teraction. In addition, $|3\cos^2\theta - 1| \simeq 2$ above the plane and only 1 in the plane. Thus the total expected pseudocontact effect will be much larger above the plane of the π system.

This interpretation makes clear predictions on the geometrical dependence of radical-induced shifts which we hope to test in porphyrins and other radicals. Coupling constant and chemical shift evidence show that the bridge in **1** is flexible, but a more rigid bridge would allow measurement of θ, r, and hence g-tensor anisotropy.

These results reveal the possibility that, in isolated photosynthetic reaction centers, chlorophyll-protein complexes, or chlorophyll-doped membranes, the chlorophyll radical cation could act as a natural in situ spin label causing shifts as well as broadening. Other biologically important radical cations and anions presumably have the same potential, although in all cases the long electron T_1 may be a problem.

Acknowledgments. We thank M. D. Turnbull and A. R. Battersby for a generous supply of the bridged porphyrin and S.R.C. for financial support.

References and Notes

(1) E. de Boer and H. Van Willigen, *Prog. Nucl. Magn. Reson. Spectrosc.*, **2**, 111 (1967).
(2) H. J. Keller and K. E. Schwartzhans, *Angew. Chem. Int. Ed. Engl.*, **9**, 196 (1970).
(3) R. W. Kreilick, *Adv. Magn. Reson.*, **6**, 141 (1973).
(4) J. Reuben, *Prog. Nucl. Magn. Reson. Spectrosc.*, **9**, 1 (1973).
(5) A very small effect has been reported: I. Morishima, K. Ishihara, K. Tomishima, T. Inubishi, and T. Yonezawa, *J. Am. Chem. Soc.*, **97**, 2749 (1975).
(6) A. R. Battersby, D. G. Buckley, S. G. Hartley, and M. D. Turnbull, *J. Chem. Soc., Chem. Commun.* 879 (1976).
(7) F. A. Bell, A. Ledwith, and D. C. Sherrington, *J. Chem. Soc. C*, 2719 (1969).
(8) (a) J. K. M. Sanders and J. C. Waterton, *J. Chem. Soc., Chem. Commun.*, 247 (1976); (b) J. K. M. Sanders and I. Baxter, *Tetrahedron Lett.*, 4543 (1974).
(9) E. de Boer and C. Maclean, *J. Chem. Phys.*, **44**, 1334 (1966).
(10) The relative importance of ring current and local anisotropy in porphyrins is not clear. See A. Agarwal, J. A. Barnes, J. L. Fletcher, M. J. McGlinchey, and B. G. Sayer, *Can. J. Chem.*, **55**, 2575 (1977).
(11) The radical cation is a 17- or 19-π-electron system which may be nonaromatic and have no ring current. This effect could account for only a small part of the observed shift.
(12) Since some radical decomposition occurs between generation and spectral acquisition, the quoted shift is a lower limit.
(13) J. H. Furhop, *Angew. Chem. Int. Ed. Engl.*, **13**, 321 (1974).
(14) Similarly, with lanthanides, ^{17}O NMR of the aquo ions implied that pseudocontact effects would be unimportantl W. B. Lewis, J. A. Jackson, and H. Taube, *J. Chem. Phys.*, **36**, 692 (1962).

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High Resolution ^{31}P and ^{13}C Nuclear Magnetic Resonance Spectra of Unsonicated Model Membranes

Sir:

Nuclear magnetic resonance has been used extensively in the last decade to study the structure and dynamics of model and biological membranes.[1] However, the complexity of these systems, which should manifest itself in a corresponding richness of their NMR spectra, has in most cases not been observed because of the substantial breadth of the NMR lines. It is now understood that this breadth is due primarily to residual chemical shift anisotropy and dipole-dipole interactions. For dilute spins, such as ^{13}C and ^{31}P, the dipolar broadening can be removed by sufficiently intense rf irradiation at the proton resonance frequency.[2-4] Nevertheless, a substantial broadening due to the anisotropy of the chemical shift remains. In order to obtain "high resolution" NMR spectra, it has become customary to subject multilamellar dispersions to prolonged ultrasonic irradiation.[5] This process, which results in particles of reduced size with reduced reorientational correlation times, does indeed improve the resolution of the NMR spectra; however, its exact physical and chemical consequences are a subject of much debate.[6] We report an approach which avoids sonication.

