

Correlation of Isotropic and Anisotropic Chemical Shifts in Solids by Two-Dimensional Variable-Angle-Spinning NMR

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Abstract. We describe a new solid-state nuclear magnetic resonance (NMR) technique for correlating anisotropic and isotropic chemical shifts in powdered samples. Two-dimensional (2D) NMR spectra are obtained by processing signals acquired in *independent* experiments for different angles between the sample spinning axis and the Zeeman magnetic field. This 2D NMR approach can therefore resolve individual static anisotropic lineshapes according to their isotropic chemical shift frequencies, without use of sudden mechanical motions or multiple-pulse irradiation schemes. Applications of the technique are illustrated with an analysis of the chemical shift anisotropy for the eight distinct ^{13}C sites in tyrosine.

The combination of cross polarization-dipolar decoupling techniques with magic-angle sample spinning, has made NMR particularly useful for the study of polycrystalline or amorphous solids.¹⁻⁴ When applied to the observation of dilute spin-1/2 nuclei, the resulting CPMAS NMR experiment affords spectra in which sharp resonances arising from chemically inequivalent sites appear resolved at distinctive isotropic chemical shifts. This spectral resolution however is achieved by elimination of the solid-state chemical shift anisotropy (CSA), a potentially valuable interaction whose lineshapes afford a detailed description of structural and dynamic properties in the systems under analysis. During the last decade, different methods for retaining at least part of the information contained in the anisotropy have been demonstrated.⁵⁻¹⁷ Particularly promising are those approaches based on two-dimensional (2D) NMR separation of interactions, which provide detailed information about anisotropic chemical shifts while maintaining the normal resolving power of MAS. Most of these experiments rely on the following idea: anisotropic information is encoded during a variable time t_1 during which spins precess with a CSA-dependent evolution frequency, followed by a second time t_2 during which signals are acquired while precessing under the effects of the fast-spinning MAS Hamiltonian. These 2D correlation techniques can be

catalogued into two groups, according to how anisotropic evolution is introduced. In one group, incomplete isotropic averaging is achieved either by spinning the sample fast but off the magic angle, by spinning it slowly at the magic angle, or by not spinning it at all.^{12,13,15,17} This class of techniques requires both sudden mechanical motions and storage of evolving magnetizations. The second group of experiments relies on synchronized mechanical and radiofrequency perturbations, for example by applying multiple-pulse sequences on samples undergoing constant MAS. This rf irradiation counteracts the rotational averaging, and provides a net anisotropic evolution.^{5,9,11,16,17}

In the present study, we describe and illustrate an alternative way of correlating isotropic and anisotropic chemical shifts which avoids both the necessity of multiple-pulse irradiation of the spins as well as sudden mechanical motions. Instead, isotropic-anisotropic 2D NMR spectra are obtained by processing a set of signals arising from a rapidly spinning sample, acquired in independent experiments as a function of the orientation of the spinning axis. To visualize the principles of this

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variable-angle correlation spectroscopy (VACS) technique, consider the evolution of spins in a powdered sample rotating about an axis inclined at an angle β with respect to the external magnetic field. In the usual rotating frame of reference, the precession frequency ω of each site can be written as⁷

$$\omega = \omega_i + P_2(\cos\beta) \cdot \omega_a(\theta, \phi) \quad (1)$$

where ω_i is the difference between the isotropic chemical shift of the site and the transmitter offset, $\omega_a(\theta, \phi)$ is the orientation-dependent anisotropic frequency in a given crystallite, and

$$P_2(\cos\beta) = (3\cos^2\beta - 1)/2 \quad (2)$$

is the second-order Legendre polynomial of $\cos\beta$. The signal detected at a time t after excitation of the spins is given by the weighted integral of eq 1, averaged over all sites and orientations in the sample:

$$S(\beta, t) = \iint I(\omega_i, \omega_a) \exp[i(\omega_i \cdot t_i + \omega_a \cdot P_2(\cos\beta)t)] d\omega_i d\omega_a \quad (3)$$

