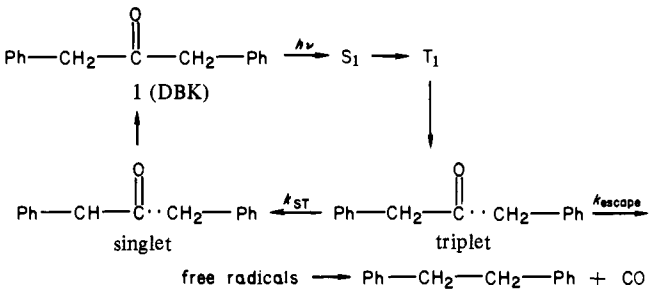
Scheme I. Transformations in Dibenzyl Ketone Photochemistry



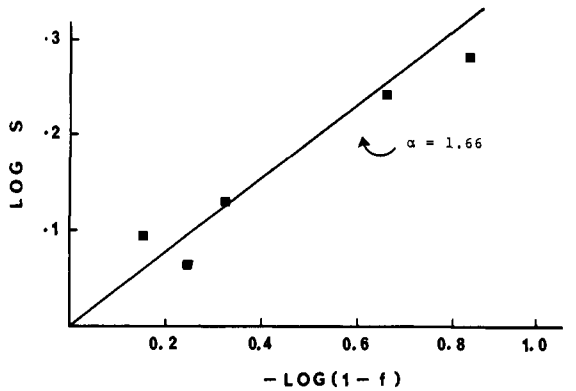


Figure 1. ^{13}C content of recovered dibenzyl ketone (DBK). Plot of separation factor, S, vs. fraction, f, of reacted DBK.

and farther apart. Since there is a great similarity in the physicochemical behavior of micelles, bilayers, vesicles, and adsorbed monolayers,[4a] we sought to probe whether a synthetic monolayer of a long-chain hydrocarbon, chemically bonded to the surface of a silica gel support, might produce a micelle "model" which could subsequently be altered systematically to find the optimum "position" for the "reflecting boundary" for highest isotopic enrichment. We prepared such a surface by the reaction of trichlorododecylsilane with silica gel,[5] giving rise to a surface which is a permanently bonded, rather than adsorbed, hydrocarbon monolayer. Adsorption of DBK onto this surface[7] followed by irradiation[8] gave rise to the same products observed in previous solution studies of DBK. The recovered DBK, as in Turro's reports, was found[9] to be enriched in ^{13}C! The results of a series of irradiations are shown in Figure 1. As Turro has noted, the use of α, the "single-stage separation factor",[10] is a convenient way to describe the efficiency of enrichment, where the larger the value of α, the larger the efficiency of ^{13}C enrichment. The value of α is obtained by noting that the slope of log S vs. -log $(1 - f)$ equals $(\alpha - 1)/\alpha$. Figure 1 leads to a value of 1.66 for α in our study, which may be compared with the value of 1.47 found[1a,c] in micelles[11] and the value of 1.03 for solution photolysis.[1a]

Thus, the synthetic monolayer surface seems to be quite analogous to a micelle in providing an artificial "reflecting boundary" which leads to an enhanced magnetic isotope effect over solution photolysis. We feel that it is unlikely that the dodecyl system and conditions reported here will be found to be the optimum environment for enrichment and hope that by varying the degree of crowding of the surface and/or length of the hydrocarbon chain, we will find significantly higher enrichment factors are possible. Work along these lines is currently in progress.

Acknowledgment. We are grateful to the University of Connecticut Research Foundation and the National Institutes of Health (AI12200) for financial support of this work. We are further grateful to Marvin Thompson and Ruven Smith for mass spectral determinations. E.F. thanks C.-J. Chung, Columbia University, and Ming Chow, IBM, Fishkill, NY, for helpful discussions.

(11) For simplicity the previous assumption[1a] that all enrichment occurred at one carbon was maintained in our work.

Quadrupolar ^{13}C–^{14}N Couplings and ^{14}N Relaxations in Aggregated and Nonaggregated Choline Phospholipids

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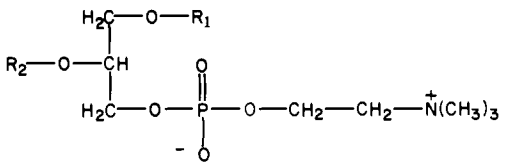
Quadrupolar relaxation (T_1) of nitrogen-14 (spin 1, positive quadrupole moment) is greatly dependent on the symmetry of substitution around the nitrogen nucleus,[1,2] and T_1, in turn, affects the resonance of directly bonded nuclei with a spin $^1/_2$.[3] Hence, ^{14}N T_1 and the quadrupolar ^{13}C–^{14}N couplings (J_{CN}) are sensitive to changes in the nitrogen environment. In the present study, we show that carbon–nitrogen couplings and quadrupolar ^{14}N relaxations of phosphatidylcholine (**1**; PC) are dependent on the



1, R_1 = acyl; R_2 = acyl
2, R_1 = acyl; R_2 = H
3, R_1 = R_2 = H

solvent, but that the magnitude of ^{14}N T_1 and J_{CN} is not so much the result of phospholipid–solvent interactions than it is a consequence of aggregation or, better, nonaggregation of the PC species in a given medium.

