts of the C_{60}.

The dramatic difference in reactivity of C_{60}^+ and C_{70}^+ relative to nearby clusters is especially remarkable considering that all of these species presumably consist of >95% fullerene structures.[28] This would seem to implicate electronic factors as the primary determinant of reactivity. Further experimental and theoretical studies are underway to try to determine the relative importance of electronic and geometric factors.

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(19) Carlin, T. J.; Freiser, B. S. *Anal. Chem.* **1983**, *55*, 571.
(20) Comisarow, M. B.; Grassi, V.; Parisod, G. *Chem. Phys. Lett.* **1978**, *57*, 413.
(21) Cody, R. B.; Burnier, R. C.; Freiser, B. S. *Anal. Chem.* **1982**, *54*, 96.
(22) Burnier, R. C.; Cody, R. B.; Freiser, B. S. *J. Am. Chem. Soc.* **1982**, *104*, 7436.
(23) Schultz, R. H.; Crellin, K. C.; Armentrout, P. B. *J. Am. Chem. Soc.* **1991**, *113*, 8591.
(24) Meckstroth, W. K.; Ridge, D. P.; Reents, W. D., Jr. *J. Phys. Chem.* **1985**, *89*, 612.
(25) Wronka, J.; Ridge, D. P. *J. Am. Chem. Soc.* **1984**, *106*, 67.
(26) Foster, M. S.; Beauchamp, J. L. *J. Am. Chem. Soc.* **1971**, *93*, 4924.
(27) Foster, M. S.; Beauchamp, J. L. *J. Am. Chem. Soc.* **1975**, *97*, 4808.
(28) Heldon, G. v.; Hsu, M.; Kemper, P. R.; Bowers, M. T. *J. Chem. Phys.* **1991**, *95*, 3835.

Two-Dimensional Hetero-TOCSY-NOESY. Correlation of ^{31}P Resonances with Anomeric and Aromatic ^1H Resonances in RNA

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The application of ^{31}P NMR to nucleic acids has been limited by the difficulty of resonance assignment, which has depended on either the correlation of resonances with assigned ribose 3′, 4′, 5′, or 5″ proton resonances[1-4] or on regiospecific ^{17}O labeling.[5-9] Assignment by proton correlation fails when the proton spectral dispersion is poor (as in RNA); the use of ^{17}O labeling is tedious. The experiment described here, 2D hetero-TOCSY-NOESY, provides a route for sequential assingment of ^{31}P resonances by correlation to H6, H8, and H1′ resonances of adjacent residues that avoids many of the difficulties of other approaches.

Recently it was demonstrated[10] that composite-pulse heteronuclear cross polarization[11-15] (hetero-TOCSY) could be used to correlate ^{31}P resonances with a number of ribose protons, including H2′ (via the pathway P–H3′–H2′). Because heteronuclear and homonuclear coherence transfer steps occur simultaneously, and because cross peaks are in-phase and absorptive, sensitivity and resolution are better than in alternatives such as INEPT-RELAY[16] and INEPT-TOCSY.[17] Moreover, since the ^{31}P–^1H coupling constants are small (<12 Hz) and ^{31}P is relatively sensitive, hetero-TOCSY is also preferable (in this case) to alternatives such as HMQC-TOCSY.[18]

Double-stranded nucleic acids exhibit strong NOEs from H2′ to intraresidue H1′ and H6 or H8, and often to sequential H6 or H8 protons in the 3′ direction. Thus we reasoned that sequential ^{31}P–aromatic and H1′ correlations would be revealed if a NOESY step were added to the ^{31}P–^1H hetero-TOCSY experiment, and

(1) Pardi, A.; Walker, R.; Rapoport, H.; Wider, G.; Wüthrich, K. *J. Am. Chem. Soc.* **1983**, *105*, 1652.
(2) Marion, D.; Lancelot, G. *Biochem. Biophys. Res. Commun.* **1984**, *124*, 774.
(3) Fu, J. M.; Schroeder, S. A.; Jones, C. R.; Santini, R.; Gorenstein, D. G. *J. Magn. Reson.* **1988**, *77*, 577.
(4) Varani, G.; Cheong, C.; Tinoco, I. *Biochemistry* **1991**, *30*, 3280.
(5) Connolly, B. A.; Eckstein, F. *Biochemistry* **1984**, *23*, 5523.
(6) Ott, J.; Eckstein, F. *Nucleic Acids Res.* **1985**, *13*, 6317.
(7) Ott, J.; Eckstein, F. *Biochemistry* **1985**, *24*, 2530.
(8) Lai, K.; Shah, D. O.; Derose, E.; Gorenstein, D. G. *Biochem. Biophys. Res. Commun.* **1984**, *121*, 1021.
(9) Gorenstein, D. G.; Schroeder, S. A.; Fu, J. M.; Metz, J. T.; Roongta, V.; Jones, C. R. *Biochemistry* **1988**, *27*, 7223.
(10) Kellogg, G. W. *J. Magn. Reson.*, in press.
(11) Bearden, D. W.; Brown, L. R. *Chem. Phys. Lett.* **1989**, *163*, 432.
(12) Zuiderweg, E. R. P. *J. Magn. Reson.* **1990**, *89*, 533.
(13) Brown, L. R.; Sanctuary, B. C. *J. Magn. Reson.* **1991**, *91*, 413.
(14) Artemov, D. Y. *J. Magn. Reson.* **1991**, *91*, 405.
(15) Morris, G. A.; Gibbs, A. *J. Magn. Reson.* **1991**, *91*, 444.
(16) Field, L. D.; Messerle, B. A. *J. Magn. Reson.* **1986**, *66*, 483.
(17) (a) Zagorski, M. G.; Norman, D. G. *J. Magn. Reson.* **1989**, *83*, 167. (b) Hiroaki, H.; Uesugi, S. *FEBS Lett.* **1989**, *244*, 43.
(18) David, D. G. *J. Magn. Reson.* **1989**, *84*, 417.

