normalized with respect to the intensity at +0.2 V in the absence of any solution species. This quantity was different for thin and thick films and, hence, only the relative magnitudes should be considered. Figure 15a,b are plots of the variation of emission intensity with H_2Q concentration for thin (a) and thick (b) films. At +0.2 V there is very little change in intensity, showing that H_2Q does not quench to any significant extent. At +0.8 V H_2Q is oxidized to benzoquinone and the decreased emission intensity shows that benzoquinone quenches $Ru(bpy)_3{}^{2+*}$. At +1.4 V, in the case of thin films, most of the $Ru(bpy)_3{}^{2+}$ is oxidized to the 3+ state as shown by the low intensity in Figure 15a. However, with thicker films a considerable amount of $Ru(bpy)_3{}^{2+}$ in the film remains in the reduced state, even when the electrode is held at +1.4 V. Quenching still occurs at thick films (Figure 15b) showing that considerable interaction still occurs between the solution- and polymer-bound species at the film/solution interface. Similar results are observed in the case of Fe^{2+}/Fe^{3+}, the only difference being that both Fe^{2+} and Fe^{3+} are capable of quenching $Ru(bpy)_3{}^{2+*}$ (Figure 15c,d).

$K_4Fe(CN)_6$ exhibits a different behavior. Neither $Fe(CN)_6{}^{4-}$ nor $Fe(CN)_6{}^{3-}$ penetrates the Nafion polymer film. However, regeneration of $Ru(bpy)_3{}^{2+}$ from $Ru(bpy)_3{}^{3+}$ can occur at the film/solution interface. This happens when the oxidized 3+ species "diffuses" to the film/solution interface, is reduced, and then diffuses back again. (This diffusion occurs by both electron hopping and mass transfer.) Thus, two rate processes can influence the emission intensity. The first one is the diffusion process, which is directly affected by the thickness of the film, and the second is the rate of reconversion at the film/solution interface, which does not directly depend on the thickness. The observed emission intensity will depend on which of the processes dominates under given conditions. The results in Figure 15e suggest that $Fe(CN)_6{}^{4-}$ is not oxidized at the filmed electrode at +0.8 V, and, hence, no change occurs. At +1.4 V most of the $Ru(bpy)_3{}^{2+}$ is oxidized to 3+ state at the thin electrode (Figure 15e). However, since electron propagation is faster in thin films, the 3+ ions are reduced back to 2+ at the film/solution interface, thus increasing the emission intensity. At higher concentrations this reconversion becomes more efficient up to 10 mM beyond which electron transport through the film becomes limiting. The situation is very different in the case of thick films (Figure 15f). The oxidation of $Ru(bpy)_3{}^{2+}$ itself is not very efficient as indicated by the small difference in the luminescent intensity between +0.2 and 1.4 V in the absence of an $K_4Fe(CN)_6$. Secondly, we do not observe any effective reconversion of $Ru(bpy)_3{}^{3+}$ into the 2+ state. Since the rate of this reaction at the film/solution interface should be independent of film thickness, the absence of effective reconversion supports slow electron propagation in the thick films.

Conclusion

The experimental results obtained in this investigation of the photoelectrochemical properties of Nafion polymer-bound $Ru(bpy)_3{}^{2+}$ indicate that the decrease in conversion efficiency with increasing film thickness arises mainly from limitations due to charge transport through the film. This transport is associated with a net motion of counterions across the polymer phase to maintain electroneutrality. Albery et al.[26] have shown that for thionine-coated electrodes increasing the anion size decreased the charge-transfer rate through the film. Thus, diffusion of counterions could effectively slow the net propagation of charge. However, previous studies of Nafion films[25c] have shown that the apparent diffusion coefficient for electrons remained the same when the anion size was increased. This result was confirmed for luminescence experiments with electrolytes having bigger anions. Increasing the counterion size did not result in any noticeable decrease in emission intensity for the thin films in the presence of $Fe(CN)_6{}^{4-}$. If counterion motion was the slow step, increasing the counterion size would hinder electron propagation even in thin films and, thus, the reconversion efficiency.