In the absence of quadrupolar relaxation, nuclei of spin $^1/_2$ coupled to nuclei of spin 1 produce simple multiplets.[4] Quadrupolar relaxation, however, modulates the line shape of the spin $^1/_2$ nucleus (e.g., ^{13}C) when random transition occurs between states 0 and ± 1 according to

$$\eta = 10\pi T_1 J_{CN} \quad (1)$$

with η being a dimensionless parameter. Pople[4] has previously calculated theoretical line shapes for conditions $\eta^2 = 10^n$ ($n = 0$,

(5) The "RP-silica gel" was prepared by using the procedure of Berendsen[6] with Merck Silica Gel 40, trichlorododecylsilane, and catalytic chlorotrimethylsilane to give a product which was found to be 11.17% carbon, indicating a very high surface coverage.

(6) (a) Berendsen, G. E.; Galan, L. *J. Liq. Chromatogr.* **1978**, *1*, 561. (b) Berendsen, G. E. "Preparation and Characterization of Well-Defined Chemically Bonded Stationary Phases for High Pressure Liquid Chromatography"; Delft University Press: Delft, Netherlands, 1980.

(7) Typically, a slurry was prepared of a solution of 1.0 g of DBK in ca. 100 mL of 3:1 pentane/ether and 10 g of the RP-silica gel. The solvent was removed in vacuo to give the dry, free-flowing solid which was used in the irradiations.

(8) Irradiating dibenzyl ketone with naturally abundant ^{13}C by using a 450-W medium-pressure Hanovia lamp, under a nitrogen atmosphere, through Pyrex.

(9) The DBK was isolated by high-performance-low-pressure LC on Merck silica gel 40, eluting with ether/pentane. The extent of enrichment was determined by GC/MS analysis.

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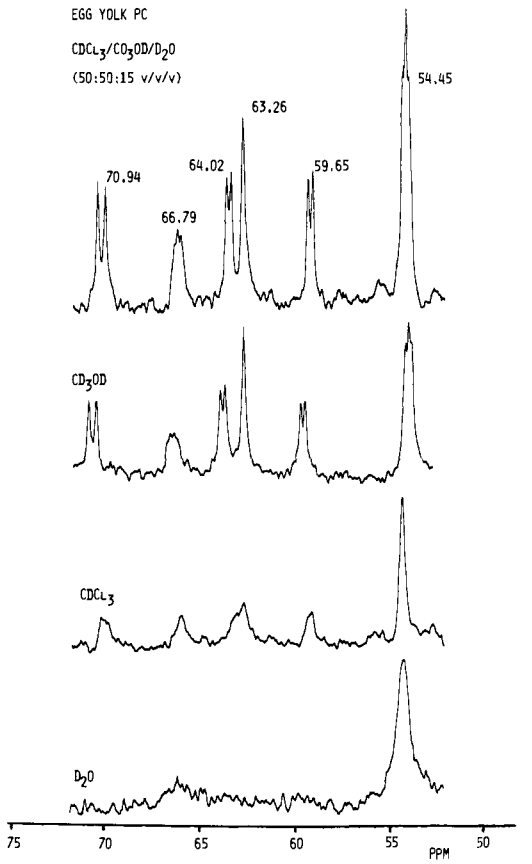

Figure 1. Proton-decoupled 20-MHz ^{13}C NMR spectra[7] of egg yolk phosphatidylcholine (**1**) in $CDCl_3/CD_3OD/D_2O$ (50:50:15, v/v/v),[8] CD_3OD, $CDCl_3$ (reverse micelles), and D_2O (liposomes). Chemical shifts (δ) downfield from internal Me_4Si are given for $N(CH_3)_3$ (54.45, J_{CN} = 3.6 Hz), P-O-CH_2 of choline (59.65, $^2J_{CP}$ = 4.8 Hz), *sn*-1-CH_2-O-R_1 (63.26), *sn*-3-CH_2-O-P (64.02, $^2J_{CP}$ = 4.8 Hz), CH_2-N (66.79, $^3J_{CP}$ = 7.0 Hz, J_{CN} = 3.6 Hz), and *sn*-2-CH-O-R_2 (70.94, $^3J_{CP}$ = 7.9 Hz).

