(72.5 ppm and 78.6 ppm, respectively) slightly upfield from those of the corresponding precursor thiophosphoryl chlorides (84.6 and 91.5 ppm). Reaction of $AlCl_3$ with an equimolar mixture of the precursors (Y_2XPCl) **e** and **e'** gives ^{31}P NMR signals at 72.3 and 78.2 ppm and shows no phosphorus-phosphorus coupling (i.e., no asymmetric dimers) indicating that these signals correspond to monomeric species. However, while the ^{27}Al NMR spectra contain a sharp signal at 102 ppm ($AlCl_4^-$),[18] the more substantial (>50%) signal is a broad peak slightly downfield (106 ppm). These ^{27}Al spectra are similar to those obtained for $(NMe_2)_3P{=}S{\cdot}AlCl_3$ and $Ph_3P{=}S{\cdot}AlCl_3$.[19] We conclude that, in solution, chloride ion reabstraction occurs allowing formation of a covalent Lewis acid-base adduct ($Y_2PSCl{\cdot}AlCl_3$)[21] and that the ionic nature of **2e** and **2e'** is dependent on a crystal lattice factor. In contrast, a single sharp signal at 102 ppm ($AlCl_4^-$)[18] is observed in the ^{27}Al NMR spectrum of solutions of **2f**. Moreover, the ^{31}P spectrum includes signals for $(Et_2N)_2P^+$ (264.0 ppm)[20] and **2f** (-7 ppm, J_{P-Se} = 341 Hz, identified as a triplet in the ^{77}Se NMR spectrum J_{P-Se} = 340 Hz) and other unidentified signals at 79-80 ppm, demonstrating complex equilibria involving only ionic species.

The identification of derivatives of **2** in the solid state further demonstrates the dominant stability of the phosphetane framework.[14] However, this new cationic system has unique dissociative properties in solution, with the nature of the species dependent upon the chalcogen (X). We anticipate the stabilization of derivatives of **1** through the suitable choice of substituents X and Y.

Supplementary Material Available: Crystal structures, experimental details, unit cell packing diagrams, and tables of crystal data, atomic positional parameters, anisotropic thermal parameters, bond lengths, bond angles, and least-squares planes for $[(Et_2N)_2PS]_2(AlCl_4)_2$ and $[(Et_2N)_2PSe]_2(AlCl_4)_2$ (16 pages); table of observed and calculated structure factors (10 pages). Ordering information is given on any current masthead page.

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(21) Similar adducts have been postulated for reaction of the nitrogen analog (Y_2XPCl, **b**) with $AlCl_3$.[5c]

Binding of 4',6-Diamidino-2-phenylindole (DAPI) to GC and Mixed Sequences in DNA: Intercalation of a Classical Groove-Binding Molecule

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DNA intercalators typically contain planar, fused-aromatic systems that slide between base pairs in the complex.[1] Molecules that bind in the minor groove generally contain unfused-aromatic systems with terminal basic functions.[2] Detailed molecular models