We have also shown that the efficiency for photosensitization with these polymer electrodes is not improved to any practically important level compared to derivatized electrodes with adsorbed monolayers. Photocurrents are still small (~nA cm^{-2}). While the efficiency increases slightly with thickness because of improved light absorbance, the increasing film thickness results in increased hindrance of electron flow through the film, which then limits the photocurrent. For Nafion films the optimum thickness appears to be about 0.3 to 0.5 μm.

In many cases the application of polymer electrodes to catalysis appears to be limited by the "film resistance" (i.e., charge propagation through the film). Improvements may be possible in these applications as well as in photosensitization experiments by improving the film conductivity via the incorporation of electronically conductive zones within the film.[25a] Experiments of this type are planned.

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Registry No. SnO_2, 18282-10-5; $Ru(bpy)_3{}^{2+}$, 15158-62-0; $FeSO_4$, 7720-78-7; $K_4Fe(CN)_6$, 13943-58-3; $Ru(bpy)_3{}^{3+}$, 18955-01-6; Nafion, 39464-59-0; hydroquinone, 123-31-9; benzoquinone, 106-51-4; carbon, 7440-44-0.

Homonuclear Decoupling in Heteronuclear Chemical Shift Correlation Spectroscopy. 1. Study of Progesterone

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Abstract: $^1H-^{13}C$ chemical shift correlation spectroscopy was improved by selective reversal of distant protons which eliminates most homonuclear J couplings. The method facilitated easy assignment and separate measurements of ^1H chemical shifts as well as geminal couplings in progesterone. Nonuniform values of one-bond coupling constants $^1J_{CH}$, strong coupling in the proton spin system, and other apparent obstacles against the routine use did not cause serious problems.

Two-dimensional (2D) NMR techniques[1] can provide valuable information about a spin system, but they also require a certain time commitment for data acquisition and processing. Measuring times are especially critical in heteronuclear ($^1H-^{13}C$) versions where signals of low-sensitivity isotopes are detected. Efficiency

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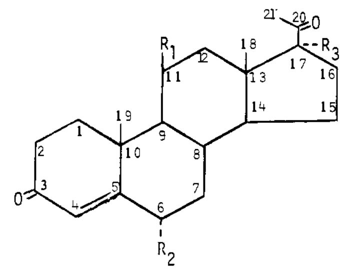

1 $R_1=R_2=R_3=H$

2 $R_1=OH$, $R_2=R_3=H$

3 $R_1=H$, $R_2=Me$, $R_3=OAc$

Figure 1. Structures of progesterone (**1**), 11β-hydroxyprogesterone (**2**), and 17α-acetoxy-6α-methylprogesterone (**3**).

can be improved by recent approaches[2-6] which are based on a relatively simple idea: suppression of the "undesired" type of interaction intentionally modifies the data sets and extraction of the "desired" exclusive information is accomplished with better resolution and within a shorter time.

Bax has reported[6] that homonuclear J coupling can be eliminated in $^1H-^{13}C$ chemical shift correlation maps of CH groups. Our paper evaluates extension to CH_2 and CH_3 groups, and it clearly shows that complete homonuclear decoupling can be achieved except in the case of CH_2 with nonequivalent protons. This new approach gives rise to superior correlation maps which will become indispensable for studies of complex molecules. As an example ^1H and ^{13}C chemical shfts are measured and unambiguously assigned in progesterone (**1** in Figure 1). We also discuss some apparent obstacles (such as nonuniform values of one-bond coupling constants $^1J_{CH}$, strong coupling in the proton spin systems, etc.) against the routine use.

Description of the Method

Traditional correlation maps[7-10] do separate ^1H and ^{13}C chemical shifts along the F_1 and F_2 dimensions, but homonuclear J couplings are also shown. Although "more information" is obtainable, practical realization of the experiment usually does not provide enough data points to resolve all lines. Resonances are simply broadened along the F_1 dimension thus hampering ^1H chemical shift measurements.

