reduced complex has a $g_\perp = 2.181$ and a $g_\parallel = 2.003$. Both signals are split into triplets ($A_\perp = 9 \times 10^{-4}$ cm^{-1} and $A_\parallel = 16 \times 10^{-4}$ cm^{-1}). In CH_3CN superhyperfine lines (triplets) are observed in the $g_\parallel$ of $[Rh_2(N_2R_2CR)_4]^-$ (Figure 3b) and may be due to a coupling of one ^{14}N nucleus ($I = 1$) of CH_3CN. However, the fact that identical g values (±0.002) and splittings are obtained in a bonding and a nonbonding solvent suggests that there may be no interaction between the negatively charged dirhodium(II,I) complex and the solvent.

The $g_\parallel = 2.003$ for $[Rh_2(N_2R_2CR)_4]^{-1}$ is consistent with the prediction that an unpaired electron in a σ_{M-M}-type orbital should have $g_\parallel \approx g_e$.[10] The LUMO of $Rh_2(O_2CR)_4$ complexes with weak or no axial ligands has generally been believed to be a σ^* Rh–Rh orbital with little or no contribution from the σ^* Rh-axial ligand.[10,13–16] Thus, in spite of the differences between $Rh_2(O_2CR)_4$ and $Rh_2(N_2R_2CR)_4$, the LUMO of $Rh_2(N_2R_2CR)_4$ appears to be primarily σ^* Rh–Rh. If this is the case, the superhyperfine structure in the $g_\parallel$ of Figure 3b may be due to a minor contribution from σ^* Rh–L where L = CH_3CN.

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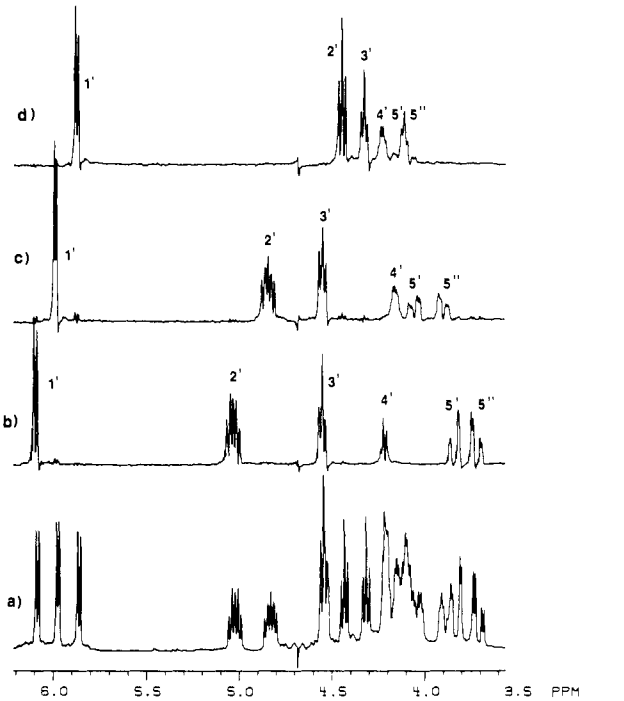

Figure 1. Spectra, 270 MHz, of the ribose region of A2′-P-5′A2′-P-5′A. (a) Regular spectrum; (b–d) selective excitation subspectra of A2′-P, P-5′A2′-P, and P-5′A nucleotides. For all three difference spectra, propagation was started at the anomeric proton and a propagation delay of 180 ms was used.

Simplification of ^{1}H NMR Spectra by Selective Excitation of Experimental Subspectra

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We propose a new method for the simplification of ^{1}H NMR spectra by the generation of experimental subspectra. The method is based on the concept of spin propagation[1–6] via a homonuclear Hartmann–Hahn coherence transfer process[6,7] and is the improved one-dimensional (1D) analogue of the 2D HOHAHA experiment, described previously.[3] For spectra of limited complexity, 1D versions of 2D experiments can have significant advantages over the 2D experiment because of shorter minimum measuring time, reduced data storage requirements, and improved digital resolution.