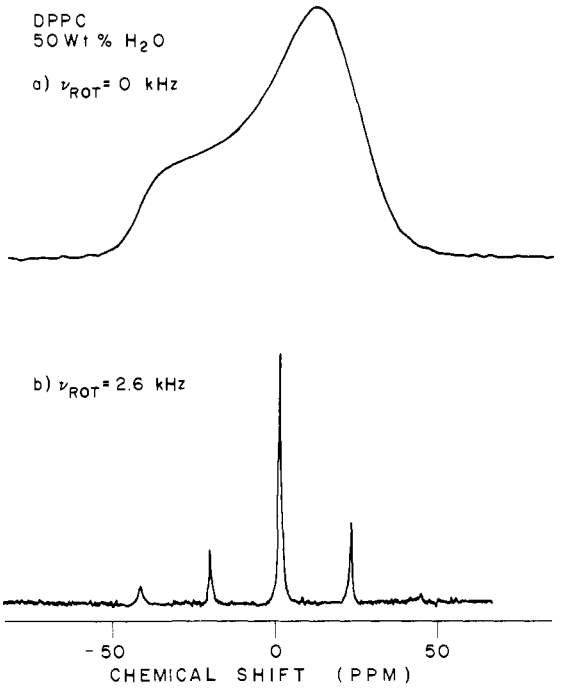

Figure 1. Proton decoupled ^{31}P spectra of DPPC in 50 wt % H_2O (T = 21 °C): (a) axially symmetric powder spectrum, $\Delta\sigma = -60$ ppm (7.2 kHz), $\nu_{rot} = 0$; (b) $\nu_{rot} = 2.6$ kHz. In (b) the side bands are spaced at the spinning frequency and the full width of the centerband is 95 Hz. Shifts are referenced to external 85% H_3PO_4.

Almost 2 decades ago Lowe[7] and Andrew et al.[8] suggested that rapid spinning of a sample about an axis inclined at the "magic angle", 54° 44′, with respect to H_0 would suffice to narrow dipolar broadened NMR lines. The criterion to achieve complete narrowing is that the rotation rate, ν_R, must be greater than the dipolar line width.[9] For this reason ^1H spinning experiments on phospholipids have not been very successful.[10] With the development of pulse methods for obtaining high resolution NMR spectra in solids,[11-14] there has been renewed interest in this technique because chemical shift tensors, like dipolar tensors, are of rank two, and can be averaged to their trace by magic angle spinning. Recently Schaefer and Stejskal [15]demonstrated that high resolution ^{13}C spectra of several polymeric samples could be obtained by combining dilute spin double resonance and sample spinning.[16] However, it was thought that ν_R must be greater than the breadth of the shift powder spectrum, $\Delta\sigma$, in order to achieve substantial narrowing. Thus, it appeared that this approach would be of utility only at relatively low magnetic fields—i.e., <2.0 T.

Recently we and others[17-19] have observed that even at $\nu_R < \Delta\sigma$ one still obtains a narrow centerband.[18] However, this line is accompanied by sidebands spaced at intervals equal to the spinning frequency, as is illustrated by the ^{31}P spectra of dipalmitoylphosphatidylcholine (DPPC), dispersed in excess water, shown in Figure 1. Here motional averaging narrows the rigid lattice axially asymmetric ^{31}P spectrum of ~190-ppm breadth[4] to an axially symmetric one of ~60 ppm,[3] which amounts to 7.2 kHz at our ^{31}P frequency of 119.05 MHz. Upon spinning this sample in an Andrew[20]-type rotor at 2.6 kHz, we observe a single line of 95-Hz full width at half-height flanked by two sets of sidebands. The residual line width is essentially