Here, $I(\omega_i, \omega_a)$ is the joint probability distribution of a spin having an isotropic frequency ω_i and an anisotropic frequency ω_a . This distribution is identical to the 2D NMR spectrum correlating purely anisotropic powder lineshapes with the isotropic chemical shift of each site, and becomes available from a Fourier analysis of $S(\beta, t)$ regarded as an implicit function of two variables $t_i = t$ and $t_a = P_2(\cos\beta)t$:

$$I(\omega_i, \omega_a) = \iint S(\beta, t) \exp[-i(\omega_i \cdot t_i + \omega_a \cdot t_a)] dt_i dt_a \quad (4)$$

Thus, although it may be very difficult to design a 2D NMR experiment in which a system evolves under the exclusive effects of ω_i and ω_a during independent times t_i and t_a , it is relatively simple to collect the corresponding data. Figure 1 illustrates the considerable region of (t_i, t_a) -space that can be sampled by acquiring the time-domain signal of the spins for different fixed angles of rotation β . From these VAS experiments, $I(\omega_i, \omega_a)$ correlation distributions can be obtained by conventional 2D Fourier transformation of the data interpolated over a regular (t_i, t_a) -grid. The $-0.5 \leq P_2(\cos\beta) \leq 1$ constraint precludes acquisition of data for the entire isotropic-anisotropic time-domain, but we find that the resulting "truncation" only leads to small diagonal ridges emerging from the main resonance peaks.

Figure 2 presents an experimental application of this approach to the separation of ¹³C isotropic and an-

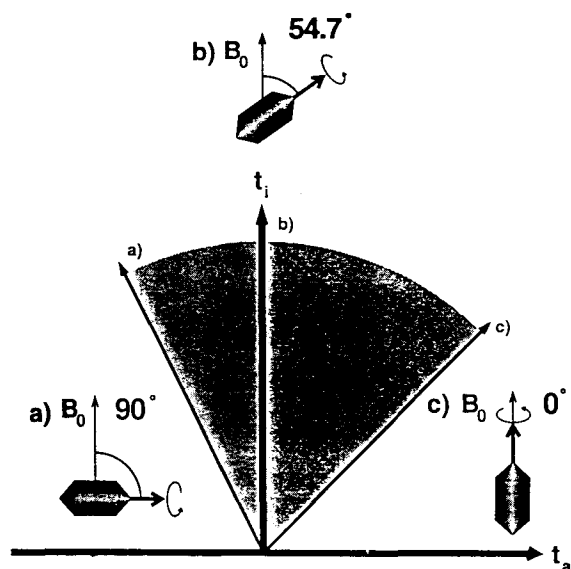


Fig. 1. Experimental principles involved in the extraction of isotropic-anisotropic 2D NMR spectra using variable-angle correlation spectroscopy. The direction along which the system evolves in the $(t_i = t, t_a = P_2 t)$ plane can be varied between $P_2(\cos\beta) = 1$ and $P_2(\cos\beta) = -0.5$ by changing the angle of sample spinning β . The shaded area illustrates the time-domain region accessible by this scheme. The final NMR spectrum is obtained by interpolating the acquired data over a regular array followed by two-dimensional Fourier transformation.

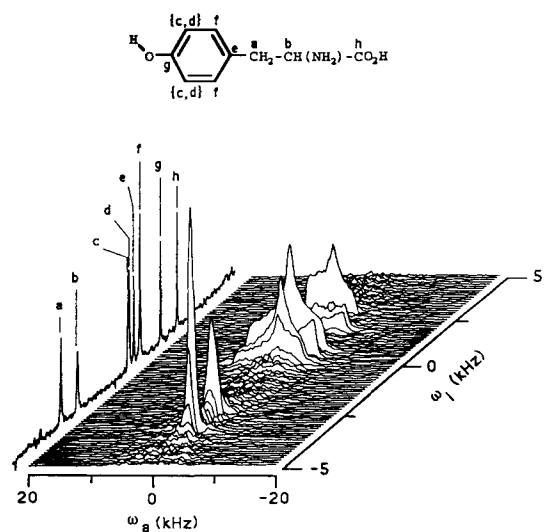


Fig. 2. Two-dimensional isotropic-anisotropic NMR chemical shift correlation spectrum of L-tyrosine, showing the isotropic projection and the assignment of the peaks to each site in the molecule. A set of 65 signals was digitized at rates of 12 kHz over a range of angles β satisfying $-0.5 \leq P_2(\cos\beta) \leq 0.5$; experimental data were then interpolated over a regular grid composed by 128×256 points and Fourier transformed.

