

Dynamic Nuclear Polarization of membrane proteins: covalently bound spin-labels at protein-protein interfaces

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Supporting Information

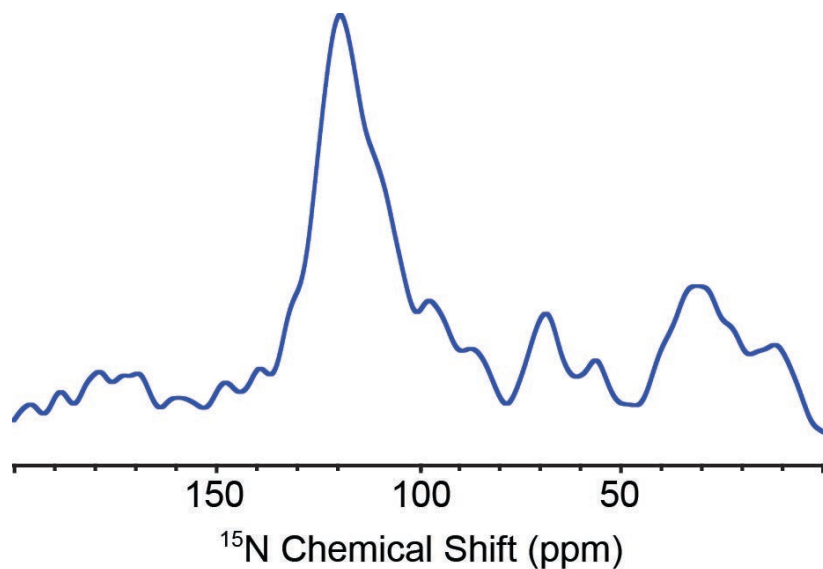


Figure S1. ^{15}N 1D of GALN. Like with most membrane proteins, few sites are cleanly resolved, clearly the presence of these peaks, cross polarized from the same ^1H bath, confirms the efficacy of covalently bound radicals for DNP enhancements in membrane proteins.

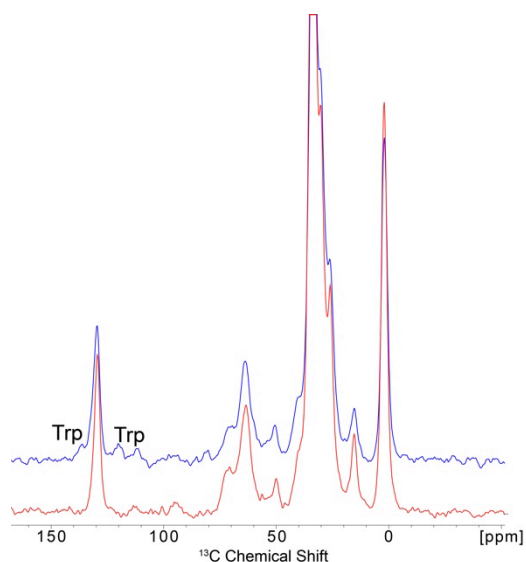


Figure S2. Overlay of ^{13}C 1D spectrum of Sample 1 (1:3.5 GCDL, red) with Sample 4. Sample 4 was prepared with two different gramicidin species, one spin-labeled gramicidin C and the other unlabeled gramicidin D (1:17 GCDL:Lipid, 1:6 gramicidin D:Lipid, blue). Overall the aliphatic regions of each spectrum are roughly identical, yet the aromatic regions are significantly different. In the spectrum of sample 4, aromatic side chain peaks arising from the tryptophan spin systems are cleanly visible, while these same peaks are either broad or non-existent in sample 1. It can be inferred that these peaks arise from the protein without a spin label. These spectra suggest two things: First, while backbone and aliphatic side chain resonances are not significantly perturbed by the presence of covalently bound spin labels, aromatic groups appear to be significantly impacted. Second, the enhancement of signals in the spectrum of sample 4 are equivalent within the signal-to-noise of the spectrum of sample 1,

suggesting a complementary means of polarization enhancement where small spin-labeled molecules are placed into vesicles along with other protein targets.