Achievements of Multiple-Pulse n.m.r. [and Discussion]

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Achievements of multiple-pulse n.m.r.

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A survey is given of the achievements of multiple-pulse n.m.r. with regard to measurements of shielding tensors of protons. The need to investigate single crystals rather than powder samples is stressed. It is useful to consider separately protons in hydrogen bonds and protons bonded directly to carbon atoms. While for the former group of protons there is now a wealth of experimental shielding data, measurements of shielding tensors of protons from the latter group are still difficult and scarce. Symmetry arguments applied to some available data show that the measurable shielding tensors are substantially influenced by the crystal lattice and do not directly reflect molecular properties. Several implications of this fact are discussed. It is pointed out that continuation of multiple-pulse n.m.r. calls for a new kind of motivation. The study of molecular motions in solids through site-specific spin relaxation measurements may become a new area of interest and activities for multiple-pulse n.m.r.

Introduction

Before the introduction of multiple-pulse techniques into the inventory of the n.m.r. spectroscopist, the information that we had about shielding anisotropies and shielding tensors in general was mainly from liquid crystal studies. The results obtained with this method for protons were judged by Appleman & Dailey in their 1974 review as 'generally unreliable'. The development of multiple-pulse techniques in the late 1960s and early 1970s gave rise to the hope that shielding tensors of protons and other abundant nuclei with strong magnetic moments do become accessible to measurement. Chemical shifts, the traces or orientational averages σ_{av} of shielding tensors $\boldsymbol{\sigma}$, have proved to be extremely useful for the chemist, and a standard phrase in the introduction of many early multiple-pulse papers was that the full shielding tensor, which contains obviously more information than just its trace, should prove to be even more useful.

Now, in 1980, we can say that the first boom of the multiple-pulse business is over (will there ever be another?) and we have to ask ourselves the question: have the hopes of the early 1970s materialized? The answer is, of course, neither a strict 'yes' nor a strict 'no'. Therefore let us consider the actual achievements of multiple-pulse n.m.r. We start with shielding tensors of protons in hydrogen bonds.

Shielding tensors for protons in hydrogen bonds

Figure 1 shows an early multiple pulse spectrum from a powder sample of oxalic acid, $(COOH)_2$. In solid oxalic acid all protons are involved in hydrogen bonds. From this spectrum we can deduce the shielding anisotropy, $\Delta\sigma$, and the shielding asymmetry factor, η:

$$\Delta\sigma = \sigma_{zz} - \tfrac{1}{2}(\sigma_{xx} + \sigma_{yy}), \tag{1}$$

where σ_{ii}, $i = x, y, z$, are the principal components of $\boldsymbol{\sigma}$; and

$$\eta = \frac{\sigma_{yy} - \sigma_{xx}}{\sigma_{zz} - \sigma_{\mathrm{av}}}. \tag{2}$$

The convention $$|\sigma_{zz}-\sigma_{\mathrm{av}}| \geqslant |\sigma_{xx}-\sigma_{\mathrm{av}}| \geqslant |\sigma_{yy}-\sigma_{\mathrm{av}}|$$ is used.

What we cannot learn from powder spectra is the orientation of the principal axis system of $\boldsymbol{\sigma}$ relative to the nuclear framework of the molecule or crystal. The orientation of this axis system contains three out of the nine shielding parameters.

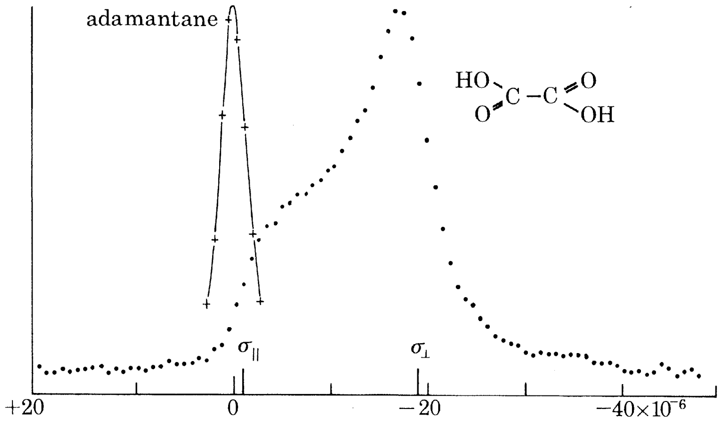

FIGURE 1. Multiple-pulse powder spectrum of α-oxalic acid.