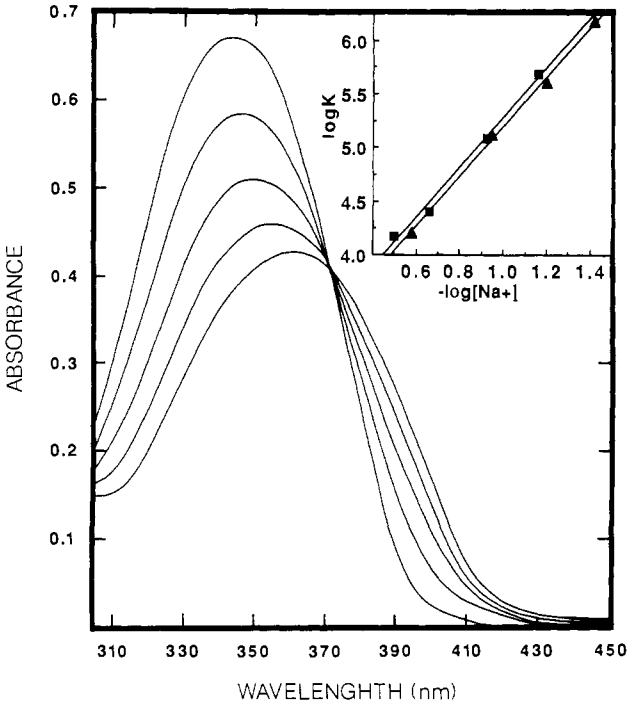

Figure 1. Spectral shifts of DAPI on titration with polyd(G-C)$_2$. The DAPI concentration was 2.5×10^{-5} M, and the polyd(G-C)$_2$ base-pair molarity increased as follows (top to bottom curves at 340 nm): 0, 1.4, 2.7, 4.0, 6.0×10^{-5}. The titration was conducted in a 1-cm cuvette in MES buffer on a Cary 2200 spectrophotometer. The inset shows plots of log K vs -log [Na^+] (NaCl added to MES buffer) for DAPI (■) and quinacrine[12] (▲).

for both binding modes have been developed from X-ray crystallographic and molecular modeling results.[3,4] For example, (i) binding in the DNA minor groove is sterically inhibited by the 2-NH_2 group of G[2,4] and (ii) several aromatic diamidines, 4',6-diamidino-2-phenylindole[5] (DAPI, Figure 2), berenil,[1b] and hydroxystilbamidine,[6] are AT specific minor groove-binding agents.[2] Early investigations with DAPI suggested that it binds to DNA by intercalation,[5e] but more detailed studies indicated that it binds specifically to AT base pairs in the minor groove.[2,5a-d]

The intercalation with DNA of a group of unfused-aromatic cations, similar in structure to classical groove-binding molecules, has led to the conclusion that intercalation and groove-binding modes should be viewed as two variable depth potential wells on a continuous energy surface.[7] This result requires the investigation

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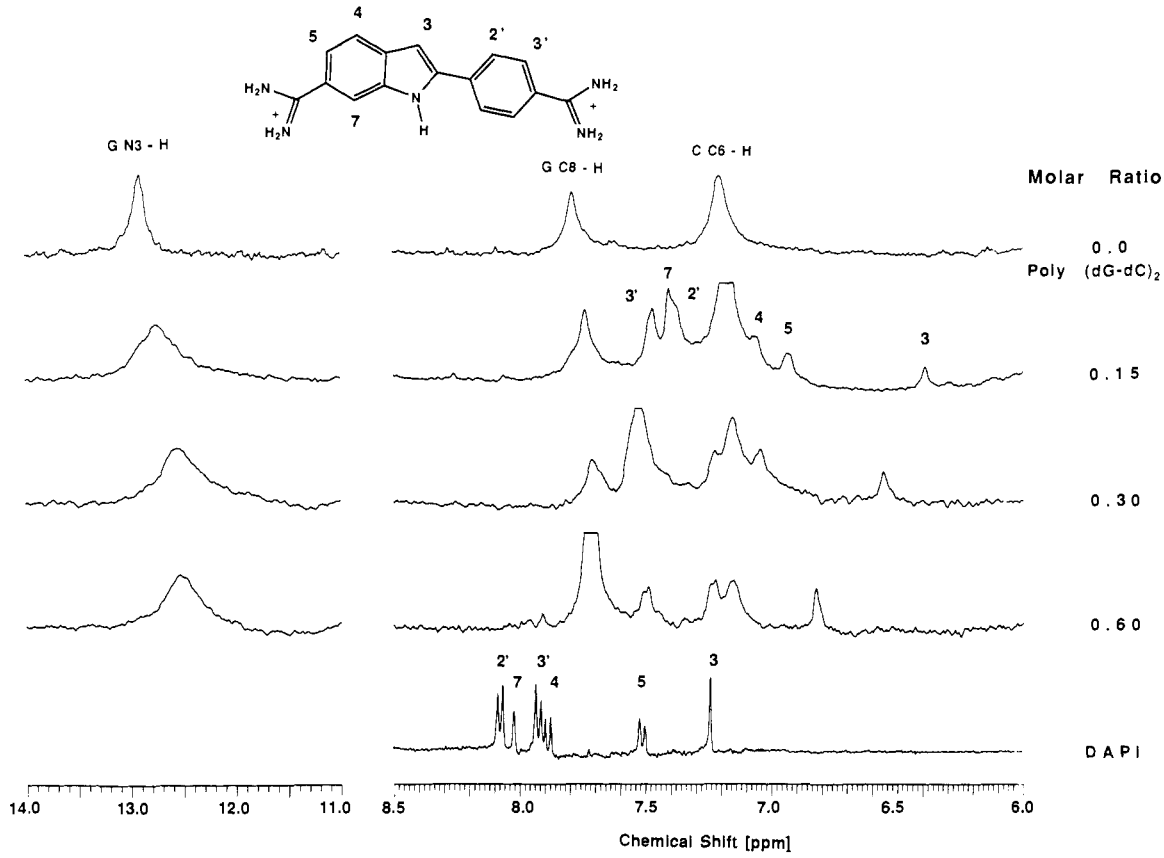