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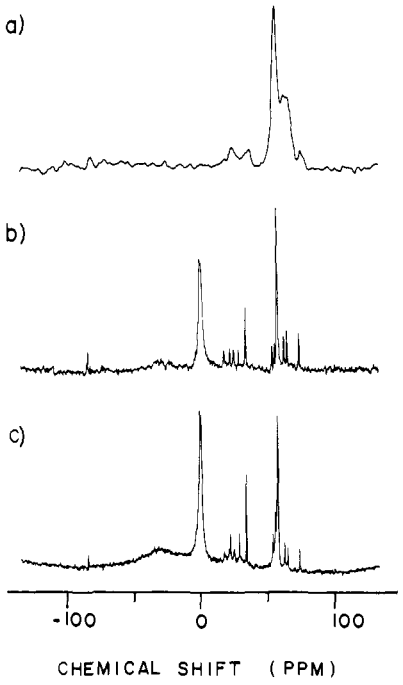

Figure 2. Proton decoupled ^{13}C spectra of DMPC and DPPC in 50 wt % H_2O (T = 21 °C): (a) DMPC, ν_{rot} = 0; (b) DMPC, ν_{rot} = 2.6 kHz; (c) DPPC, ν_{rot} = 2.6 kHz. Line assignments are based on those given in ref 23 and 26 and shifts are referenced to external delrin. Full widths of the single carbon resonances are ~20 Hz. The broad line at −30 ppm also arises from the delrin rotor. $\nu_{^{13}C}$ = 73.966 MHz.

Table I. ^{13}C Chemical Shifts for Unsonicated DMPC and DPPC in Excess Water at $T \sim 21$ °C with Respect to External Delrin Reference[a]

Carbon position	Chemical shifts, ppm DMPC	DPPC
Acyl chain		
C-1 (C=O)	−84.9	−84.6
C-2	54.4	54.7
C-3	64.0	63.7
C-4-C-11 (13)	58.0	58.3
C-12 (14)	56.1	56.6
C-13 (15)	65.6	66.0
C-14 (16)	74.5	75.0
Glycerol		
C-1 (CH_2OP)	25.2	25.4
C-3 (CH_2OCOR)	25.2	21.5
C-2 (CHOCOR)	17.7	17.9
Choline		
CH_2OP	28.8	29.3
CH_2N	22.3	22.7
$N(CH_3)_3$	34.3	34.6

[a] In the acyl chains C=O and $-CH_3$ are labeled C-1 and C-16, respectively, and in the glycerol C-1 and C-3 are esterified with the acyl chain and PO_4, respectively. The numbers in parentheses refer to the carbon position in the DPPC acyl chains.

identical with that observed in sonicated DPPC vesicles at roughly an equivalent magnetic field and it may be determined by shift anisotropy relaxation effects.[21]

The effect of sample spinning is more dramatically illustrated by the proton-decoupled ^{13}C spectra of dimyristoylphosphatidylcholine (DMPC) and DPPC, dispersed in excess water, displayed in Figure 2. Figure 2a shows a spectrum obtained with a stationary sample of DMPC and again the rigid lattice ^{13}C shift tensors are partially averaged to the point that assignment of bands to the carbonyl, the $-N(CH_3)_3$, glycerol backbone, head group $-CH_2-$ and the acyl chain carbons can be accomplished, but there remains a considerable amount of shift anisotropy broadening. Upon spinning, the spectral bands narrow dramatically as is shown in Figure 2b and, at a spinning rate of 2.6 kHz, we resolve all of the lines that are observed when the lipid is dissolved in $CHCl_3$.[22,23] The assignment of the lines is given in Table I referenced to the strong line in the center of our spectrum which arises from the ^{13}C nuclei in our plastic (Delrin) rotor. Since we have observed the broad line spectra of Figure 2a both prior to and following spinning, it appears that sample rotation at 2.5 kHz does not degrade the structure of the liposomes.

