Communication

Constant-time method for measuring inter-nuclear distances in static powders

Jae-Seung Lee, A.K. Khitrin *

Department of Chemistry, Kent State University, Kent, OH 44242-0001, USA

Received 24 January 2007; revised 22 February 2007
Available online 1 March 2007

Abstract

We present a constant-time modification of a single-echo 2D technique for measuring the constants of spin–spin interaction between heteronuclei in static powder samples. Compared to the sequence with variable echo time, the new scheme provides higher resolution. Another important improvement is a possibility to eliminate the central peak from a natural-abundance matrix. The experimental results are presented for glycine-$[^{13}C_\alpha, ^{15}N]$ and glycine-$[^{13}C', ^{15}N]$.
© 2007 Elsevier Inc. All rights reserved.

Keywords: Heteronuclei; Adiabatic cross-polarization; SPINAL; Static solid; Inter-nuclear distance

Solid-state NMR can be a very accurate tool to measure inter-nuclear distances in solids. The best results were obtained for static solids, where the nutation spectroscopy [1] or the Carr–Purcell sequence have been used to eliminate the chemical shift interactions in homonuclear systems [2], and the MLEV-8 sequence with composite pulses has been applied to a heteronuclear system [3]. Recently, we introduced the least-perturbing single-echo 2D pulse sequence, which allows reaching an unprecedented accuracy in measuring the inter-nuclear distances, comparable to that of the X-ray and neutron scattering [4].

The pulse sequence [4] is shown in Fig. 1a. It resembles the pulse sequences used in two-dimensional heteronuclear *J* spectroscopy [5] and solid-state NMR measurement of ^{13}C–^{15}N dipolar couplings in a single crystal [6]. Our sequence starts with adiabatic cross-polarization (ACP) [7], performed with two frequency-sweeping pulses, to boost the ^{13}C polarization. Two simultaneous 180° pulses on ^{13}C and ^{15}N spins in the middle of the varying evolution period t_1 refocus dephasing by chemical shifts and form an echo at the end of the evolution period. The dipolar powder pattern is obtained as a Fourier transform with respect to t_1.

The major factor limiting the precision is line broadening, which comes from the homonuclear dipole–dipole interactions and non-perfect heteronuclear decoupling. A constant-time [8] scheme can significantly decrease line broadening. The pulse sequence with the constant echo time τ is shown in Fig. 1b. It works as follows. The "sliding" 180° ^{15}N pulse, applied at $t_1/2$ ($t_1 < \tau$), changes the sign of the heteronuclear ^{13}C–^{15}N dipole–dipole and *J*-coupling. One hundred and eighty degree ^{13}C pulse at $\tau/2$, in the middle of the evolution period, restores the sign of the ^{13}C–^{15}N spin–spin interaction. As a result, the total evolution "in one direction" corresponds to the evolution time t_1. The Pake powder pattern in this experiment is also obtained as a Fourier transform over t_1. Ideally, the constant-time scheme may completely eliminate line broadening for the directly detected nucleus (at the expense of sensitivity). Another important improvement, offered by the scheme in Fig. 1b, is a significant decrease of the central peak from the natural-abundance matrix. Elimination of the central peak is achieved by a standard dc correction in a 2D Fourier transform. This is valuable since, in many cases, using isotopically depleted matrices may be cost-prohibitive.