0002-7863/92/1514-2727$03.00/0

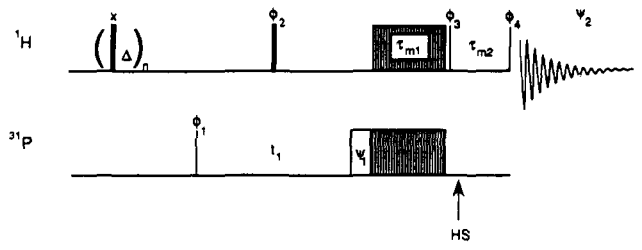

Figure 1. Pulse sequence diagram for 2D hetero-TOCSY-NOESY. The hatched boxes represent a matched multiple pulse mixing sequence for cross polarization. The choice of mixing sequence is discussed elsewhere;[10,12,13,28] the sequence used here was DIPSI-2. The phase cycling is as follows: $\phi_1 = (y, -y)$; $\phi_2 = (x, x, -x, -x)$; $\psi_1 = x$; DIPSI2 = x; $\phi_3 = y$; $\phi_4 = -x$; $\psi_2 = (+, -, +, -)$.

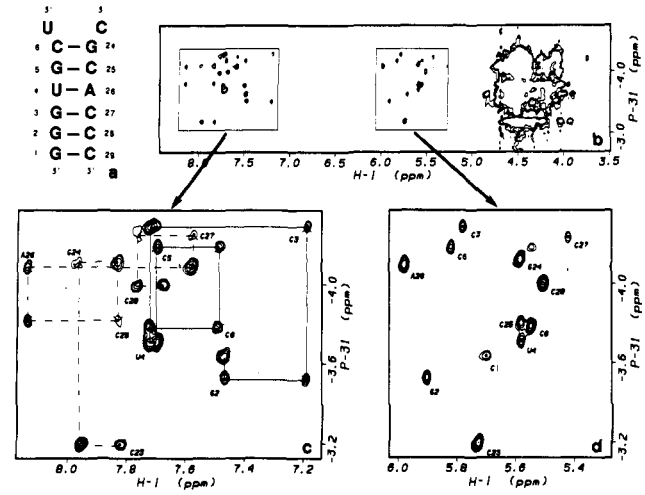

Figure 2. (a) Sequence and secondary structure of the RNA molecule used in this work. The phosphorus atoms are numbered according to the nucleoside on their 5′ side. (b) 2D hetero-TOCSY-NOESY spectrum for the molecule illustrated in part a. The spectrum was recorded using a 3 mM RNA sample in D_2O at 30 °C on a Bruker AM-500 spectrometer (500 MHz, protons; 202 MHz, phosphorus). Other parameters were as follows: τ_{m1} = 72 ms, τ_{m2} = 500 ms, ^{1}H width = 4000 Hz, ^{31}P width = 600 Hz: 94 increments of 320 scans each were recorded. (c) Expanded plot of the ^{1}H aromatic region of the spectrum in part b. For both strands the sequential connectivities are traced with either a solid or a dashed line. Each intraresidue ^{31}P–^{1}H8 or ^{31}P–^{1}H6 cross peak is labeled. (d) Expanded plot of the ^{1}H anomeric region of the spectrum in part b. Each of the 12 intraresidue ^{31}P–^{1}H1′ cross peaks is labeled.

that a powerful ^{31}P assignment method would result.

The 2D hetero-TOCSY-NOESY pulse sequence is shown in Figure 1. ^{31}P magnetization is excited after elimination of ^{1}H magnetization by saturation and is allowed to evolve with refocusing of heteronuclear coupling.[10] The two mixing periods are then applied sequentially. A 3D version of this experiment could be achieved by inserting a second incrementable delay between the mixing times. The 2D experiment was carried out on the oligoribonucleotide whose sequence is shown in Figure 2a. The entire spectrum is shown in Figure 2b. The cross peaks of most interest are in the ^{1}H aromatic (7.0–8.2 ppm) and anomeric (5.0–6.0 ppm) regions.