Based on a measurement of the temperature of the gas employed to drive the spinner, we believe the sample temperature is close to 21 °C. For DMPC the pretransition temperature, T_p, is 14 °C and the gel-liquid crystalline transition temperature, T_c, is 24 °C; so we believe the lipid is in its monoclinic phase.[24,25] However, if there were a slight amount of heating due to ^1H decoupling, then the spectrum of Figure 2b could be due to the liquid crystalline phase. In order to obtain a gel phase spectrum, we have studied DPPC which has T_p = 35 °C and T_c = 41 °C and its spectrum is shown in Figure 2c; the chemical shifts are also given in Table I. A comparison of Figures 2b and 2c shows that the acyl chain and carbonyl regions of the spectra are rather similar, but the glycerol and choline regions are somewhat different. Specifically, in the DPPC spectrum the α- and β-choline carbon lines sharpen slightly and all three glycerol carbon lines are now resolved. On the basis of DPPC spectra obtained in methanol,[26] we assign the glycerol lines, in order of decreasing shielding, to C-1, C-3, and C-2, respectively. The fact that the three glycerol lines are resolved in DPPC, but not in DMPC, suggests the possibility of slightly different conformations in the glycerol segment below and above T_c.

The ^{13}C spectra in Figures 2b and 2c do not exhibit rotational sidebands, in spite of the fact that they were obtained at a 6-8-T high magnetic field. This can be understood by noting that the apparently large breadths of the powder spectra of Figure 2a are due in part to overlap of residual shift anisotropy patterns. In fact the powder spectra from individual carbons are probably ~1 kHz wide so that we are in the limit where the narrowing is complete. This also suggests that ^{13}C lipid spectra which are free of rotational sidebands could be obtained at two-three times higher magnetic fields with currently available rotor designs. On the other hand, information on the residual shift anisotropy should be available by decreasing the spinning rate to the point where sidebands appear.[17] This is an appealing approach to studying these parameters because the lines will be sharp and, consequently, the problem of signal overlap can be partially circumvented.

Acknowledgments. Special thanks are accorded to J. Chandnoit and P. Sassone for their very patient assistance in the construction of the probe and spinners used in these experiments. This research was supported by the National Science Foundation, through Contract C-670 to the FBNML, and by the National Institutes of Health through Grants GM-23316 and GM-23289.

References and Notes

(1) For a review of this field see A. G. Lee, N. J. M. Birdsall, and J. C. Metcalfe, "Methods of Membrane Biology", Vol. II. E. D. Korn, Ed., Plenum Press, New York, N.Y., 1974, pp 1-156.
(2) J. Urbina and J. S. Waugh, *Proc. Natl. Acad. Sci. U.S.A.*, 71, 5062-5067 (1974).
(3) R. G. Griffin, *J. Am. Chem. Soc.*, **98**, 851-853 (1976).
(4) S. J. Kohler and M. P. Klein, *Biochemistry*, **15**, 967-973 (1976); J. Herzfeld, R. G. Griffin, and R. A. Haberkorn, *ibid.*, in press.
(5) C. H. Huang, *Biochemistry*, **8**, 344-352 (1969).
(6) For recent references, see for instance N. O. Petersen and S. I. Chan, *Biochemistry*, **16**, 2657 (1977); G. W. Stockton, C. F. Polnaszek, A. P. Tulloch, F. Horsan, and I. C. P. Smith, *ibid.*, **15**, 954 (1976).
(7) I. J. Lowe, *Phys. Rev. Lett.*, **2**, 285-287 (1959).
(8) E. R. Andrew, A. Bradbury, and R. G. Eades, *Nature (London)*, **182**, 1659 (1958).

