the second moment contributions of each particular nuclear species. Equations 2b and 2c are for natural abundance ^{25}Mg and eq 2d applies to 99% enriched ^{25}Mg. Because g anisotropy typically broadens lines asymmetrically, and even in spectra of fully deuterated algae,[15] which give very narrow Gaussian esr lines, the spectrum is highly symmetric, we assume g anisotropy is negligible here. With this premise, and using the ΔH_{pp} values of ^2H and ^1H Chl a$\cdot^+$ of 4.2 and 9.3 G,[15] respectively, solution of eq 1, 2a, 2b, and 2c yields a value of about 15% for the contribution to the line width from sources other than protons.

We expect an increase in line width for highly enriched ^{25}Mg Chl if the unpaired electron of the free radical interacts with Mg. An increase of 0.5 G in line width would be detectable. Since the line shape of Chl a$\cdot^+$ is Gaussian and we have only a single ^{25}Mg atom per unpaired spin, second moment theory can be applied rigorously to determine the minimum coupling constant observable for ^{25}Mg. By definition the second moment for a stick spectrum of ^{25}Mg (spin $^5/_2$) is

$$\langle \Delta H^2 \rangle_{^{25}Mg} = {}^{35}/_{12} A_{^{25}Mg}{}^2 \qquad (3)$$

where $A_{^{25}Mg}{}^2$ is the root-mean-square ^{25}Mg hyperfine coupling constant. We assume that the minimum line-width change that we could reliably detect would result in a 9.8-G line width, *i.e.*, $\langle \Delta H^2 \rangle_{total,\,^1H}$ is $^1/_4$(9.3 G)^2 and $\langle \Delta H^2 \rangle_{total,\,^2H,\,^{25}Mg}$ is $^1/_4$(9.8 G)^2. Thus, eq 1, 2c, 2d, and 3 indicate that an increase of 0.5 G in line width would be produced by a 0.9-G ^{25}Mg hyperfine coupling constant.

We have grown *Phormidium luridum* on a medium in which the magnesium (obtained from Oak Ridge National Laboratory) was 99% ^{25}Mg, according to the procedures of DaBoll, *et al.*[18] Chlorophyll was extracted by the method of Strain and Svec.[19] Preparation of chlorophyll samples for esr was performed as previously described.[15] Spectra were recorded on a Varian E9 spectrometer with 100-kHz modulation and a maximum modulation amplitude of one-third of the line width in a TE_{104} mode cavity equipped with a low-temperature quartz dewar insert. Microwave power was 2 mW. Calibration procedures were those of Norris, *et al.*[15] For increased precision, data were acquired with a Fabritek time-averaging device (Model 1072) on-line to the Argonne Chemistry Division central Sigma V computer.

Esr spectra of monomeric Chl a oxidized with I_2 in CH_3OH–CH_2Cl_2 (1:1) at both room temperature and $-160°$ consisted of single Gaussian lines, whether the chlorophyll contained ^{24}Mg or ^{25}Mg. At $-160°$, the line width for [^{25}Mg]Chl a was found to be 9.3 ± 0.5 G. [^{24}Mg]Chl a yields a line width of 9.3 ± 0.3 G at that temperature.[15]

Esr spectra of aggregated Chl a hydrates[20,21] containing ^{24}Mg or ^{25}Mg were also found to be indistinguishable. [^{25}Mg](Chl a$\cdot H_2O)_n$ oxidized by exposure to red light or chemically with I_2 has a line width of ~1.2 G (based on two determinations). [^{24}Mg](Chl a$\cdot H_2O)_n$ under the same conditions has the same line width for the photosignal.

The esr spectra of oxidized Chl a with natural abundance Mg and those of 99% [^{25}Mg]Chl a are thus indistinguishable. If significant spin density were in the magnesium 3s orbital, a difference in spectra from the two isotopic species should have been observed. Our data indicate that the ^{25}Mg coupling constant is less than 1 G; how much less is not known. The free ion coupling constant of ^{25}Mg(II) is 247.2 G.[22] Thus, our data indicate that the unpaired electron spin has little magnesium s character (less than 1/250). To relate this figure to another example of an unpaired electron interacting with ^{25}Mg, we note that the isotropic coupling constant in F centers with ^{25}Mg (4 G)[23] is consistent with an electron with very little s character (~1/60). Our data confirm the π nature of the Chl a free radical, and are thus consistent with conclusions arrived at in other recent studies on porphyrin π cations of biochemical significance.[24]

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Mary Ellen Druyan,[25] James R. Norris, Joseph J. Katz*
Chemistry Division, Argonne National Laboratory
Argonne, Illinois 60439
Received November 16, 1972

Nuclear Magnetic Resonance Studies of a Polypeptide in a Nonprotonating Solvent System

Sir:

Many polypeptides undergo a helix to coil transition as the solvent composition is varied, and trifluoroacetic or dichloroacetic acids (TFA, DCA) are the most commonly used helix breakers for inducing the transition. A strong solvation of the peptide groups by hydrogen bonding with acid in the coil form is the usual explanation offered for breakdown of the helix.[1] However, it has been proposed that the acid protonates the amide groups and the resulting electrostatic repulsions result in helix breakdown[2] and a coil form at least partially protonated. Nmr spectra in this journal and elsewhere[3] have been adduced as supporting protonation, although the spectral interpretations have been questioned.[4] The evidence opposing the idea of protonation is considerable.[1,5] However, none has been unequivocal and this communication presents nmr data that prove beyond doubt that protonation is not an essential step in helix breakdown.