Table 1. ^{13}C NMR shielding parameters in L-tyrosine^a

Carbon atom	Isotropic shift ^b (± 0.5 ppm)	$\Delta\omega$ (± 1 ppm)	η
a	37.1	12.2	0.45 \pm 0.2
b	57.3	-22.1	0.70 \pm 0.1
c	115.0	-99.6	0.67 \pm 0.05
d	117.9	-101.8	0.63 \pm 0.03
e	123.7	-108.4	0.67 \pm 0.03
f	131.0	-106.2	0.88 \pm 0.02
g	156.0	-92.9	0.95 \pm 0.02
h	175.5	-68.6	0.87 \pm 0.04

^aIn ppm, using the usual convention $|\omega_{33}| \geq |\omega_{22}| \geq |\omega_{11}|$.

^bDownfield from TMS, measured using adamantane as external reference.

sotropic chemical shifts in a powdered sample of L-tyrosine. Data were collected in a 4.2 T magnet using a home-built spectrometer and probe, equipped with a computer-controlled stepping motor for changing the orientation of the spinning sample. The instrumental setup and computational procedures used for obtaining this spectrum will be described more completely in a forthcoming publication.¹⁸ Apart from minor distortions in the lineshapes of non-protonated carbons, the spectrum in Fig. 2 presents all the features expected from a 2D VACSYS NMR experiment. Since time-domain signals were acquired for both positive and negative values of $P_2(\cos\beta)$, a series of "echoes" formed along the t_a axis for positive values of t_1 . This simplifies the extraction of a purely absorptive $I(\omega_i, \omega_a)$ spectrum, whose isotropic resolution is therefore the same as that obtained under normal MAS conditions. The singularities displayed by the resolved traceless powder patterns afford a direct measurement of the CSA values, and a complete list of the ^{13}C NMR shielding parameters of tyrosine is given in Table 1.

When compared with previously proposed isotropic-anisotropic correlation techniques, the main advantage of the present approach is its experimental simplicity. Since no sudden mechanical switching between interactions is involved, the method avoids both special mechanical designs as well as signal-to-noise problems associated with magnetization storage. Furthermore, since neither rotor-synchronous irradiation nor detection is required, the present method is well-suited for recording spectra on samples requiring large numbers of signal-averaging scans. The only complication of the VACSYS technique is the necessity for interpolating the time-domain data before transformation into the frequency-domain. Nevertheless, once implemented, this is a straightforward computational procedure which takes only a small fraction of the time involved in, for example, the Fourier transformation itself.

The most significant aspect of the experiment is that it extracts 2D NMR data by scaling rather than by switching nuclear spin interactions. Although similar approaches have been proposed and used before in the design of magnetic resonance imaging experiments,¹⁹ their application to the correlation of internal spin interactions seems to have been hitherto overlooked. From a spectroscopic point of view, the effects introduced by changing the axis of sample rotation on the anisotropic chemical shift evolution of spin-1/2 nuclei are analogous to those obtained when changing gradients in NMR imaging experiments. A consequence of this is the apparent similarity between the sampling of (t_1, t_a) -space shown in Fig. 1, and the sampling of k -space in back-projection imaging experiments.

This parallelism opens the possibility of applying widely used and successful imaging techniques, such as spin-warp and echo-planar-imaging, to the design of more efficient solid-state correlation experiments. Use of time as a parameter for the non-cartesian sampling of multi-dimensional domains may also find a general application for the simplification of other solution and solid-state NMR experiments. Possible examples include the extraction of chemical shielding parameters in single-crystal studies, more efficient ways of correlating CSA and dipole interactions, and experiments involving scaling or modulation of isotropic spin interactions in solution NMR. These potential applications are currently under investigation.

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