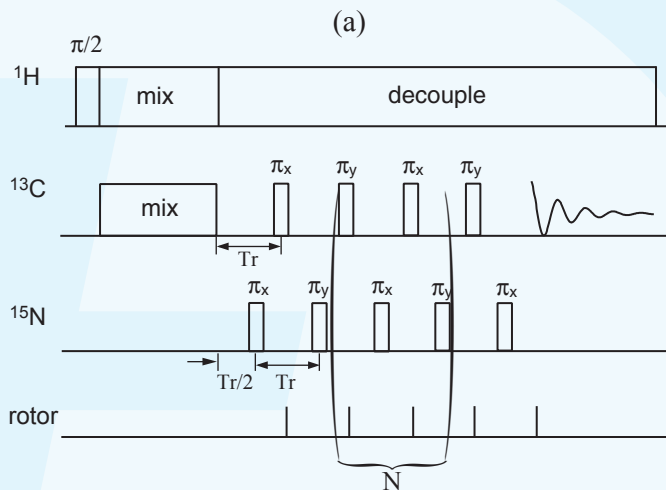


1. Introduction

Rotational-Echo **DO**uble-Resonance nuclear magnetic resonance (REDOR) is a magic-angle spinning (MAS) experiment for measuring internuclear distances between heteronuclear spin pairs through dipolar interaction.<sup>[1,2]</sup> The spin pair consists of two enriched sites, e.g., <sup>13</sup>C and <sup>15</sup>N in the same molecule. In the REDOR experiment, the sample is spun about the magic angle and cross-polarization from protons is applied to enhance the signal intensity. The <sup>13</sup>C and <sup>15</sup>N dipolar interaction is averaged due to MAS, thus removed, and reintroduced by a set of rotor-synchronized  $\pi$  pulses on both <sup>13</sup>C and <sup>15</sup>N which results in a signal intensity reduced spectrum.

2. Pulse sequence

- <sup>1</sup>H<sub>90</sub>:   4  $\mu$ s
- Mixing rf field:  50 kHz
- <sup>13</sup>C<sub>180</sub>:   8 $\mu$ s
- <sup>15</sup>N<sub>180</sub>:   18 $\mu$ s
- <sup>1</sup>H decoupling:  55 kHz
- Phase cycling for  $\pi$  pulses:  xy



(b)

Event Number	1	2	3	4	5	6	7	8	9	10	11	12	13	14
Name:														
Delay	pd	H90	mix	delay1	N180	delay2	C180	delay2	N180	delay2	C180	delay2	N180	delay2
F1_Ampl		H90 amp	Hmix amp	Hdec amp	Hdec amp	Hdec amp	Hdec amp	Hdec amp	Hdec amp	Hdec amp	Hdec amp	Hdec amp	Hdec amp	Hdec amp
F1_Ph		X	Y	Y	Y	Y	Y	Y	Y	Y	Y	Y	Y	Y
F1_Attn		H90 attn	Hmix attn	Hdec attn	Hdec attn	Hdec attn	Hdec attn	Hdec attn	Hdec attn	Hdec attn	Hdec attn	Hdec attn	Hdec attn	Hdec attn
F1_TxGate														
F2_Ampl			F2 amp				F2 amp				F2 amp			
F2_Ph			X				X				Y			
F2_Attn			F2 attn				F2 attn				F2 attn			
F2_TxGate														
Loop														
Acq														
Acq_phase														
F3_Ampl					F3 amp				F3 amp					F3 amp
F3_Ph					X				Y					X
F3_Attn					F3 attn				F3 attn					F3 attn

Parameter	Value	Parameter	Value	Parameter	Value
pd	50u	rd	4u	Hmix attn	16
H90	4u	ad	4u	Hdec attn	17
mix	8u	Acq. Time	20.48m	F2 amp	87
delay1	=1000000/(2*[spin speed])-[N180]/2	Last Delay	2s	F2 attn	10
N180	18u	H90 amp	82.2	N	11
delay2	=1000000/(2*[spin speed])-[C180]+[N180]/2	Hmix amp	82.2	F3 amp	100
C180	8u	Hdec amp	80	F3 attn	6
delay3	=1000000/(2*[spin speed])-[N180]/2-[rd]-[ad]	H90 attn	16	spin speed	2000Hz

Fig. 1a: REDOR pulse sequence. b: Actual sequence in the NTNMR sequence editor. Delays between  $\pi$  pulses are calculated conveniently by means of the mathematical expressions in the dashboard(Fig.1 b). Update automatically occurs upon entry rotor speed and  $\pi$  pulse width.

### 3. Experiments

Sample preparation: 1 portion of glycine-1-<sup>13</sup>C, <sup>15</sup>N (>98% Cambridge Isotope Laboratories, Inc.) and 9 portions of natural abundance glycine dissolved in 50°C water. The solution is left standing at room temperature and air. Crystals precipitate after a few hours. Spectra were acquired using a 7 Tesla spectrometer and a Doty 5 mm HXY triple resonance MAS probe. The number of rotor cycles during the <sup>13</sup>C/<sup>15</sup>N dipolar evolution period was up to 32. The rotor speed was 1.3 kHz.