1, 2, and 3) and predicted collapsed triplets resulting in singlets or broadened singlets for $n = 0$ and 1 and triplets for $n = 2$ and 3.

T_1 of ^{14}N, in turn, is given by

$$1/T_1 = \tfrac{3}{8}(e^2Qq/\hbar)^2\tau_q \qquad (2)$$

when the environment of ^{14}N is axially symmetrical;[3] $e^2Qq/\hbar$ is the quadrupolar coupling constant in solid state; τ_q, the correlation time, refers to reorientations which affect the direction of the field gradient. The contribution from quadrupolar relaxation to T_1 is rather large compared to that from dipole–dipole and spin–rotation processes. Because of the molecular symmetry around the nitrogen, ^{14}N quadrupolar relaxation of choline derivatives is not affected by methyl rotation or internal rotation about the CH_2–N bond but will be dominated by rotation around the CH_2–CH_2 linkage of the choline moiety,[5,6] thus affecting the orientation of the CH_2–N vector. Hence, nonaggregated phosphatidylcholines, for example, in which the motion of the choline CH_2–CH_2 bond is not restricted, should show longer ^{14}N T_1 values and better-defined ^{13}C–^{14}N couplings.

As we show in Figure 1, the ^{13}C NMR spectrum (50–75-ppm region) of phosphatidylcholine (**1**) from egg yolk is very much affected by the medium. Resolved ^{13}C–^{14}N couplings for the choline methyls are observed in CD_3OD and particularly in $CDCl_3/CD_3OD/D_2O$ (50:50:15, v/v/v).[8] Yet in $CDCl_3$ and D_2O,

Table I. ^{14}N Chemical Shifts and Relaxations of Cholines in Various Solvents[a]

choline derivative	solvent	ppm[b]	T_1[c]	T_2*[d]
phosphatidylcholine from egg yolk (1)	CD_3OD	−327.20	0.150	0.177
	CMW-*d*[e]	−327.50	0.136	0.121
	$CDCl_3$	−328.82		0.041
	D_2O		~0.060[f]	~0.0006[f]
lysophosphatidylcholine (2)	CD_3OD	−327.89	0.219	
	CMW-*d*[e]	−327.36	0.184	
	D_2O	−329.40		0.036
sn-glycero-3-phosphocholine (3)[g]	CMW-*d*[e]	−327.42	0.267	
	D_2O	−329.04	0.404	
choline phosphate[h]	D_2O	−329.19	0.686	
choline chloride[i]	CD_3OD	−327.18	1.974	
	CMW-*d*[e]	−327.01	1.068	
	D_2O	−328.90	2.835	

[a] 5.742-MHz ^{14}N NMR spectra were recorded at 35 ± 1 °C on samples that were not degassed; see also ref 7. [b] Chemical shifts (ppm) are relative to KNO_3/D_2O used as external standard. [c] ^{14}N T_1 values (±10%) were obtained using the (PD-180°-τ-90°)$_n$ pulse sequence and the Varian T_1 Calc and Analyzer Programs. [d] $T_2^* = 1/\pi\nu_{1/2}$. [e] $CDCl_3$:CD_3OD:D_2O (50:50:15, v/v/v) (CMW-*d*) served as solvent; see also ref 8. [f] T values at 35 °C were read from published data; see ref 14. [g] See ref 11. [h] Sigma Chemical Co., St. Louis, MO. [i] Aldrich Chemical Co., Milwaukee, WI.