Pulse sequence

$$\begin{array}{lll} ^1H: & \pi/2(\phi_1)\text{-}t_1/2\text{-}\pi/2(\phi_2)\text{-}\tau\text{-}\pi(\phi_1)\text{-}\tau\text{-}\pi/2(\phi_2)\text{-}t_1/2\text{-}\tau\text{-}\pi/2(\phi_3)\text{-}\tau'\text{-dec.} & \\ ^{13}C: & \qquad\qquad \text{-}\pi(\phi_3)\text{-} \qquad\qquad\qquad \text{-}\pi/2(\phi_2)\text{-} \quad \text{-acq.}(\pm) & \\ & | \quad \text{evolution} \qquad\qquad\qquad | \text{ mixing } | & [A] \end{array}$$

combines chemical shift correlation with pulses

$$\begin{array}{lll} ^1H: & \ldots\text{-}\pi/2(\phi_2)\text{-}\tau\text{-}\pi(\phi_1)\text{-}\tau\text{-}\pi/2(\phi_2)\text{-}\ldots & [B] \\ ^{13}C: & \qquad\quad \text{-}\pi(\phi_3)\text{-} & \end{array}$$

at the middle of the evolution time. Since quadrature phase detection must be accomplished along both dimensions[10] and multiple quantum coherence transfer echoes[11] should be suppressed, a four-step phase cycle (Table I) has been used together with the standard quadrature phase detection and automatic baseline correction.

Table I. Relative Phases of Radio Frequency Pulses in [*A*] As Denoted in the Doubly Rotating Frame

step	φ_1	φ_2	φ_3
1	$-y$	x	x
2	$-x$	$-y$	$-x$
3	y	$-x$	x
4	x	y	$-x$

Table II. ^1H Chemical Shifts and Geminal Couplings in Cyclohexanol

position	chemical shift, ppm	geminal coupling, Hz
1	3.542[a]	
2α	1.253	12.5[b]
2β	1.867	12.5
3α	1.241	13.1
3β	1.714	13.1
4α	1.148	13.2
4β	1.526	13.2

[a] Accuracy ±0.005 ppm. [b] Accuracy ±0.5 Hz.

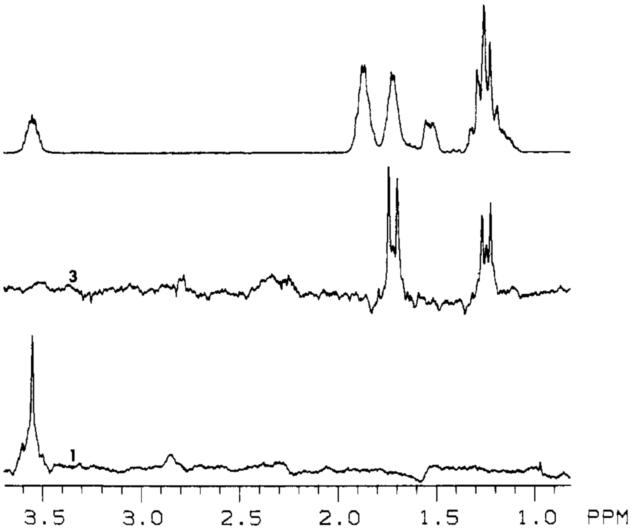

Figure 2. Comparison between the one-dimensional ^1H spectrum of cyclohexanol (upper trace) and cross sections along the F_1 dimension as obtained by pulse sequence [*A*]. If one proton is attached to a ^{13}C spin, selective reversal of distant protons eliminates all homonuclear J couplings (bottom trace). When two nonequivalent protons are bonded to the same carbon, pulse sequence [*A*] does not suppress their geminal coupling and the cross section shows four peaks (middle trace). The broad tail which is superimposed on the sharp peak in the bottom trace is due to the fact that distant protons partially evolve during the intervals 2τ and they are not completely reversed by pulses [*B*].

Pulse sequence [*A*] detects signals of ^{13}C spins in isotopically labeled molecules, therefore, protons are divided into two groups: (i) directly attached ones experience one-bond coupling $^1J_{CH}$ with ^{13}C spin, while (ii) other protons in the molecule are distant and their homonuclear and heteronuclear couplings are significantly smaller than $^1J_{CH}$. If the interval τ is set equal to $\tau_0 = 1/(2^1J_{CH})$, pulses [*B*] apparently do not affect attached protons.[4,5] On the other hand, distant protons are selectively reversed, and precession resulting from coupling between attached and distant protons is refocused during the second half of the evolution time ($t_1/2$).