Figure 2. ^{1}H NMR titrations of (right side) the aromatic proton region of DAPI with sonicated polyd(G-C)$_2$ at 80 °C in D_2O and (left side) the imino proton region of sonicated polyd(G-C)$_2$ with DAPI at 30 °C in H_2O/D_2O. The molar ratio of DAPI to base pairs is shown. Spectra were collected[11] on a Varian VXR 400 MHz spectrometer in MES buffer with 0.1 M NaCl. Assignments (figure) were made with standard COSY and NOESY 2D methods.

of the mode by which AT-specific, groove-binding molecules bind to GC-rich regions in DNA to determine whether the groove-specific agents bind in the GC minor groove, simply interact electrostatically with the DNA phosphate groups, or intercalate at GC base pairs. Since a significant effort is being expended to design and synthesize base-pair, particularly GC, specific groove-binding agents,[2,8] it is extremely important to answer these questions. The structure of DAPI is similar to some unfused intercalators,[7] and it was selected for initial analysis with polyd(G-C)$_2$ as a model system. We present here the first conclusive evidence that AT-specific, groove-binding agents can bind strongly to GC-rich regions of DNA by intercalation.

The interaction of DAPI with polyd(G-C)$_2$ results in large visible spectral shifts (Figure 1), a significant induced visible CD signal, and enhanced fluorescence. Spectral changes were used to construct Scatchard plots,[9] and the results are well-fitted by the site exclusion model of McGhee and von Hippel.[10] In a buffer with 0.1 M Na^+, for example, the binding constant (K) is 1.2×10^5 M^{-1} with 2–3 base pairs per binding site, similar to results for ethidium under these conditions.[11] A plot of log K vs −log [Na^+] (inset, Figure 1) has a slope of 2.3 suggesting two ion pairs in the DAPI complex with ion release as expected for intercalation.[11] The DAPI binding constants and slope are similar to those observed for intercalation of the dicationic acridine quinacrine with DNA[11b] (inset, Figure 1). The binding of DAPI to AT base pairs[5] is stronger than the binding to GC base pairs, but this is because the binding in AT regions is very strong, not because the binding in GC regions is weak.

Viscometric and NMR titrations of polyd(G-C)$_2$ with DAPI were used to evaluate the binding mode.[9] Increases in viscosity, up to a ratio of 0.2–0.3 (compound/base pair), are very similar for DAPI and intercalators.[12] ^{1}H NMR experiments to monitor the chemical shifts of aromatic protons of molecules on addition of sonicated DNA provide a sensitive, direct method of distinguishing intercalation from groove-binding modes.[7,13] All proton signals of DAPI shift significantly upfield and broaden on addition of sonicated polyd(G-C)$_2$, and the imino and aromatic protons of polyd(G-C)$_2$ also shift upfield on addition of DAPI (Figure 2) as expected for an intercalation binding mode.[7,13]