---

* Corresponding author. Fax: +1 330 672 3816.
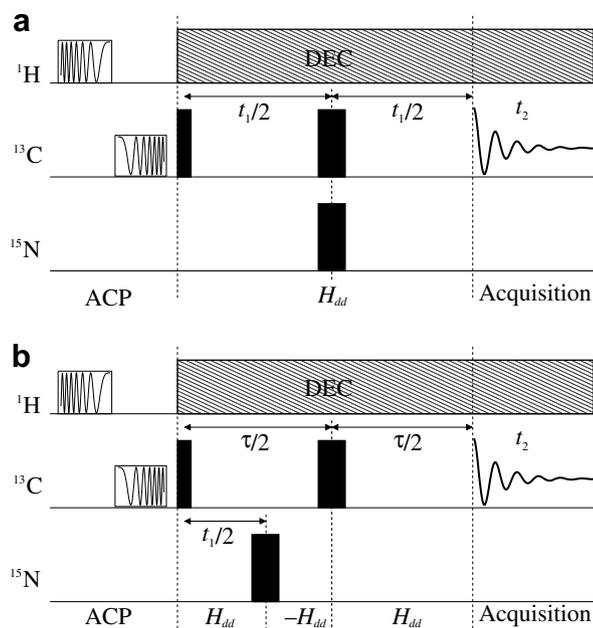 *E-mail address:* akhitrin@kent.edu (A.K. Khitrin).

Fig. 1. NMR pulse sequences for the ^{13}C–^{15}N distance measurement: (a) the sequence with varying echo time and (b) the constant-time sequence.

The experimental results are presented for α-crystalline form of glycine-[^{13}C$_\alpha$,^{15}N] (ISOTEC). Labeled glycine was diluted to 2% in natural abundance (n.a.) glycine (Aldrich) and to 1% in isotopically depleted glycine-[^{12}C,^{14}N] (ISOTEC). All the samples were recrystallized from aqueous solutions. The experiments have been performed at room temperature (25 °C) using a Varian Unity/Inova 500 MHz NMR spectrometer with a triple-resonance "indirect" probe for liquids. The first ^{1}H adiabatic pulse had 100 ms duration and 100 kHz frequency-sweeping range, and the second ^{13}C pulse had 100 ms duration and 60 kHz frequency-sweeping range. ACP provided about sevenfold increase of the ^{13}C polarization compared to its thermal equilibrium value. Proton decoupling, at 50 kHz decoupling power, has been performed with the SPINAL-64 heteronuclear decoupling sequence [9]. The 90° and 180° pulses for a ^{13}C spin echo were, respectively, 14 and 28 μs long, and the 180° pulse on ^{15}N spin was 80 μs long. The fixed echo times τ were 25.6 ms for 2% glycine-[^{13}C$_\alpha$, ^{15}N] in n.a. glycine and 32.0 ms for 1% glycine-[^{13}C$_\alpha$,^{15}N] in glycine-[^{12}C$_2$,^{14}N].

The experimental dipolar powder spectra for glycine-[^{13}C$_\alpha$,^{15}N] are shown in Fig. 2. The spectra are the so-called "sky projections" obtained from the real (cosine) Fourier transforms with respect to the evolution time t_1, using the Varian's VnmrJ software. Compared to the experiment with variable echo time [4], performed with the same probe and decoupling parameters, the Lorentzian broadening factor [4] decreased from 36 to 16 Hz for 1% glycine-[^{13}C$_\alpha$,^{15}N] in glycine-[^{12}C$_2$,^{14}N] (Fig. 2a), and from 59 to 24 Hz for the 2% sample in n.a. matrix (Fig. 2b). ^{13}C–^{15}N NMR distance, estimated by using a simple data processing scheme of Ref. [4], is 0.149 nm for both samples.
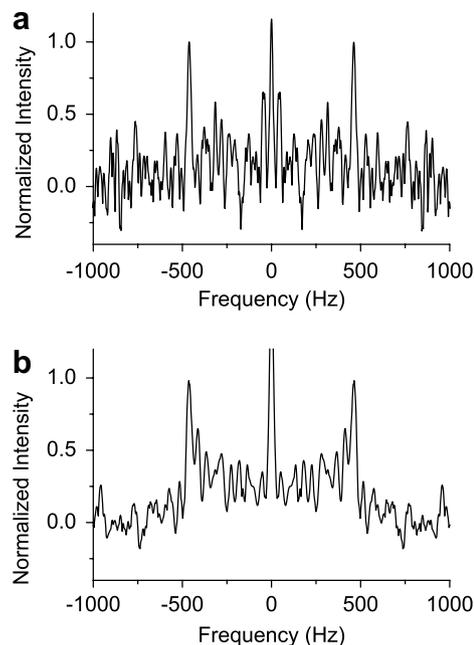

Fig. 2. The dipolar powder spectra obtained with the constant-time experiment. The number of t_1 increments was 128. (a) One percent glycine-[^{13}C$_\alpha$,^{15}N] in glycine-[^{12}C$_2$,^{14}N] (exp. time ~52 h, τ = 32.0 ms). (b) Two percent glycine-[^{13}C$_\alpha$,^{15}N] in natural-abundance glycine (exp. time ~76 h, τ = 25.6 ms).