There is another important lesson to learn from figure 1. In oxalic acid all protons are crystallographically equivalent, i.e. the space group of the crystal contains symmetry elements that relate each proton with all others. As a consequence the shielding tensors of all protons in oxalic acid are congruent, i.e. they possess identical sets of principal components, and all protons contribute to one and the same 'powder pattern'. Now suppose that we want to investigate a powder sample of calcium formate, $\mathrm{Ca(HCOO)_2}$. In solution all formate ions are chemically equivalent and therefore give rise to just one proton n.m.r. line. In crystals of $\mathrm{Ca(HCOO)_2}$, however, the formate ions occupy two crystallographically inequivalent sites. Therefore, we can have, and in fact we do have, two non-congruent shielding tensors (Post *et al.* 1980). The powder spectrum would consist of the superposition of two slightly different patterns such as that shown in figure 1. To disentangle the two patterns would obviously be virtually impossible. The lesson to learn is thus that with the exception of very few cases the study of proton shielding tensors with powder samples is of very limited value and that it is practically essential to study single crystals. Shielding tensors of protons in hydrogen bonds have been studied by multiple-pulse techniques in more than a dozen compounds. Usually, rotation patterns, i.e. the angular variation of shifts on rotating crystals about a fixed axis which itself is perpendicular to the applied field, are recorded for three crystals. Such rotation patterns are easily analysed in terms of shielding tensors.

What have we learnt from these studies?

First, and I think this should not be underestimated, we have now accurate knowledge of the shielding tensors of protons in hydrogen bonds in some 20 specific compounds.

Secondly, from these sample cases we are able to derive a general picture of the shielding of protons in hydrogen bonds. This picture has been described before (Haeberlen 1976) and so I shall be very brief here:

(*a*) for protons in hydrogen bonds, $\boldsymbol{\sigma}$ is almost axially symmetric, but deviations from axial symmetry are definitely measurable;

(*b*) the most shielded axis is approximately along the hydrogen bond;

(*c*) the anisotropies $\Delta\sigma$ range from about 17×10^{-6} to up to 44×10^{-6}. There have been attempts to correlate the shielding anisotropies with the lengths and other characteristics of hydrogen bonds (Berglund & Vaughan 1980). These attempts have met with some success but it should be pointed out that their basis is completely empirical. What is lacking to a deplorable extent is a theoretical appraisal of the material accumulated by the experimentalists.

Shielding tensors for protons bound to carbons

A characteristic feature of the shielding of protons bound to carbons is the small shielding anisotropy. Measured values of $\Delta\sigma$ range from about 4 to 10×10^{-6}. This fact has important consequences on several levels for the spectroscopist.

First, for meaningful measurements, ellipsoidal, in practice spherical, samples must be used to avoid line shifts in the rotation patterns resulting from bulk susceptibility shape effects, which may easily become comparable in size to the line shifts owing to the chemical shift anisotropy. While it may be inconvenient to have to work with crystals rather than with powders it can be extremely tedious and difficult to have to shape single crystals into spheres, to orientate them subsequently by means of X-rays and to mount them on an n.m.r. goniometer.

Secondly, the small anisotropies, combined with the fact that the spectral resolution achievable with multiple-pulse techniques is limited, impose strict limitations on the choice of compounds amenable to a multiple-pulse study: the chemical composition of the compound *and* its crystal structure should be such that the number of lines in the multiple pulse spectrum does not exceed, say, eight or, better, four. To the best of my knowledge the largest number of clearly resolved lines in a proton multiple-pulse spectrum from a solid sample is six. It is from a single crystal of lead formate (Post *et al.* 1980).

For these reasons the number of single-crystal multiple-pulse studies of protons bound to carbon atoms is still rather small.