The mixing period accomplishes polarization transfer via INEPT[12,13] and the last delay $\tau' = 1/(4^1J_{CH})$ establishes partial phase coherence of ^{13}C components which are detected during the ac-

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quisition time t_2. Signals are first separated with respect to ^{13}C chemical shifts (F_2 dimension), and then they are followed as a function of the variable evolution time t_1.

If equivalent protons are bonded to a given carbon, only one peak is detected along the F_1 dimension and the appropriate cross section is utilized for an accurate measurement of the ^{1}H chemical shift. The situation becomes entirely different when two nonequivalent protons (H_α and H_β) are attached to the same carbon in a CH_2 group. Since both are apparently not affected by pulses [*B*], precession resulting from the geminal coupling $^2J_{H_\alpha H_\beta}$ is not refocussed during the second half of the evolution time. The cross section along the F_1 dimension shows pairs of peaks centered at chemical shifts δ_{H_α} and δ_{H_β} and further split by $^2J_{H_\alpha H_\beta}$ (Figure 2).

This fact can be considered as a disadvantage, because the intensity is dispersed into four resonances and the measuring time becomes significantly longer. On the other hand, pulse sequence [*A*] represents a unique approach which facilitates separate determinations of geminal couplings even without resolving the complete pattern (Figure 2). Table II summarizes results for cyclohexanol. Even for this relatively small molecule one-dimensional ^{1}H spectra are very difficult to interpret,[14] while the indirect detection via pulse sequence [*A*] provides exclusive information in a simple way.

Pulses [*B*] have to discriminate attached and distant protons. Since the values of $^1J_{CH}$ can range from 125 up to 250 Hz, it is impossible to adjust the delay $\tau = \tau_0$ accurately for all spins. Missetting of τ hampers efficiency for pulses [*B*] and spurious peaks can appear in the spectra, but their positions are predictable and they do not interfere with the intended measurements.[15]

Attached protons are evolving due to $^1J_{CH}$ during the time 2τ and they have a probability[15,16]

$$p_0 = (1 - \cos(\pi\tau/\tau_0))/2 \qquad (1)$$

for being brought to the initial direction when pulses [*B*] are applied. On the other hand, there is also a probability

$$p_1 = (1 + \cos(\pi\tau/\tau_0))/2 \qquad (2)$$

that they are flipped over. If $\tau \neq \tau_0$, p_1 becomes different from zero and a part of the ^{1}H magnetization is reversed at the middle of the evolution time. Precession resulting from chemical shift dispersion is refocussed, while precession resulting from homonuclear and heteronuclear *J* couplings is continued during the second half of the evolution time thus leading to spurious peaks which are centered at $\pm^1J_{CH}/2$ and further split by homonuclear *J* couplings. It is sometimes possible to observe the spurious peaks, but in typical spin systems too many homonuclear couplings are present and the intensity drops below the noise level.

The majority of the attached protons (probability p_0), however, behaves as supposed in the description of pulse sequence [*A*], and they give rise to a limited number (≤ 4) of real peaks which are determined by ^{1}H chemical shifts and geminal couplings. In most cases the missetting of τ should not exceed $\sim$25% and the intensity will drop for less than $\sim$15% due to partial reversal of attached protons. It must be mentioned that nonuniform values of $^1J_{CH}$ also impede polarization transfer during the INEPT mixing period,[13] and a small additional loss cannot be avoided. Our experiments using pulse sequence [*A*] with variable τ values confirm that when τ is within 20–25% of τ_0, spurious signals at $\pm^1J_{CH}/2$ could hardly be detected and the loss of intensity of the real signal is within 15–20%.[17]

Since $^1J_{CH}$ is not always an order of magnitude larger than the sum of homonuclear and long-range heteronuclear couplings, it is also obvious that distant protons do evolve during the time 2τ. They are not completely reversed by pulses [*B*], and precession resulting from homonuclear couplings is partially continued during the second half of the evolution time. These spurious resonances are centered at the ^{1}H chemical shift, but they show a homonuclear coupling pattern thus contributing to a "tail" which is superimposed on the sharp "real" peak (Figure 2).