(9) U. Haeberlen and J. S. Waugh, *Phys. Rev.*, **185**, 420–424 (1969).
(10) D. Chapman, E. Oldfield, D. Doskocilova, and B. Schneider, *FEBS Lett.*, **25**, 261–264 (1972).
(11) U. Haeberlen, "High Resolution NMR in Solids", Academic Press, New York, N.Y., 1976.
(12) M. Mehring, "High Resolution NMR Spectroscopy in Solids", Springer-Verlag, New York, N.Y., 1976.
(13) J. S. Waugh, L. M. Huber, and U. Haeberlen, *Phys. Rev. Lett.*, **20**, 180–182 (1968).
(14) A. Pines, M. G. Gibby, and J. S. Waugh, *J. Chem. Phys.*, **59**, 569–590 (1973).
(15) J. Schaefer and E. O. Stejskal, *J. Am. Chem. Soc.*, **98**, 1031–1032 (1976).
(16) Combined multiple pulse and sampling spinning experiments have also been reported: B. C. Gerstein, R. G. Pembleton, R. C. Wilson, and L. M. Ryan, *J. Chem. Phys.*, **66**, 361–362 (1977).
(17) E. Lippmaa, M. Alla, and T. Tuherm, *Proc. Congr. Ampere, 19th, 1976*, 113–118 (1976).
(18) M. Maricq and J. S. Waugh, *Chem. Phys. Lett.*, **47**, 327–329 (1977).
(19) E. O. Stejskal, J. Schaefer, and R. A. MaKay, *J. Magn. Reson.*, **25**, 569–573 (1977).
(20) E. R. Andrew, L. F. Farnell, M. Firth, T. D. Gledhill, and I. Roberts, *J. Magn. Reson.*, 1, 27–34 (1964).
(21) J. A. Berden, P. R. Cullis, D. I. Hoult, A. C. McLaughlin, G. K. Radda, and R. E. Richards, *FEBS Lett.*, **46**, 55–58 (1974).
(22) The ^{13}C spectrum of DMPC in $CHCl_3$ is essentially identical with Figure 2b; so it is not included here.
(23) Y. K. Levine, N. J. M. Birdsell, A. G. Lee, and J. C. Metcalfe, *Biochemistry*, 11, 1416–1421 (1972); E. Oldfield and D. Chapman, *Biophys. Biochem. Res. Commun.*, **43**, 949–953, (1971).
(24) M. Janiak, D. M. Small, and G. G. Shipley, *Biochemistry*, **15**, 4575–4580 (1976).
(25) E. J. Luna and H. M. McConnell, *Biochem. Biophys. Acta*, **466**, 381–392 (1977).
(26) N. J. M. Birdsall, J. Feeney, A. G. Lee, Y. K. Levine, and J. C. Metcalfe, *J. Chem. Soc., Perkin Trans. 2*, 1441 (1972).
(27) Francis Bitter National Magnet Laboratory.
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Received September 22, 1977

Photocalorimetry. Enthalpies of Photolysis of *trans*-Azobenzene, Ferrioxalate and Cobaltioxalate Ions, Chromium Hexacarbonyl, and Dirhenium Decarbonyl

Sir:

We report here on a new type of calorimetry, *photocalorimetry*. The procedure determines the ordinary ΔH of a reaction, but is special in that the reaction is light induced. If light of flux $F°$ (defined here as J s^{-1}) is absorbed by a solution, then in the absence of photochemical (or thermal) reaction and of emission, the energy $F°t$ must appear as heat, where t is the time of irradiation in seconds. If, however, a photoinduced reaction occurs, the observed rate of heat production will be some different value, F, and the quantity $(F° - F)t$ gives the enthalpy change associated with the amount of reaction that has occurred. The ΔH of reaction may be calculated as $(F° - F)/n$, where n is the moles of such reaction per second. Alternatively, the fractional discrepancy due to the heat of reaction is $f = (F° - F)/F°$; if positive, f is the efficiency of conversion of light to chemical energy. Further, if monochromatic light is used, $\Delta H = fE/\phi$, where E is the energy per mole of light quanta, and ϕ, the quantum yield for the photoreaction. Note that if the experimental n is used in calculating ΔH, the result will be correct even if absorption by the cell window or by photoproducts occurs, although f and ϕ will be low.