Methylene groups, —CH_2—

In single crystals, methylene protons have only been studied in malonic acid, $CH_2(COOH)_2$ (Sagnowski *et al.* 1977). All methylene groups in the crystal are equivalent, but the two protons of the CH_2 group are inequivalent and have, in fact, non-congruent shielding tensors. The shielding anisotropies are small, 3.8 and 4.8×10^{-6}, respectively. The assignment of the two measured $\boldsymbol{\sigma}$-tensors to the two proton sites is very difficult. The most shielded direction is close to the C—H bond.

Double bond systems, —HC=CH—

Full information about the shielding of double bond systems is available only for KH-maleate (Achlama *et al.* 1978) and for transdiiodoethylene (Spiess *et al.* 1977). Again, the anisotropies are small, $\Delta\sigma$ (KH maleate) $= 3.5 \times 10^{-6}$, $\Delta\sigma$ $(C_2H_2I_2) = 3.8 \times 10^{-6}$.

The formate ion, $[HCOO]^-$

The shielding tensor of the proton in the formate ion has been fully measured in two different crystal sites in $Ca(HCOO)_2$ and in two sites in $Pb(HCOO)_2$, and partially in $Cd(HCOO)_2$ (Post *et al.* 1980). The shielding tensors are substantially different in these four sites. The anisotropies measured in $Ca(HCOO)_2$ and $Pb(HCOO)_2$ are -5.0, -5.2, -9.0 and -9.2×10^{-6}, respectively. The orientation of the principal axis system relative to the formate ion differs from site to site.

Aromatic protons

These have been studied in single crystals of ferrocene (Spiess *et al.* 1976), pyromellitic acid dianhydride (PMDA) (Aravamudhan *et al.* 1979), pyromellitic acid (Tegenfeldt *et al.* 1980) and terephthalic acid (Wendling 1979). The most surprising observation, at first sight, is that none of the principal shielding directions coincides with the C—H bond direction, even in PMDA, where the bond direction is a symmetry axis of the isolated molecule.

This observation, together with the strong dependence of the shielding of the proton in the formate ion on the site of the ion in the crystal lattice, points to substantial intermolecular contributions to the *observable* shielding of protons. Note, however, that these intermolecular shielding contributions can become substantial only because the *intra*molecular shielding anisotropies of these protons are so small. The intermolecular shielding contributions are not *absolutely* but only *relatively* strong.

INTERMOLECULAR CONTRIBUTIONS TO PROTON SHIELDING TENSORS

Remember, when we developed the multiple-pulse techniques we hoped that this tool would make proton shielding tensors measurable quantities. What we meant implicitly were, of course, shielding tensors characterizing proton sites in molecules. Now, in a sense, we have reached our initial goal: we can measure and have measured proton shielding tensors but we must face the situation that what we can measure is not a property of the molecule but to a large extent the consequence of the rather accidental *packing* of molecules in crystals. Thus our goal seems to elude us once again.

It does so not because of ineffective pulse sequences, inherent limits to resolution of the multiple pulse method or imperfections of the equipment used, but simply because Nature is just as it is.

However, the situation is not as disappointing as it may look. With a simple magnetostatic model it is possible to account for the intermolecular shielding contributions with reasonable accuracy. In this model the molecules, or functional atomic groups of molecules in the neighbourhood of the proton considered are treated as classical, magnetizable entities. In the magnetic field they acquire magnetic dipole moments $\boldsymbol{\mu}_i = L^{-1}\boldsymbol{\chi}_i \cdot \boldsymbol{B}_0$, where $\boldsymbol{\chi}_i$ is the molar susceptibility of the ith molecule (or functional group) and L is Avogadro's number. The proton considered 'feels' the field originating in these dipoles. Its effect can be expressed as an intermolecular shielding contribution

$$\boldsymbol{\sigma}_{\text{inter}} = \sum_i \boldsymbol{\sigma}_{i,\,\text{inter}}, \tag{3a}$$

with

$$\boldsymbol{\sigma}_{i,\,\text{inter}} = \frac{1}{L}\left(\frac{\boldsymbol{\chi}_i}{r_i^3} - \frac{3(\boldsymbol{\chi}_i \cdot \boldsymbol{r}_i)\,\boldsymbol{r}_i}{r_i^5}\right). \tag{3b}$$

$\boldsymbol{r}_i$ is the vector from the proton to the dipole i, $r_i = |\boldsymbol{r}_i|$.