The basic idea is to invert the resonances of an isolated spin multiplet with a selective 180° pulse and then to let this inverted magnetization propagate through the ^{1}H coupling network. A difference spectrum (with the decoupler switched off during the selective pulse interval) then yields a subspectrum of all hydrogens directly or indirectly scalar coupled to the inverted ^{1}H resonance. By studying the subspectrum as a function of propagation time, a straightforward spectral assignment within the subunit is obtained.

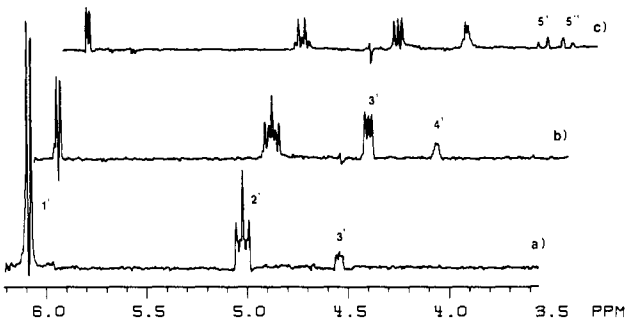

Figure 2. Propagation subspectra of the ribose ring of the A2′-P nucleotide, obtained for propagation times of (a) 40, (b) 80, and (c) 120 ms. Spectra b and c are horizontally displaced to improve the visual appearance. For increasing propagation delay, the 1′ proton magnetization propagates into the 5′,5″ direction at a rate determined by the various coupling constants between vicinal protons.

We demonstrate the method for the trinucleotide A2′-P-5′A2′-P-5′A, a potent inhibitor of protein synthesis. The regular 270-MHz ^{1}H spectrum (Figure 1a) of the ribose region shows a substantial amount of overlap. The anomeric protons, however, are well separated in the 5.85–6.1 ppm region and serve as labels that can separately be inverted by a frequency-selective 180° pulse. By use of 180-ms propagation periods, the experimental subspectra of the three ribose rings (Figure 1b–d) were obtained by starting the propagation from the anomeric protons. The individual rings are also readily identified: H2′ in Figure 1d is a triplet and does not show coupling to ^{31}P, assigning this spectrum to the P-5′A nucleotide. Similarly, the H5′ and H5″ resonances in Figure 1b (when examined at higher resolution than shown) do not exhibit coupling to ^{31}P, assigning this spectrum to A2′-P.

Spectral assignment within each subspectrum can be obtained by variation of the duration of the propagation period. For short propagation times (~40 ms) magnetization has not propagated

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much further than to its direct coupling partner (H2′). For longer times, the magnetization propagates further away from the starting point (the anomeric hydrogen in this case) into the H5′,5″ direction (Figure 2b,c). Spectral assignments are in agreement with those proposed by Doornbos et al.,[8] who based their results on 360-MHz double-resonance experiments and on comparison with spectra of the dinucleotide A2′-P-5′A.

The actual pulse sequence used in the present work is as follows: $180°_{sel}$ (on/off), $90°_x$, $(SL_y60°_{-y}300°_ySL_{-y}60°_y300°_{-y})_n$, Acq(±). The sequence between parentheses is the actual propagation scheme, where SL_y and SL_{-y} denote spin lock along the y and $-y$ axis, respectively. As explained elsewhere,[3,6] the duration τ of the spin lock period SL_y (and SL_{-y}) should obey

$$\tau < \nu/2\Delta^2$$

where ν is the nominal rf field strength during the spin lock, and Δ is the largest offset of a resonance of interest to the carrier frequency. The carrier frequency is placed at the center of the region of interest to minimize the value of Δ. The extra pulse pair, $60°_{-y}300°_y$ (and its inverse) is a composite pulse that rotates magnetization from the $+y$ effective rf field to the $-y$ effective rf field direction. This limits the loss of spin-locked magnetization during phase alternation of the spin-lock field to less than 1% for Δ/ν ratios of up to 0.3. This permits the use of rather weak rf fields (2–10 kHz) for most cases of interest (covering 1200–6000-Hz spectral width). Therefore, several watts of rf power is generally sufficient to induce effective spin propagation. Considering the rf duty cycle over the entire experiment ($<\sim$10%), this is well within the safe range for most commercial high-resolution spectrometers. On some spectrometers, the low-power mode provides a sufficiently strong rf field; on our spectrometer a ENI Model 420 L rf power amplifier was used to further amplify the low-power mode rf. The ^1H heteronuclear decoupler amplifier can also be used for this purpose. Use of the high-power observe amplifier for generating the spin-lock field should be discouraged since this may lead to probe or attenuator damage.