Photocalorimetry suffers in precision because it depends on the difference $(F° - F)$ and is insensitive if ϕ is small. It can, however, be a convenient alternative to other methods of ΔH determination, and in some cases may be the only practical method. Certainly, current thermochemical complications[1-3] allow rather few enthalpies of reactions between organometallic compounds in solution to be assembled; the same is true for kinetically stable Werner-type coordination compounds. A further point is that there can be cases where the photochemical reaction is complex or not fully understood, and yet is one for which the enthalpy change is desired. Potential solar energy storage reactions may fall in this category.[4] The equipment described here is well suited to the testing of such systems since f is determined.

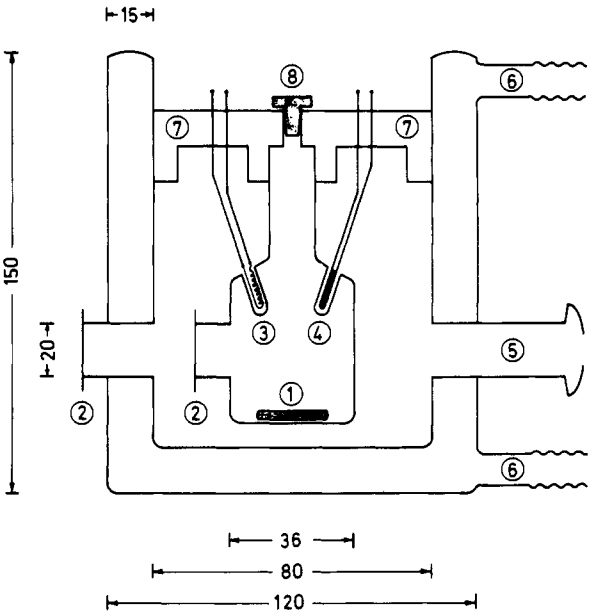

Figure 1. Photocalorimeter cell (dimensions in mm): (1) coated magnetic stir bar, (2) quartz windows, (3) heating coil, (4) thermistor, (5) to vacuum pump, (6) to thermostat, (7) metal cover, (8) cap to inner cell.

The essential features of our equipment and procedure are as follows. The photocalorimeter cell is shown in Figure 1. The inner cell is magnetically stirred and its temperature is sensed by a thermistor and associated bridge and chart recorder. A precise amount of electrically generated heat can be delivered by means of a resistance coil, using a constant current generator. The space around the cell is evacuated and the outer space thermostated. The cover is of heat-conducting metal, effectively to complete the constant temperature environment. Monochromatic light, controlled by a shutter, is used. Many of the features of the equipment are similar to those for the calorimetric determination of fluorescence yields.[5]

An experiment consisted of the following sequence. The cell was first filled with an absorbing but nonphotoactive reference solution, and the heat capacity, C, determined as joules per chart division (the chart readout being proportional to temperature). The rate of light heating, R, was determined by opening the shutter for the appropriate time and is reported in chart divisions per second. The product CR gives $F°$. The cell was then rinsed and filled with the solution of interest, C and R again determined, and thus F. While not in principle necessary, we made the practice of using as reference solution a similar, but nonphotoactive system in the same solvent. Irradiations were sufficiently limited to avoid significant secondary photolysis; none of our solutions emitted to any visually detectable extent; the data are for temperatures falling in the range 24.5–25.5 °C.

The results for the five reactions studied are summarized in Table I. Since the method was new, it was desirable to see if a known $\Delta H°$ could be confirmed. The reaction chosen is

$$\textit{trans}\text{-azobenzene} \xrightarrow{h\nu} \textit{cis}\text{-azobenzene} \qquad (1)$$

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