For a dication a plot of log τ (the average dissociation kinetics half-life) as a function of log [Na^+] will have a slope of approximately −0.6 for intercalation and of −1.6 for groove-binding.[11,14] For the SDS driven dissociation of DAPI from polyd-(G-C)$_2$ the slope of such a plot is -0.8 ± 0.1 (τ of 3.0 ms, 0.1 M Na^+). Dissociation experiments with polyd(A-T)$_2$ yielded a slope of -1.8 ± 0.1 (τ of 150 ms, 0.1 M Na^+). These results for DAPI strongly support an intercalation binding mode at GC sites and groove-binding interactions at AT sites.[1,2,7,14]

Stereochemically reasonable complexes can be constructed with DAPI and an intercalation site of standard geometry.[15] As

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(12) Viscometric titrations were also performed with sonicated calf thymus DNA and Col E_1 superhelical DNA. The viscosity increases with calf thymus DNA were slightly less than one-half those obtained with polyd(G-C)$_2$. Ethidium gave a viscosity maximum at a ratio of 0.05 with the superhelical DNA, while DAPI had a maximum at approximately 0.3 and did not rewind the DNA as efficiently as ethidium. Both of these results are consistent with intercalation of DAPI at GC sites on natural DNA after saturation of the strong AT binding sites.

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observed with the cationic side chains of an unfused intercalator,[7] the amidine groups of DAPI cannot sterically fit into the minor groove but can fit quite well into the DNA major groove in a CG intercalation site (not shown). The phenyl and indole rings of DAPI stack well with base pairs at the intercalation site, and the cationic amidines associate with phosphate groups on opposite chains of the complex by H-bonding and electrostatic interactions.

Our results clearly demonstrate that DAPI binds to polyd(G-C)$_2$ by intercalation with binding strength and kinetics in the range typical of intercalators. More limited results indicate that DAPI binds to polyd(A-C)·polyd(G-T) in a manner similar to that for polyd(G-C)$_2$ but quite different from its groove-binding mode with polyd(A-T)$_2$. This is consistent with observations that groove-binding of DAPI requires three to four consecutive AT base pairs.[5] DAPI can thus intercalate at a broad range of sites, with little specificity, as with most other intercalators. More GC and mixed base-pair intercalation sites are available than consecutive AT base-pair, groove-binding sites on any natural DNA sequence. DAPI should thus be viewed as an intercalator that has unusual and very favorable interactions in the minor groove at AT sequences. The intercalation binding mode would not be easily detected by methods such as DNA footprinting due to its short lifetime. Preliminary modeling studies with other classical AT specific-groove-binding molecules suggest that they can also bind to GC base pairs by intercalation.

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(15) Molecular mechanics calculations were conducted on a MicroVaxII-Evans and Sutherland PS390 system with the program MACROMODEL (Professor Clark Still, Columbia University). DAPI was docked in a central CG intercalation site (to be described elsewhere) in the dodecamer sequence d(CGTACGTACG) and minimized to a gradient of less than 0.1 kcal/mol·Å. Various orientations of DAPI, with the amidine groups in both the minor and the major grooves, were considered in the initial docking experiments. In all minor groove complexes the DAPI was forced out of the intercalation site by unfavorable van der Waals contacts.

Metalloradical Activation of CO: Formation and Carbonyl Coupling of a Bent 17-Electron M–CO Unit

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Carbonyl carbon centered radical reactivity in organotransition-metal complexes has been previously observed only for transient 19-electron species.[1-3] Abstraction of hydrogen from metal hydrides by $[Fe(CO)_5]^-$ to form an intermediate 18-electron formyl complex, $[Fe(CO)_4CHO]^-$, is a prominent example of this type of reactivity.[1] The characteristic reactions of 17-electron metal carbonyl species are one-electron reduction, M–M and M–X bond formation which occur at the metal center to produce 18-electron species.[4-7] Studies of rhodium porphyrins with CO that produce metalloformyl,[8-10] dimetal ketones,[11,12] and dimetal α-diketones[12] have suggested that a 17-electron metalloorganic radical, (por)Rh–$\dot{C}$O,[10] may function as an intermediate, but this type of species has eluded direct observation. This article reports on the reversible reaction of tetramesitylporphyrinrhodium(II), (TMP)Rh$^\bullet$, **1**, with CO to form (TMP)Rh–$\dot{C}$O, **2**, which dimerizes by C–C bond formation. EPR and reactivity studies of **2** indicate the presence of a bent CO fragment with a partially rehybridized CO unit that facilitates one-electron reactions at the carbonyl carbon center.

