Supporting Information for

Solid-State NMR of Membranous ^{13}C-Labeled Cholesterol

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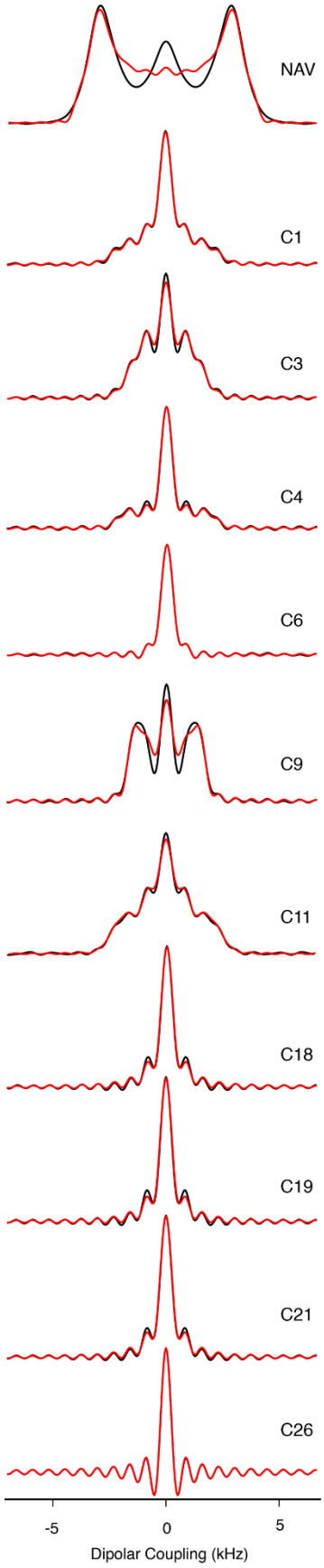

Figure 1S: Representative Dipolar Coupling Spectra

Experimental (black) and simulated (red) ^{1}H-^{13}C dipolar coupling line shapes, acquired at 600 MHz using direct ^{1}H-^{13}C polarization and the $R48_3^{18}$–symmetry recoupling sequence. The line shape for NAV (rigid lattice standard) is shown at the top.

Table 1S: Order Parameters from $R48_3^{18}$ 2D experiments

Average order parameters for various ^{1}H-^{13}C sites along cholesterol including standard deviation. The order parameters are calculated from both DP and CP $R48_3^{18}$ experiments. Our measured order parameters are compared to plotted values (± 0.03) reported by Ferreira et al. in Figure 5D [28]. Note that for CH_2 groups, previous measurements report the largest of the two values.

Site	Average Order Parameter	Standard Deviation	Order Parameters from Ferreira et al. [28]
1a	0.26	0.05	-
1b	0.39	0.07	0.38
3	0.38	0.03	0.39
4a	0.42	0.01	0.35
4b	0.25	0.04	-
6	0.12	0.01	-
9	0.47	0.01	0.44
11a	0.43	0.07	0.45
11b	0.27	0.07	-
18	0.21	0.01	0.12
19	0.20	0.01	0.14
21	0.18	0.02	0.14
26	0.03	0.01	-
27	0.03	0.01	-

^{13}C-labeled isotopomers of C13

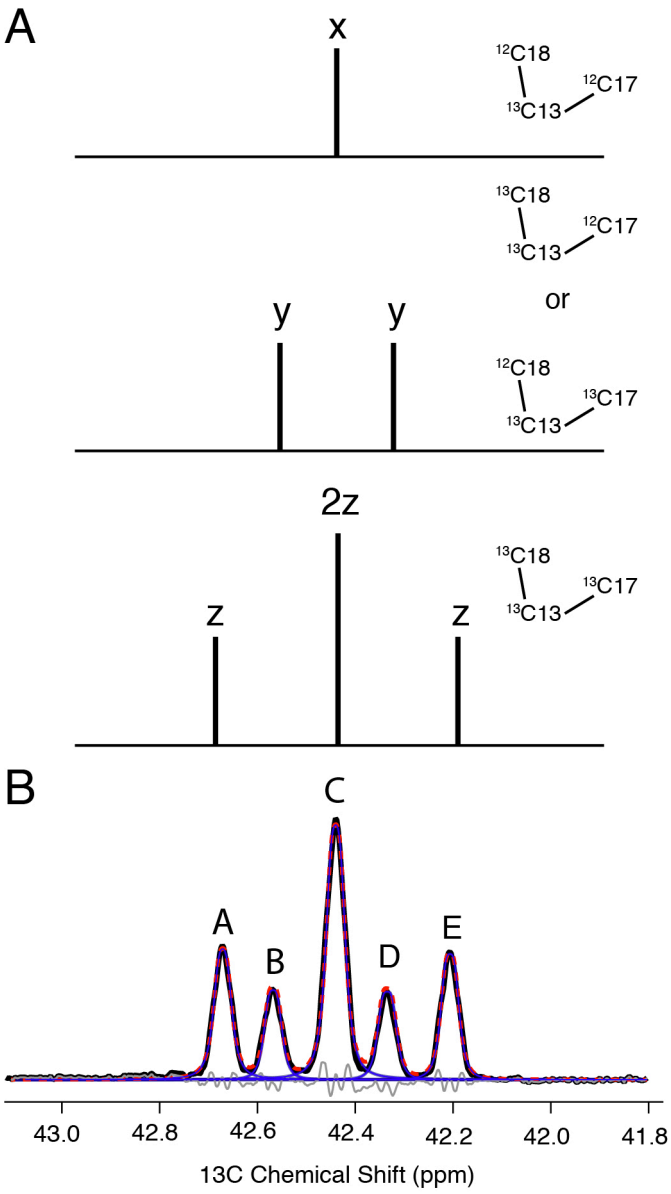

Figure 2S: Multiplet Structure and Analysis of C13

A) Sum of multiplets from various C13 populations resulting in the B) observed multiplet, analyzed by global spectral deconvolution. The experimental data is shown in black, the peaks A-E are drawn in blue, and the sum of the fits are represented by the dotted red line. The residual between the experimental (black) and the sum (dotted red) is shown in grey.

The C13 multiplet from Figure 5D from the main text was used to determine the various 13C-labeled populations of C13. As depicted in Figure 2S below, Peaks B and D represent the splitting which occurs when a labeled C13 is next to either a labeled C17 or C18. Peaks A and E occur when a labeled C13 is next to both a labeled C17 and a C18 molecule. The overall multiplet structure can be represented as the sum of the possible multiplets which occur, shown in part A of Fig 2S. The middle peak C is a sum of 2z + x. Where x represents the amount of labeled C13 next to both 12C C17 and 12C C18. The areas of the peaks were determined using MestreNova's Select Line Fitting Region(s) tool. The breakdown of the analysis is demonstrated below in Table S2. The grey line in Figure 2S represent the residual between the data and the sum (dotted red) of the line fitting (blue) of each peak.

Thus, the following correlations are:

C = x = 2z
B, D = y
A, E = z

Table 2S: Integration values of peaks A-E shown in Figure 2S.

Peak	Area
A	14118.3
B	8702.7
C	28690.6
D	8941.7
E	12674.1

It was then possible to solve for the various populations, in which 3% of the population C13 is not near any labeled C17 and C18, 24% of the population C13 is connected to either a labeled C17 or C18, and 73% of C13 is connected to both a labeled C17 and C18 atoms.