Strong coupling in the proton spin system gives rise to systematic errors and spurious peaks in 2D homonuclear spectroscopy.[1] Indirect detection of ^{1}H resonances by pulse sequence [*A*] utilizes signals only from ^{13}C-labeled molecules, and the presence of the magnetically active isotope changes energy levels of the attached proton (H_a). The essential difference will be illustratred by an analysis of the so called ABX spin system[18] with $A = H_a$, $B = H_d$, and $X = {}^{13}C$ where the homonuclear coupling $J_{H_aH_d}$ between the attached and distant proton is comparable to the difference of frequencies $\nu_{H_a} - \nu_{H_d}$ arising from chemical shifts.

During the first half of the evolution time the precession frequency ν_1 of the attached proton is equal to[18]

$$\nu_1 = \tfrac{1}{2}(\nu_{H_a} + \nu_{H_d} + J_{H_aH_d} + \tfrac{1}{2}(J_{H_aC} + J_{H_dC}) + R_1) \qquad (3)$$

where

$$R_1 = ((\nu_{H_a} - \nu_{H_d} + \tfrac{1}{2}(J_{H_aC} - J_{H_dC}))^2 + J_{H_aH_d}{}^2)^{1/2} \qquad (4)$$

Application of pulses [*B*] selectively reverses ^{13}C and distant ^{1}H spin, while the attached one is apparently not affected and precession frequency ν_2 becomes during the second half of the time $t_1/2$

$$\nu_2 = \tfrac{1}{2}(\nu_{H_a} + \nu_{H_d} - J_{H_aH_d} - \tfrac{1}{2}(J_{H_aC} + J_{H_dC}) + R_2) \qquad (5)$$

where

$$R_2 = ((\nu_{H_a} - \nu_{H_d} - \tfrac{1}{2}(J_{H_aC} - J_{H_dC}))^2 + J_{H_aH_d}{}^2)^{1/2} \qquad (6)$$

The resulting evolution is described by a frequency

$$\langle\nu\rangle = \tfrac{1}{2}(\nu_1 + \nu_2) \qquad (7)$$

which averages contribution from the first and second half of the evolution time. By substituting eq 3 and 5 into eq 7

$$\langle\nu\rangle = \tfrac{1}{2}(\nu_{H_a} + \nu_{H_d}) + \tfrac{1}{4}(R_1 + R_2) \qquad (8)$$

it becomes obvious that scalar spin–spin couplings affect net precession only via R_1 and R_2. Since $^1J_{H_aC}$ is supposed to be significantly larger than $J_{H_aH_d}$ and J_{H_dC}, expressions 4 and 6 are approximated by the first two terms in their series thus leading to

$$\langle\nu\rangle \approx \nu_{H_a} + (\nu_{H_a} - \nu_{H_d})J_{H_aH_d}{}^2/(4(\nu_{H_a} - \nu_{H_d})^2 - (J_{H_aC} - J_{H_dC})^2) \qquad (9)$$

The result clearly indicates that the indirectly measured frequency $\langle\nu\rangle$ slightly deviates from the chemical shift frequency ν_H. The correction is proportional to $J_{H_aH_d}{}^2$ and inversely proportional to the dominant coupling $J_{H_aC}{}^2$, therefore, the accuracy of the chemical shift is not seriously hampered.

Strong coupling between protons does not represent an obstacle against the routine use of pulse sequence [*A*], because presence of the ^{13}C spin efficiently reduces inaccuracy of results. On the other hand, a more severe problem can be faced when protons are weakly coupled, but accidentally meet the following condition

$$|\nu_{H_a} - \nu_{H_d}| \approx \tfrac{1}{2}(J_{H_aC} - J_{H_dC}) \qquad (10)$$

In this case the typical correction is of the order of $^1/_4J_{H_aH_d}$, and it may be significant in some unfavorable samples. Fortunately, the indirect results can be compared with the direct homonuclear 2D experiment which represents an excellent complement by detecting ^{1}H signals of nonlabeled molecules.