Subtracting $\boldsymbol{\sigma}_{\text{inter}}$ from $\boldsymbol{\sigma}_{\text{exptl}}$ should result in the *intra*molecular shielding contribution $\boldsymbol{\sigma}_{\text{intra}}$.

We have applied this procedure to the formate ion in the four different crystal sites studied and to the aromatic protons in PMDA, pyromellitic acid and terephthalic acid. Intrinsic tests of the procedure are whether or not $\boldsymbol{\sigma}_{\text{intra}}$ reflects molecular symmetry and whether or not $\boldsymbol{\sigma}_{\text{intra}}$ is dependent on the crystal site. In the cases studied so far these tests are not passed with the highest possible marks but we feel they are passed well enough to allow extraction of intramolecular shielding tensors σ_{intra} from the data, suited for comparison with molecular orbital calculations of shielding tensors which, alas, are not available, and, as it seems, not feasible at the moment.

To summarize this part of the paper we may say the following. The multiple-pulse method has been successful in the sense that accurate measurements of shielding tensors of protons in widely varying bonding situations could be carried out. From the point of view of basic research, progress is hampered by poor theoretical understanding of the experimental results. The theoretical community has practically ignored the experimental progress achieved in the last 5 years. Empirical applications comparable to those of chemical shifts σ_{av} are severely limited by the stringent requirements a sample must meet to qualify as a candidate for a successful multiple-pulse study (chemical composition, size of the molecule, crystal structure, possibility of growing crystals and of shaping them into spheres, etc.).

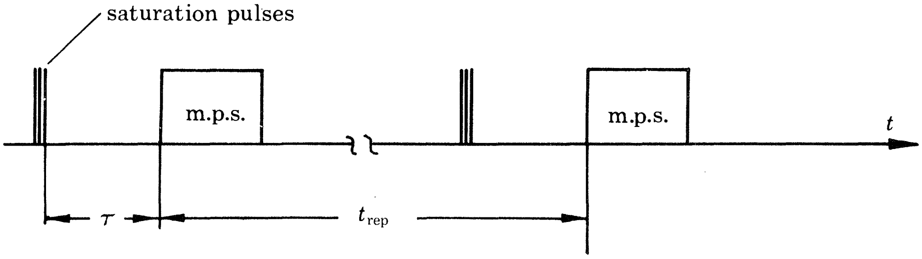

FIGURE 2. Scheme used for measuring relaxation times of individual protons in single crystals of malonic acid.

OTHER APPLICATIONS OF MULTIPLE-PULSE N.M.R.

There are applications of multiple-pulse n.m.r. that do not depend on an understanding of shielding tensors. I shall briefly discuss two of them.

Spin diffusion

In liquids, nuclei in different positions of a molecule may have different spin lattice relaxation times, T_1. This is well known and widely exploited in the use of ^{13}C nuclei. By contrast, the dipolar couplings of the protons in solids should lead quickly, after a perturbation of the spin system, to the establishment of a common spin temperature of all the spins and this common spin temperature implies a common T_1 for all the protons in the sample. This spin-temperature concept is well accepted but experimental tests of it are mostly indirect. The multiple-pulse method offers the opportunity for an accurate test of the equality of relaxation times of protons in different crystal sites. We have carried out such a test on malonic acid. We used the scheme shown in figure 2.