The spin propagation method is not restricted to systems that consist of well-defined subunits that are not mutually coupled (peptides, oligonucleotides, oligosaccharides) but can also be used in very complex coupling networks as often found in steroids and alkaloids. In these cases one can use the propagation time dependence to assign hydrogen resonances that are an increasing number of bonds removed from the starting point since the relay of magnetization proceeds sequentially between coupled pairs of spins, with each step occurring at a rate determined by the coupling constant. It should be pointed out that although the subspectra appear to be absorptive, there is also antiphase dispersive character within the multiplet.[2,9] This partial antiphase dispersive nature of the individual multiplet components can lead to errors if one attempts to measure precise coupling constants from such subspectra. Computer simulations and experimental results suggest that for systems containing four or more nonequivalent hydrogens, the dispersive components tend to disappear for long propagation times, and the multiplets approach their "natural shape". Currently, we are investigating whether it is possible to measure accurate J values from the time dependence of the propagation rather than from the multiplet splittings.

Cross-relaxation among spin-locked magnetization components[10] also forms a transfer mechanism. However, this effect is relatively small and the resulting resonances in the difference spectrum are of negative sign.[7]

The one-dimensional propagation method provides a very simple and fast way to obtain spectral assignment in complex overlapping spectral regions. The major limitation is the decay of magnetization, with time constant $T_{1\rho}$, which approximately equals T_2 and which prohibits the use of long propagation times for macromolecules. New propagation schemes, based on composite pulse decoupling cycles,[11] may alleviate this problem.[12]

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Synthesis of 9-Decarboxymethoxatin. Metal Complexation of Methoxatin as a Possible Requirement for Its Biological Activity

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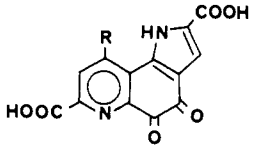
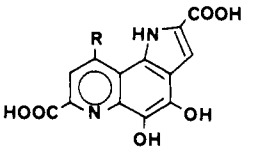
The pyrroloquinoline quinone methoxatin[1-3] ($\mathbf{1}_{ox}$) is the cofactor for certain non-flavin or nicotinamide dependent bacterial dehydrogenases known as quinoenzymes. The Cu^{II} requiring bovine serum amine oxidase[4] has recently been reported to be a quinoenzyme. Substantiation of this claim would almost certainly amount to the finding that $\mathbf{1}_{ox}$ is a human cofactor and, therefore, plausibly a vitamin.



$\mathbf{1}_{ox}$, R = COOH
$\mathbf{2}_{ox}$, R = H



$\mathbf{1}_{red}$, R = COOH
$\mathbf{2}_{red}$, R = H

It has gone without notice that structural features of $\mathbf{1}_{ox}$ are shared by known chelating agents. With attention to the 7-carboxyl group, $\mathbf{1}_{ox}$ may be viewed as a derivative of the metal complexing agent[5] α-picolinic acid. Two-electron reduction of $\mathbf{1}_{ox}$ by substrate provides the dihydrodiol $\mathbf{1}_{red}$. The latter is both a derivative by substitution and annelation of α-picolinic acid and the metal complexing agent[6] 8-hydroxyquinoline. Perhaps these features are germane to the mechanism of metal-requiring quinoenzymes.

Methoxatin must still be considered as a rather rare and generally unobtainable compound. In order to ascertain the structural requirements for biological activity and study the chemistry[7-9] and in particular the metal-binding properties of $\mathbf{1}_{ox}$ and $\mathbf{1}_{red}$ we have designed a synthesis of an easily attainable analogue, 9-decarboxymethoxatin ($\mathbf{2}_{ox}$).

8-Hydroxyquinoline was converted to 2-cyano-8-hydroxyquinoline (**3**) in three steps by a literature route.[10] Nitration of **3** in concentrated nitric–acetic acid at 20 °C produced a mixture of the 5- and 7-nitro isomers from which the 5-nitro isomer[11] **4**

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