It should be noted that apparent strong coupling can influence also selective reversal of distant protons, but these effects are not very important. One component of proton magnetization will still contribute to the expected real peaks, while the "spurious" components will not refocus precession resulting from *J* couplings and the intensity will be distributed among a very large number of

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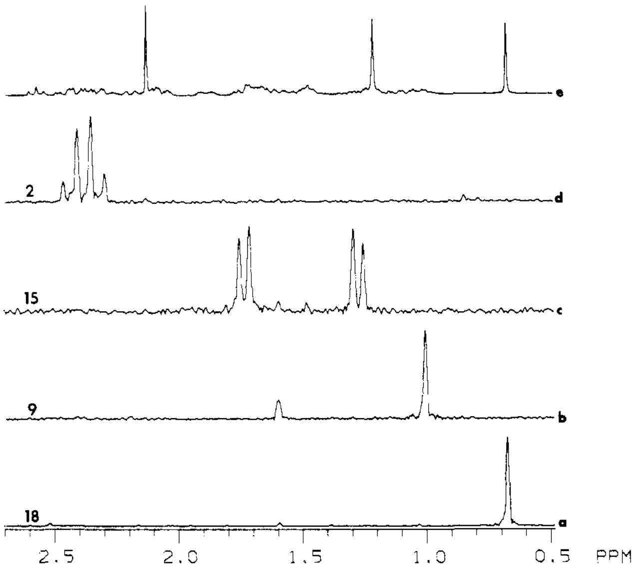

Figure 3. Comparison between the one-dimensional ^1H spectrum (upper trace) and typical cross section along the F_1 dimension in progesterone (**1**). The proton irradiation frequency was set at 1.6 ppm. Note that the peak overlap does not occur when ^1H resonances are detected via pulse sequence [*A*].

resonances. Therefore, pulse sequence [*A*] will give reliable values of ^1H chemical shifts and geminal couplings, although the signals may be severely reduced.

Finally it should be pointed out that replacement of the abundant ^{12}C nucleus with the magnetically active ^{13}C isotope changes resonance frequencies of attached protons[19] and chemical shifts are determined with uncertainty of a few ppb.

Study of Progesterone

One-dimensional ^1H spectra of progesterone (pregna-4-ene-3,20-dione) (**1**) and its derivatives are very complicated. Resonances of about 20 protons are distributed mostly in a narrow range between 0.5 and 2.7 ppm downfield from internal Me_4Si. Except for the methyl and olefinic protons, the ^1H spectra of these steroids have not been assigned and their use in determining the molecular structure and effects of substitution has been limited to monitoring the chemical shifts of the methyl groups.[20]

Recently Hall and Sanders[21,22] and Barrett et al.[23] used a combination of ^1H 2D *J* resolved spectroscopy and NOE difference measurements to assign spectra of a steroid 1-dehydrotestosterone[21] and two substituted progesterones: 11β-HO-**2**[22] and 17α-AcO-6α-Me-**3**.[23] The ^1H chemical shifts, *J* couplings, and NOE differences were then utilized to determine the conformations of these steroids in the solution. However, for molecules with this degree of complexity, even the "proton-decoupled" proton spectrum as obtained at a higher frequency (400 MHz) may suffer from overlapping of peaks thus causing uncertainties in the assignment. Progesterone itself represents a more challenging spin system, because the spectrum is not simplified by substitutions as in the case of **2** and **3**.

Since the ^{13}C signals of these molecules are relatively easy to interpret,[24] we decided to generate ^1H–^{13}C correlation maps by pulse sequence [*A*]. Cross sections along the assigned ^{13}C chemical shifts have been facilitated for studies of ^1H resonances, and Figure 3 shows some characteristic examples.