To avoid problems resulting from heating the sample crystal by the approximately 8000 pulses of the multiple-pulse sequence (m.p.s.), we worked with a constant repetition time $t_{\text{rep}} = 200$ s $\gg T_1$. Before firing the m.p.s. we saturated all protons with one or several 90° saturation pulses. τ was varied in an appropriate range around T_1. The heights of the lines in the multiple-pulse spectra were recorded and plotted in the usual semilogarithmic manner against τ to get the individual relaxation times. A representative plot is shown in figure 3. It demonstrates the equality of the relaxation times of the protons in the four inequivalent positions in malonic acid and thus confirms in a direct manner the spin-temperature concept. The deuterons in a sample of fully deuterated malonic acid do *not* approach equilibrium with a common spin lattice relaxation time.

Flip motions of water molecules

Many crystals incorporate water molecules into their lattice. In some instances the protons of the water molecules stay at their lattice sites, in others the water molecules undergo 180° rotational jumps about their twofold axes. These motions are called 'flips'. They are particularly remarkable since the water molecules in question are usually hydrogen-bonded to other molecules of the crystal. The rate of the flips ranges, depending upon the temperature of the crystal, from virtually zero to more than $10^6\,\mathrm{s}^{-1}$. Their dynamics (Arrhenius equation obeyed? If yes, activation energy? Pre-exponential factor?) can be studied with multiple-pulse techniques.

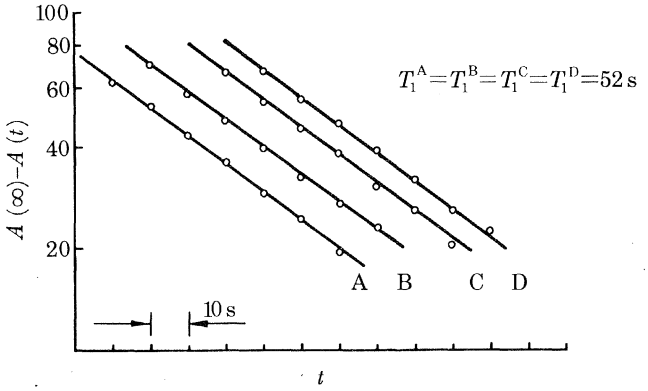

FIGURE 3. Demonstration of the equality of the individual spin lattice relaxation times at 293 K for the four inequivalent protons labelled A, B, C and D in single crystals of malonic acid.

Chemical shift differences of the two water protons for appropriate orientations of the crystals relative to the applied field are exploited: for small flip rates, separate lines are observed for the two protons, for large flip rates the two lines merge into a single line. In deuterated crystals, flip motions of D_2O molecules can be studied by deuteron n.m.r. Differences of the nuclear quadrupole moment – electric field gradient interactions at the two deuteron sites are exploited. For protons, only the limiting cases for slow and fast flip motions have been demonstrated so far (Rosenberger *et al.* 1975; Berglund & Vaughan 1980; McKnett *et al.* 1975; Müller *et al.* 1978; Feucht 1978), while for deuterons a complete study from the slow to the fast exchange limit has been carried out for the D_2O molecules in single crystals of deuterated pyromellitic acid dihydrate (Schajor 1980).

THE ANTISYMMETRIC PART OF THE SHIELDING TENSOR

In this paper I have stated several times that by multiple-pulse techniques shielding tensors $\boldsymbol{\sigma}$ of protons have become accessible to measurement. This is not quite true. What has become measurable is the symmetric part $\boldsymbol{\sigma}^{\mathrm{s}}$ of the full tensor $\boldsymbol{\sigma}$. $\boldsymbol{\sigma}^{\mathrm{s}}$ has six independent components, $\boldsymbol{\sigma}$ has nine. What has so far escaped measurement is the antisymmetric part, $\boldsymbol{\sigma}^{\mathrm{a}}$, of $\boldsymbol{\sigma}$. It has three independent components. Its measurement is still a challenge to the n.m.r. spectroscopist. I am afraid that I cannot propose a workable strategy to measure the individual components of $\boldsymbol{\sigma}^{\mathrm{a}}$ but I think that the sum of the squares of the components of $\boldsymbol{\sigma}^{\mathrm{a}}$ can be measured. The key is the spin lattice relaxation rate resulting from chemical shift interactions, $1/T_{1,\,\mathrm{cs}}$. For isotropic, diffusive molecular tumbling and under extreme narrowing conditions $1/T_{1,\,\mathrm{cs}}$ is given theoretically by (Spiess 1978)