in which PC is known to exist as reverse micelles or as liposomes,[9] respectively, the choline methyl triplets collapse. In order to determine whether this collapse would be due to interactions of the choline nitrogen with a specific solvent or whether it is caused by rotational restrictions of the choline CH_2–CH_2 bond due to aggregation, the line shape of the choline methyl signals as a function of solvent was further investigated. In $CDCl_3/CD_3OD/D_2O$ (CMW-*d*)[8] the amphiphilic egg yolk PC (**1**) and LysoPC (**2**) as well as the hydrophilic GlyceroPC (**3**) and choline chloride show well-resolved ^{13}C–^{14}N couplings of 3.6–3.8 Hz (see Figure 2). In contrast, only the water-soluble GlyceroPC and choline chloride produce well-resolved $N(CH_3)_3$ resonances in D_2O (J_{CN} = 3.7–3.9 Hz), whereas PC and LysoPC, which aggregate in water, show singlets only. Therefore, the collapse of the quadrupolar coupling cannot be the result of interactions with the solvent per se, but must be due to molecular aggregation. In fact, in reexamining the published ^{13}C NMR spectra of long-chain alkyltrimethylammonium bromides[12] in D_2O, we observed that in this case resolved $N(CH_3)_3$ triplets occured below the critical micelle concentration (CMC), but singlets occurred only above the CMC, which is consistent with the above concept developed for choline derivatives. Similarly, reverse micelles of PC in $CDCl_3$ also produce a choline $N(CH_3)_3$ singlet only (Figure 1). Observation of a PC $N(CH_3)_3$ triplet in methanol and collapse of the triplet in $CDCl_3$ had previously been noted by Birdsall et al.,[9] who suggested that this may be the result of shorter ^{14}N relaxations in the micellar structures. On the other hand, London et al.[6] demonstrated that PC vesicles in D_2O produce a triplet for N-$(CH_3)_3$ at and above 60 °C, but not below, and they attributed the disappearance of C–N splittings at lower temperature to either

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(7) Natural abundance ^{13}C (20 MHz) and ^{14}N (5.742 MHz) spectra were recorded on a Varian FT-80A pulse Fourier transform instrument at 37 and 35 ± 1 °C, respectively, using a 10-mm broadband probe.
(8) $CDCl_3/CD_3OD/D_2O$, 50:50:15, v/v/v, (CMW-*d*) readily dissolves phospholipids at concentrations (~100 mg/mL) that permit rapid recording of natural abundance ^{13}C NMR spectra. The well-resolved resonances consistently show large $^3J_{^{13}C-^{31}P}$ couplings of 7–8 Hz, indicating essentially unhindered trans orientation of both the headgroup and the backbone of PC with respect to the phosphorus (Murari, R.; Wedmid, Y.; Baumann, W. J., manuscript in preparation).
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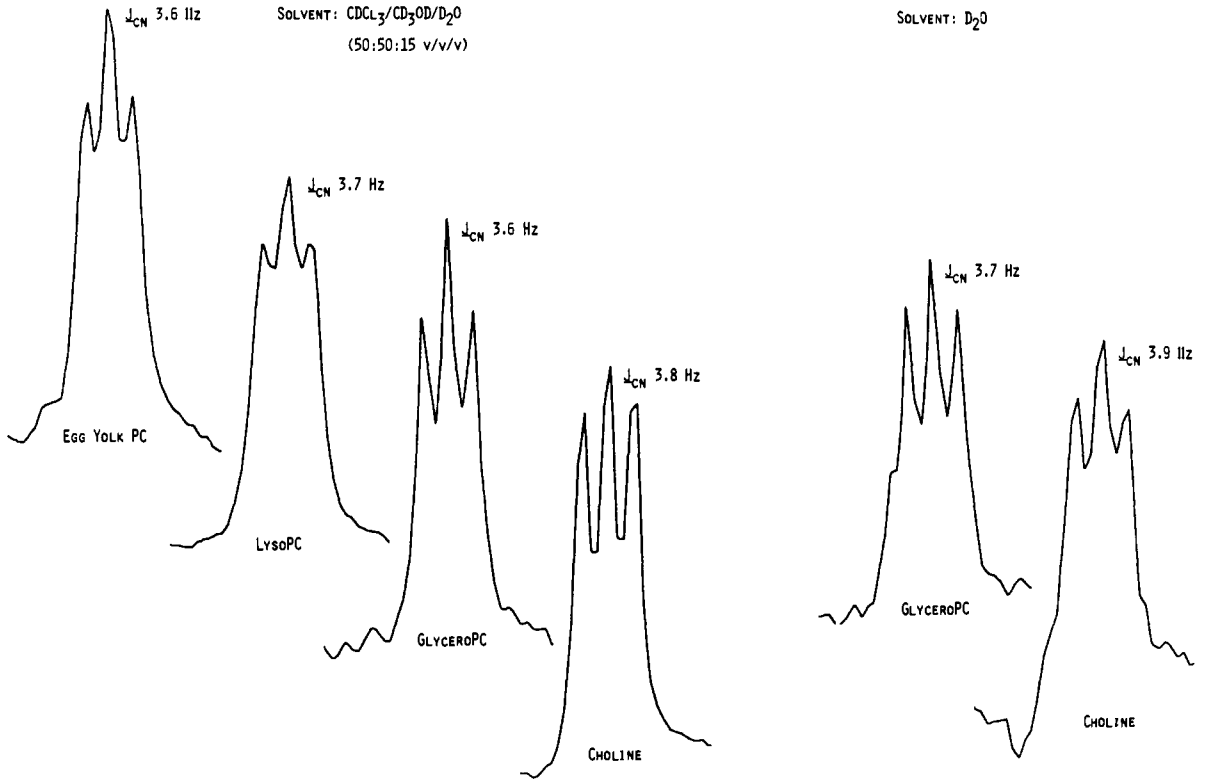