It is now well established[6] that for low molecular

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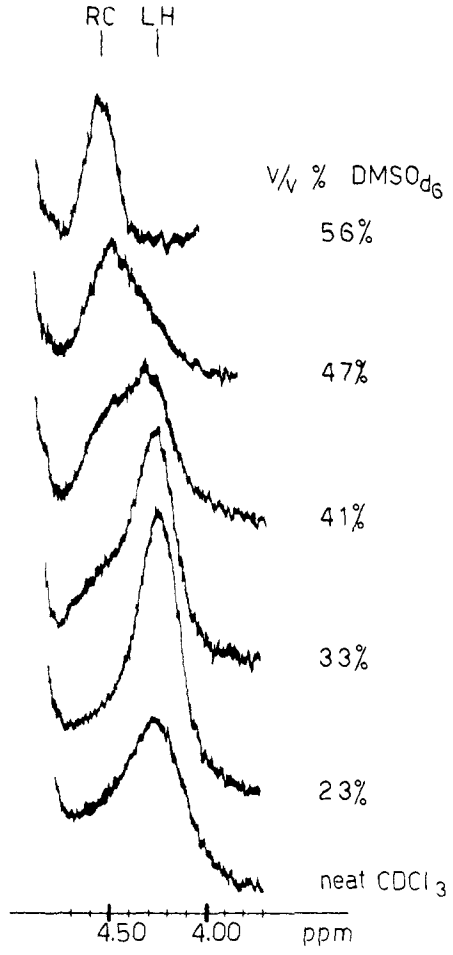

Figure 1. 100-MHz spectra, α-CH region, of poly-β-benzyl-L-aspartate in $CDCl_3$–Me_2SO-d_6; RC = random coil, LH = left-handed helix.

weight polypeptide samples the helix–coil transition is manifested in the nmr by the characteristic double-peak α-CH and amide NH resonances. If the two components of the main-chain doublets are well separated then in the case of the α-CH the up-field resonance may be assigned to helix and the low field to coil on the basis of constancy of total area[4] and a good correlation of peak areas with b_0.[7] The shift difference between the peaks $\Delta_{H/C}(\alpha\text{-}CH)$ is solvent dependent[8] and therefore represents the sum of a solvation effect (which might or might not be a result of protonation) and an intrinsic conformational effect.

Poly-β-benzyl-L-aspartate (PBLA) is random coil in dimethyl sulfoxide (Me_2SO)[9] and addition of chloroform induces transition to the left-handed (LH) helical form. The transition midpoint lies at about 42% Me_2SO-d_6. Figure 1 shows the PBLA α-CH spectrum at 100 MHz which is seen to exhibit the characteristic "double-peak" phenomenon. The LH helix shift is 4.30 ppm (with respect to internal TMS) as previously reported[10] and the coil shift in 56% Me_2SO-d_6 (and in pure Me_2SO-d_6) is 4.64 ppm. When TFA is used as the random coil inducing solvent, the coil shift in $CDCl_3$–5% TFA is 4.80 ppm.[8] This difference of 0.16 ppm in the coil shifts illustrates the importance of solvation in the α-CH helix–coil shift difference $\Delta_{H/C}(\alpha\text{-}CH)$.

Now Me_2SO-d_6, which in this experiment simply replaces TFA as the coil-inducing solvent, is not acidic and has no exchangeable protons. In the presence of fairly strong acids it even acts as a weak Lewis base. There can therefore be no protonation at all of the amide groups by Me_2SO-d_6 and it follows that protonation is not essential either for inducing the helix to coil transition or for the observation of different and characteristic helix and coil shifts for the main chain protons in the spectra of polypeptides.

PBLA is an atypical polypeptide in that it forms LH helices in chloroform. We have obtained similar results to the above using right-handed poly-γ-benzyl-L-glutamate (PBLG) samples of several molecular weights and also a number of PBLG–PBLA copolymers. The conclusions therefore appear general for this solvent system. An account of the complete series of studies will appear elsewhere.

Acknowledgment. E. M. Bradbury and C. Crane-Robinson are grateful to the CNR of Italy for fellowships during the tenure of which this work was carried out.

E. M. Bradbury,* C. Crane-Robinson
Biophysics Laboratories, Physics Department
Portsmouth Polytechnic, Portsmouth PO1 2DZ, England

L. Paolillo, P. Temussi
Laboratorio per la Chimica e Fisica di Molecole
di Interesse Biologico del C.N.R.
Arco Felice, Naples, Italy

Received October 21, 1972

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Characterization and X-Ray Structure of $(Me_3SiOC)_4Fe_2(CO)_6$

Sir:

An unusual product assigned the dimeric formula $[(Me_3Si)_2Fe(CO)_4]_2$ has been obtained from the reaction of Me_3SiI with $Na_2Fe(CO)_4$ in tetrahydrofuran.[1,2] An interesting structure based on an Fe_2C_2 tetrahedron was proposed,[1,2] and it appeared that the reaction provided a further example of the sometimes unexpected course of reactions between organosilicon halides and metal carbonyl anions.

An anomalous feature of the reported $(Me_3Si)_4Fe_2(CO)_8$ compound was the observation in its mass spectrum of peaks due to $[(Me_3Si)_4Fe_2(CO)_9]^+$ and $[(Me_3Si)_4Fe_2(CO)_{10}]^+$ in greater abundance than those of the presumed molecular ion. The earlier assignment[2] of these peaks as (P + CO) and (P + 2CO) seemed improbable,[3] and we considered it more likely that the

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