$$1/T_{1,\,\mathrm{cs}} = \omega_0^2\tau_\mathrm{c}[2\{(\sigma^\mathrm{a}_{xy})^2+(\sigma^\mathrm{a}_{xz})^2+(\sigma^\mathrm{a}_{yz})^2\}+\tfrac{2}{15}\Delta\sigma^2(1+\tfrac{1}{3}\eta^2)], \tag{4}$$

where τ_c is the correlation time for 2nd rank spherical tensors. In (4) all quantities except $\{(\sigma^\mathrm{a}_{xy})^2+(\sigma^\mathrm{a}_{xz})^2+(\sigma^\mathrm{a}_{yz})^2\}$ can be measured separately, partly in the liquid $(1/T_{1,\,\mathrm{cs}}, \tau_\mathrm{c}, \omega_0)$ and partly in the solid state $(\Delta\sigma, \eta)$ so that eventually it can be solved for $\{(\sigma^\mathrm{a}_{xy})^2+(\sigma^\mathrm{a}_{xz})^2+(\sigma^\mathrm{a}_{yz})^2\}$.

The procedure probably works better for ^{13}C than for ^{1}H, but this is also the field where practical interest in $\boldsymbol{\sigma}^\mathrm{a}$ exists. In ^{13}C relaxation studies the chemical shift relaxation mechanism is often invoked as a *deus ex machina* to 'explain' relaxation rates of carbons that do not bind protons. Inserting reasonable numbers for τ_c and $\Delta\sigma$ into (4), but neglecting the antisymmetric terms gives calculated rates that most often fall short of the measured rates. Note, however, that for a comparable size of the components of $\boldsymbol{\sigma}^\mathrm{a}$ and $\boldsymbol{\sigma}^{\mathrm{s,\,traceless}}$ the former are much more effective than the latter in relaxing the nucleus; therefore, knowledge, even if only approximate, about $\boldsymbol{\sigma}^\mathrm{a}$ *is* important. It could release the chemical shift relaxation mechanism from its role as a *deus ex machina*.

The work that I have reported here is, of course, the work of many. Most of their names appear in the reference list. I wish to express my gratitude to all of them. The measurements of spin lattice relaxation times of individual protons in malonic acid were done by N. Pislewski.

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Discussion

R. K. HARRIS (*School of Chemical Sciences, University of East Anglia, Norwich, U.K.*). Dr Haeberlen has presented a somewhat gloomy picture of multiple-pulse experiments. However, it seems to me that this pessimism should perhaps be restricted to proton work on single crystals. One might be more optimistic about studies with the use of, say, ^{19}F or ^{31}P. It is also surely feasible that multiple-pulse experiments for protons may prove to be very useful when combined with magic angle rotation. In this respect the situation for high-resolution abundant-spin n.m.r. spectroscopy seems to be similar to that for rare spins 5 years ago, and perhaps the future will be as bright. Would Dr Haeberlen agree?

U. Haeberlen. I did not intend to paint the future of the multiple-pulse technique as an experimental technique in dark colours. What I wished to emphasize is that the original stimulus for the development of multiple-pulse techniques – the measurement of shielding tensors – does not any longer provide sufficient motivation for doing such experiments and for striving for new records in terms of resolution. I also feel that seeing manifestations of chemical shift anisotropy in other kinds of experiments, e.g. multiple-quantum spectroscopy and multiple-pulse experiments combined with (near) magic angle sample spinning, should not be taken as evidence that the particular technique is useful for probing matter. Such evidence must be derived from other sources. With existing techniques enough shielding tensors have been measured in a broad scope of environments of the respective nuclei that interest in shielding tensors as such approaches saturation. The situation may change when meaningful comparisons of experimental and theoretical shielding tensors become possible.

Being a 'multiple-pulser' I certainly welcome any applications of the technique. Some nice work has been done in the area of molecular motions in solids and it is there that I see a bright future for the multiple-pulse techniques.