### 4. Results

Figure 2 shows the spectra acquired at the end of the 8th rotor cycle as shown in the pulse sequence in Fig.1, without (a) and with (b)  $\pi$  pulses. The ratio of the total intensity of all sidebands in the REDOR spectrum ( $S_r$ ) to the corresponding spectrum without  $\pi$  pulses ( $S_0$ ) is plotted as function of  $\lambda_d = N_c D T_r$  (Fig.2c).  $N_c$  represents the number of rotor cycles,  $D$  the dipolar constant, and  $T_r$  the rotor period. The solid line in Fig.2c is calculated according to [2]:

$$\frac{S_r}{S_0} = \frac{\sqrt{2}\pi}{4} J_{1/4}(\sqrt{2}N_c D T_r) J_{-1/4}(\sqrt{2}N_c D T_r)$$

The best fit to the experimental data occurs for  $D = 195$  Hz corresponding to the bond length of 2.50 Å. This compares favorably with the X-ray structure (2.486 Å)<sup>[3]</sup>

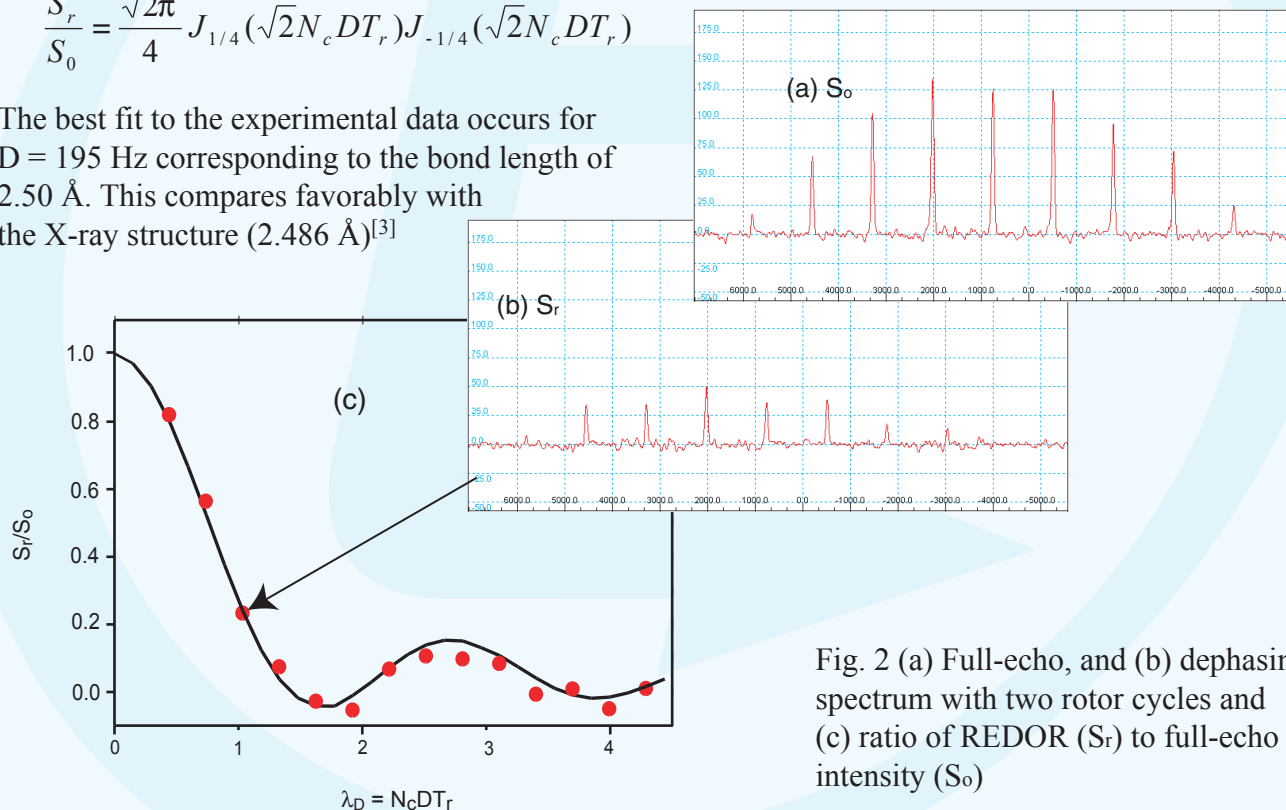


Fig. 2 (a) Full-echo, and (b) dephasing spectrum with two rotor cycles and (c) ratio of REDOR ( $S_r$ ) to full-echo intensity ( $S_0$ )

### 5. References

- (1) Gullion, T., J. Schaefer, *J. Magn. Reson.* **81**, 196 - 200, 1989.
- (2) Gullion, T., *Concepts in Magn. Reson.* **10**, 277-289, 1998.
- (3) Legros, J.-P. A. Kivick, A., *Acta Cryst., Sect. B* **36**, 3052, 1980.