Table III. The ^{13}C and ^1H Chemical Shifts (in ppm) and the ^1H Geminal Coupling (in Hz) for Progesterone in $CDCl_3$

	$\delta(^{13}C)^a$	$\delta(^1H)^a$			$^2J_{HH}{}^b$
		α		β	
1	35.68	1.716		2.047	13.6
2	33.91	2.337		2.441	17.7
3	199.30				
4	123.87		5.730		
5	170.87				
6	32.75	2.280		2.412	14.8
7	31.85	1.065		1.870	12.7
8	35.50		1.570		
9	53.59		0.989		
10	38.53				
11	20.98	1.645		1.461	13.7
12	38.60	1.456		2.079	12.4
13	43.87				
14	55.97		1.181		
15	24.33	1.724		1.267	11.8
16	22.78	1.676		2.189	13.8
17	63.43	2.548			
18	13.30		0.669		
19	17.35		1.196		
20	209.18				
21	31.47		2.127		

[a] Estimated standard deviation 0.005 ppm. [b] Estimated standard deviation 0.5 Hz.

CH_3 groups in progesterone are not coupled to other protons, and they give rise to sharp single peaks in any version of the heteronuclear correlation spectroscopy. A cross section along the F_1 dimension (Figure 3a) reveals an intense "real" resonance at $\delta_H = 0.669$ ppm for the protons attached to the methyl carbon C-18. Spurious peaks at the middle are very close to the ^1H carrier frequency, and they can be attributed to electronic instabilities, while two small signals, which appear symmetrically at $\pm^1J_{CH}/2 = 62.9$ Hz, evidently arise from missetting of the delay τ in [*B*]. Protons in CH groups experience several *J* couplings which are selectively suppressed by pulse sequence [*A*], and therefore sharp single peaks are detected again (Figure 3b). It is also obvious that obstacles mentioned in the description of the method have decreased the signal-to-noise ratio.

All protons in CH_2 groups are nonequivalent, and the cross sections reveal two pairs of peaks which facilitate measurements of ^1H chemical shifts and geminal couplings $^2J_{HH}$ (Figure 3c). The last cross section (Figure 3d) shows the pattern for protons attached to the carbon C-2. Since the chemical-shift difference is comparable to the geminal coupling, intensity of the inner signals has increased thus resembling the typical strongly coupled quartet (i.e., AB spin system in one-dimensional NMR).

Experimental results are summarized in Table III. It should be mentioned that the method provides accurate values of ^1H chemical shifts and their correlation with ^{13}C, while assignment of resonances of two nonequivalent protons in CH_2 groups must be based on other techniques or studies of related compounds. In the present case the results were compared with assigned spectra of **2** and **3** where the general framework was established by NOE difference spectroscopy.[22,23]

The results for progesterone (**1**) have been compared with previous studies of **2** and **3** in the same solvent. Taking into account substitution effects, which can be readily discerned, a good agreement has been found, but there are also some significant discrepancies. The most notable one is the assignment of proton H-14 in **3** to 1.65 ppm along with H-15α and H-16α.[23] Our study clearly shows that the chemical shift of H-14 in **1** is 1.181 ppm (Table III), while for **2** the value 1.11 ppm was reported.[23] Although the remote possibility of substitution effects should not be ignored completely, we attribute the descrepancy to missassignment in the previous study[23] which utilized "proton-decoupled" spectrum with several overlapping peaks. If the assignment of

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H-14 is corrected to 1.181 ppm in **3**, the resonance would be buried under the strong signal of the methyl H-19 in the "proton-decoupled" spectrum. The other less obvious discrepancy between **1** and **3** occurs at H-8. The difference of 0.12 ppm at a site which is relatively unaffected by substitution may again be due to the uncertainty in the assignment of overlapping peaks in **3**.

These examples illustrate the advantage of the indirect detection by pulse sequence [*A*] which leads to correlation maps and cross sections without any overlap of ^1H peaks. Moreover, in some cases the method can be utilized to ascertain ^{13}C resonances. In **1** and related compounds the assignment of C-15 and C-16 (at approximately 22 and 24 ppm, respectively) is not unanimous,[24a,d,26] but in our study H-15 and H-16 can be distinguished clearly on the basis of NOE difference measurements[22,23] in **2** and **3**. Further confirmation is derived by observing the effect of OAc substitution at position 17 which changes the geminal coupling $^2J_{HH}$ of protons H-15 slightly from 11.8 Hz in **1** to 11.3 Hz in **3**. For protons H-16 the substitution *increases* $^2J_{HH}$ from 13.8 Hz in **1** to 16.1 Hz in **3**. Thus the correlation between the ^1H and ^{13}C chemical shifts facilitates the conclusion that C-15 and C-16 assignment of Highet et al.[24d] is the correct one.