For 2% glycine-[^{13}C$_\alpha$,^{15}N] in n.a. glycine (Fig. 2b), the line-width of the central peak decreased about two times compared to the variable-time measurement. In addition, a standard dc correction in a 2D Fourier transform suppressed the peak height by about 4.5 times.

An example of measuring a longer C–N distance is shown in Fig. 3 for 2% glycine-[^{13}C',^{15}N] in n.a. glycine. With the constant-time scheme, the Lorentzian broadening factor has decreased from 26 to 12 Hz. The estimated ^{13}C–^{15}N NMR distance, 0.253 nm, is consistent with the one determined in Ref. [4].

The spectra shown in this paper were too noisy to improve the accuracy of NMR distances, even though the line broadening has been significantly decreased. True advantages of the constant-time technique can be realized by using
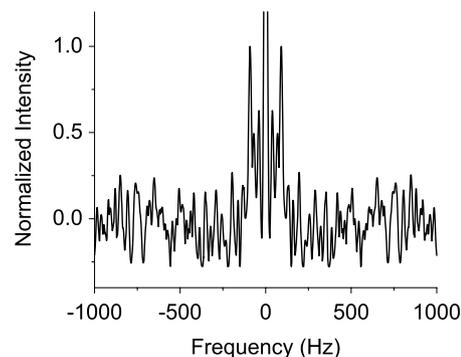

Fig. 3. The dipolar powder spectrum obtained with the constant-time experiment for 2% glycine-[^{13}C',^{15}N] in natural-abundance glycine (exp. time ~89 h, τ = 32.0 ms). The number of t_1 increments was 128.

a probe with better ^{13}C sensitivity and higher proton decoupling power.

References

[1] C.S. Yannoni, R.D. Kendrick, NMR nutation spectroscopy: a method for determining molecular geometry in amorphous solids, J. Chem. Phys. 74 (1981) 747–749.

[2] M. Engelsberg, C.S. Yannoni, The determination of bond lengths in solids using the Carr–Purcell sequence, J. Magn. Reson. 88 (1990) 393–400.

[3] Y. Ishii, T. Terao, Determination of interheteronuclear distances by observation of the Pake-doublet patterns using the MLEV-8 sequences with composite pulses, J. Magn. Reson. Ser. A 115 (1995) 116–118.

[4] J.-S. Lee, A.K. Khitrin, Accurate measurement of ^{13}C–^{15}N distances with solid-state NMR, J. Chem. Phys. 124 (2006) 144508.

[5] R. Freeman, H.D.W. Hill, High-resolution study of NMR spin echoes: "J Spectra", J. Chem. Phys. 54 (1971) 301–313.

[6] D.M. Schneider, R. Tycko, S.J. Opella, High-resolution solid-state triple nuclear magnetic resonance measurement of ^{13}C–^{15}N dipole–dipole couplings, J. Magn. Reson. 73 (1987) 568–573.

[7] J.-S. Lee, A.K. Khitrin, Adiabatic cross-polarization via intermediate dipolar-ordered state, J. Magn. Reson. 177 (2005) 152–154.

[8] A. Bax, R. Freeman, Investigation of complex networks of spin–spin coupling by two-dimensional NMR, J. Magn. Reson. 44 (1981) 542–561.

[9] B.M. Fung, A.K. Khitrin, K. Ermolaev, An improved broadband decoupling sequence for liquid crystals and solids, J. Magn. Reson. 142 (2000) 97–101.