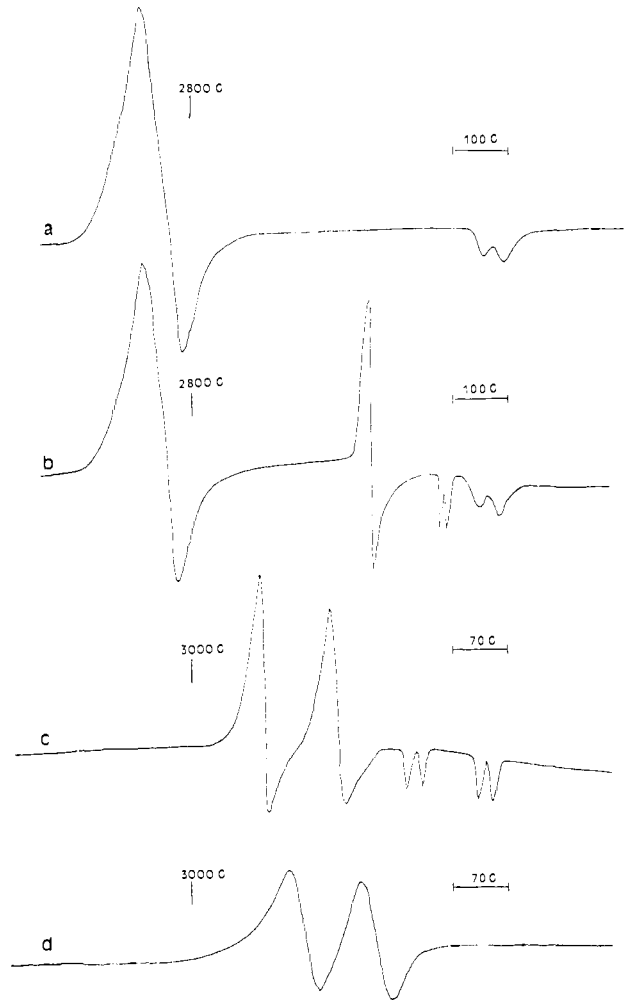

Figure 1. EPR spectra for (TMP)Rh^{II} and (TMP)Rh–$\dot{C}$O in toluene. (a) Anisotropic EPR spectrum for (TMP)Rh^{II} (90 K) [$g_{1,2}$ = 2.65, g_3 = 1.915; $A^{103}Rh(g_{1,2})$ = 197 MHz, $A^{103}Rh(g_3)$ = 158 MHz]. (b) Anisotropic EPR spectrum that results from exposing a frozen solution of (TMP)Rh^{II} to ^{12}CO (P_{CO} = 200 Torr, T = 100 K), warming to 195 K and refreezing to 100 K. (c) Anisotropic EPR spectrum for (TMP)-Rh–$^{13}\dot{C}O$ (90 K) [$g_1 \simeq 2.17$, $A^{13}C(g_1) \simeq 307$ MHz; g_2 = 2.14, $A^{13}C(g_2)$ = 330 MHz; g_3 = 1.995, $A^{13}C(g_3)$ = 299 MHz; $A^{103}Rh(g_3)$ = 67 MHz]. (d) Isotropic EPR spectrum for (TMP)Rh–$^{13}\dot{C}O$ (90 K) [$\langle g\rangle_{iso}$ = 2.101; $\langle A^{13}C\rangle$ = 312 MHz].

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