An expansion of the ^{1}H aromatic region is shown in Figure 2c. Except for G1, each ^{31}P resonance correlates with two aromatic ^{1}H resonances, and each internal aromatic resonance correlates with two ^{31}P resonances. Therefore it was possible to draw a complete set of connectivities for the length of each strand, and all 12 ^{31}P resonances were unambiguously identified. The accuracy of this procedure was confirmed by conventional sequential resonance assignment procedures.[4,19-23] Thus sequential ^{31}P resonance assignments were obtained for this sample without isotopic labeling, even in the absence of prior information about ribose ^{1}H assignments.

In instances where aromatic–^{31}P correlations are less complete, ambiguities can be resolved by comparing the ^{1}H anomeric region (Figure 2d) with assignment data obtained by homonuclear ^{1}H methods. Each ^{31}P resonance should give rise to a single, strong anomeric ^{1}H peak, via the pathway ^{31}P–H3′–H2′–H1′, which is precisely what is seen in Figure 2d.

In summary, a single hetero-TOCSY-NOESY spectrum can lead to sequential assignments for ^{31}P resonances, and for H1′, H8, and H6 resonances in RNA oligonucleotides. The assignment strategy depends, of course, on reasonably efficient transfer from ^{31}P to the 3′ and 2′ protons and NOE connectivities onward to anomeric and aromatic protons. The experiment is necessarily less sensitive than ^{1}H–^{1}H spectroscopy, and the sensitivity of the first step will decrease for larger macromolecules. Nonetheless, it has provided crucial assignment information for a 29-nucleotide RNA under study in this laboratory.[21] This experiment is likely to be most useful for the study of unusual nucleic acid structures,[24-27] drug–nucleic acid interactions, and protein–nucleic acid interactions.

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(19) Hare, D. R.; Wemmer, D. E.; Chou, S.; Drobny, G.; Reid, B. R. *J. Mol. Biol.* **1983**, *171*, 319.
(20) Scheek, R. M.; Boelens, R.; Russo, N.; van Boom, J. H.; Kaptein, R. *Biochemistry* **1984**, *23*, 1371.
(21) Szewczak, A. A.; Chan, Y.-L.; Wool, I. G.; Moore, P. B. *Biochimie* **1991**, *73*, 871.
(22) White, S. A.; Nilges, M.; Brünger, A.; Huang, A.; Moore, P. B. *Biochemistry*, in press.
(23) Cheong, C.; Varani, G.; Tinoco, I. *Nature* **1990**, *345*, 680.
(24) Chou, S.-H.; Flynn, P.; Wang, A.; Reid, B. *Biochemistry* **1991**, *30*, 5248.
(25) Taylor, J.-S.; Garrett, D. S.; Brockie, I. R.; Svoboda, D. L.; Tesler, J. *Biochemistry* **1990**, *29*, 8858.
(26) Pieters, J. M. L.; de Vroom, E.; van der Marel, G. A.; van Boom, J. H.; Konig, T. M. G.; Kaptein, R.; Altona, C. *Biochemistry* **1990**, *29*, 788.
(27) Powers, R.; Gorenstein, D. G. *Biochemistry* **1990**, *29*, 9994.
(28) Kellogg, G. W. *J. Magn. Reson.*, in press.

Oligothiophene Cation Radical Dimers. An Alternative to Bipolarons in Oxidized Polythiophene

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Current understanding of the structure, spectra, and conductivity of oxidized polythiophene and other conducting polymers is dominated by the theory that polarons and bipolarons are formed along single conjugated chains.[1] Because polarons and bipolarons have small-molecule analogues in cation radicals and dications, it is natural to explore that theory by generating these cations and studying their properties.[2] We report that the cation radicals of terthiophenes reversibly dimerize even at low concentration. We suggest that these are π-dimers and that such dimers are reasonable alternatives to bipolarons. As a consequence, π-dimers and π-stacks deserve attention as entities responsible for the properties of oxidized polythiophene and other conducting polymers.

(1) (a) Ferraro, J. R.; Williams, J. M. *Introduction to Synthetic Electrical Conductors*; Academic Press: New York, 1987. (b) *Handbook of Conducting Polymers*; Skotheim, T. A., Ed.; Dekker: New York, 1986; Vols. 1 and 2.
(2) (a) Chang, A.-C.; Miller, L. L. *Synth. Met.* **1987**, *22*, 71. (b) Fichou, D.; Horowitz, G.; Xu, B.; Garnier, F. *Synth. Met.* **1990**, *39*, 243. (c) Fichou, D.; Horowitz, G. *Mater. Res. Soc. Symp. Proc.* **1990**, *173*, 379. (d) Caspar, J. V.; Ramamurthy, V.; Corbin, D. R. *J. Am. Chem. Soc.* **1991**, *113*, 600.

0002-7863/92/1514-2728$03.00/0