Figure 2. Line shape of $N(CH_3)_3$ signals ($J_{CN} \pm 0.1$ Hz) of choline derivatives in proton-decoupled 20-MHz ^{13}C NMR spectra[7] of egg yolk PC (**1**), LysoPC (**2**),[10] GlyceroPC (**3**),[11] and choline chloride in $CDCl_3/CD_3OD/D_2O$ (50:50:15, v/v/v)[8] and in D_2O.

dipolar broadening or a decrease in ^{14}N relaxation times.

In order to differentiate between the two effects, we measured ^{14}N T_1 and T_2* values of several choline derivatives in various solvents (Table I).[13] As one would expect, T_1 values of compounds in solution decreased with increasing molecular size in a given solvent, for example, in the series choline, GlyceroPC, LysoPC, PC in CMW-*d*[8] or similarly in CD_3OD. Further reduction in relaxation time, for example, for PC in D_2O (0.06 s) vs. CMW-*d* (0.136 s), can be attributed to liposome formation; reverse-micellar aggregation causes a similar reduction in relaxation times as is shown for PC in $CDCl_3$ (T_2* = 0.041 s)[13] vs. CMW-*d* (T_1 = 0.136 s) or CD_3OD (T_1 = 0.150 s) (Table I).

From the observed $^{13}C-^{14}N$ couplings of $N(CH_3)_3$ in solution, which average 3.7 Hz (Figure 2 and ref 15), and the limiting conditions $\eta^2 = 10^2$ at which broadening of the triplet begins,[4] T_1 of ^{14}N can be calculated according to eq 1. The calculations predict that triplet broadening would occur for T_1 values shorter than 0.086 s which would lead to the eventual collapse of the splittings. The ^{14}N relaxations for various choline derivatives (Table I) and the line shapes of the respective $N(CH_3)_3$ signals (Figures 1 and 2) are consistent with these calculations. This also demonstrates that nonobservation of C-N splittings can well be rationalized on the basis of a reduction in ^{14}N T_1 alone rather than by dipolar broadening.[16]

From our data we conclude that restrictions in mobility of the CH_2-CH_2 bond, that affect the CH_2-N vector, can be attributed to polar headgroup interactions. ^{14}N T_1 values and J_{CN} couplings are diagnostic of headgroup association and should prove useful in monitoring changes in the state of aggregation of choline phospholipids.

Acknowledgment. This study was supported in part by U.S. Public Health Service Research Grant HL 08214 from the Program Projects Branch, Extramural Programs, National Heart, Lung and Blood Institute, USPHS Research Grant NS 14304 from the National Institute of Neurological and Communicative Disorders and Stroke, and The Hormel Foundation. We are particularly grateful to Dr. S. Parthasarathy for preparing the choline derivatives used in this study.

(13) It has previously been noted that for reverse micelles of PC, T_2* is only slightly shorter than T_1, and hence either value can be used for approximating ^{14}N correlation times.[14]

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(15) $J_{^{13}C-^{14}N}$ values (±0.1 Hz) were for egg yolk PC (**1**), CD_3OD, 3.4 Hz, CMW-*d*,[8] 3.6 Hz; LysoPC (**2**), CMW-*d*, 3.7 Hz; choline chloride, CD_3OD, 3.9 Hz, CMW-*d*, 3.8 Hz, D_2O, 3.9 Hz.

(16) Observation of $N(CH_3)_3$ triplets in ^{13}C spectra of PC vesicles at elevated temperature[6] could be related to the increase in surface area per molecule with temperature[17] which would result in faster CH_2-CH_2 rotation and longer ^{14}N T_1 values due to decreased headgroup interactions. Koga and Kanazawa[14] also found that above 60 °C the T_1 values of PC vesicles were greater than the 0.086 s which we have calculated.

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NMR Investigation of a Surface Compound on Colloidal Silica

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Adsorption of inorganic and organometallic compounds on the surfaces of oxides is a process of fundamental importance in the preparation and operation of heterogeneous catalysts. Investigation of such systems by nuclear magnetic resonance is nontrivial, because restricted molecular motion in the solid state frequently leads to nonaveraged static interaction[1] although workers using Magic-angle spinning and multiple pulse techniques have made significant contributions to this area.[2-4] We wish to report a novel approach which uses NMR as a surface-sensitive technique in fluid

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