Experimental Section

Progesterone (**1**) was dissolved in $CDCl_3$ to obtain a 0.6 M solution which was put into a 5-mm tube for one-dimensional ^1H and into a 20-mm tube for ^{13}C measurements. The Nicolet NT-300 spectrometer was operating at 300 MHz for protons and at 75.45 MHz for ^{13}C NMR. The routine one-dimensional spectra provided all ^{13}C as well as methyl proton chemical shifts which are reported with respect to internal Me_4Si. Multiplicity of carbon signals was verified by DEPT pulse sequence.[25]

Pulse sequence [*A*] generated 512 blocks each containing 2048 data points with the spectral width ±2976 Hz. The fixed delays were optimized for CH_2 groups ($\tau = 3.8$ ms and $\tau' = 1.9$ ms). The evolution time t_1 was incremented in steps of 1.42 ms. An exponential window function (line broadening 1.0 Hz) apodized ^{13}C signals, and subsequent Fourier transformation separated characteristic resonances along the F_2 dimension. To avoid limitations imposed by a finite size of the computer memory only 8 data points around each ^{13}C signal were transposed. Along the F_1 dimension sinusoidal apodization eliminated broad "tails" around the peaks and zero-filling up to 8192 data points improved digital resolution. The spectra were displayed in the magnitude mode, and their projection onto the F_1 axis was considered as a cross section. This procedure appears more reliable for accurate chemical-shift measurements than the common searching for the maximum signal.

The ^1H chemical shifts were calibrated with respect to the C-18 methyl signal at 0.699 ppm, and the accuracy is approximately 0.005 ppm. The limitations are imposed mostly by inhomogeneity of the external magnetic field, strong coupling, and $^{12}C/^{13}C$ isotope shifts.

Cyclohexanol was studied in a similar way. The only important difference is the fact that traditional cross sections are plotted in a phase-sensitive mode (Figure 2) to show realistic line shapes.

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Special Effects of an Unusually Large Neutral to Radical Cation Geometry Change. Adiabatic Ionization Energies and Proton Affinities of Alkylhydrazines

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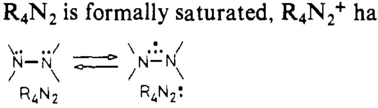
Abstract: High-pressure mass spectrometry was used to measure adiabatic ionization potentials and proton affinities for hydrazine, methylhydrazine, 1,1-dimethylhydrazine, and six tetraalkylhydrazines. Unusually large relaxation energies in the radical ions, which have vIP − aIP = 1.4–1.6 eV when only H and normal alkyl substituents are present, compared to 0.7 eV for typical amines, reflect the stabilization of the nearly planar hydrazine radical cations by charge delocalization in their three-electron π bonds. The proton affinities of hydrazines are lower than those of comparable amines by 4–6 kcal mol^{-1}. The combined effects of lowered aIPs and lowered PAs decrease the B^+–H bond-dissociation energies in protonated alkylhydrazines by ca. 20 kcal mol^{-1} compared to those of alkylamines.

Previous work on the tetraalkylhydrazine, tetraalkylhydrazine radical cation electron transfer reaction (eq 1) has been recently reviewed.[1] Although R_4N_2 is formally saturated, $R_4N_2^+$ has its



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positive charge delocalized over both nitrogens and is substantially stabilized compared to radical cations of most saturated compounds. Long lifetimes are observed for $R_4N_2^+$ in solution, allowing measurement of the thermodynamically significant formal potential for electron transfer, $E^{\circ\prime}$, for a wide variety of alkyl substituents. Unusual characteristics of this electron transfer include a lack of correlation between vertical ionization potential (vIP) and the electrochemical formal potential $E^{\circ\prime}$, a large dependence of the rate of electron loss upon the lone pair–lone pair dihedral angle θ, and very low rates for $R_4N_2,R_4N_2^+$ electron exchange. Those special characteristics result from an unusually

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