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Expanding the ^{13}C Observe Toolbox: Introducing "*DEPTQ*"

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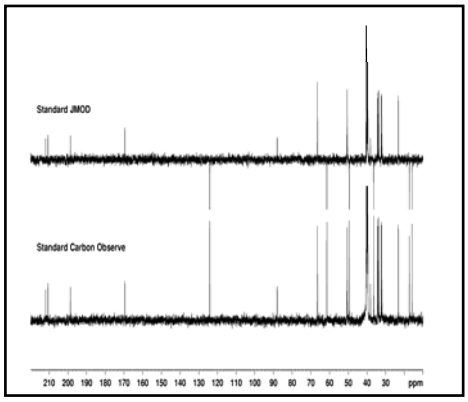
Observation of the ^{13}C nucleus continues to be of paramount importance in the suite of NMR experiments researchers routinely use to characterize their synthetic targets/intermediates. Although I do not have hard numbers, it would be fair to say that a significant portion of the usage of the AVANCE 500 is devoted to observing some form of ^{13}C NMR spectrum. For many of you, this involves a normal ^{1}H decoupled ^{13}C spectrum, sometimes in combination with an edited spectrum such as JMOD and DEPT135 which provides information on the multiplicity of the ^{13}C signals (CH_3, CH_2, etc.). Although considerable sensitivity improvement and therefore time savings can often be achieved by obtaining ^{13}C shifts indirectly via the indirect dimension of 2D experiments like HSQC/HMBC, this approach still remains relatively unpopular for a number of reasons.



Edited ^{13}C spectroscopy permits the recovery of multiplicity information of the carbon signals which is lost in the normal ^{13}C spectrum due to the application of broadband ^{1}H decoupling. One popular editing technique, the JMOD experiment, uses a spin-echo to extract multiplicity information. CH_3 and CH signals end up anti-phase with respect to CH_2 and C_q carbons. However, this information comes at a cost for at least two reasons. Compared to the normal ^{13}C{^{1}H} technique, one will lose some sensitivity in the JMOD due to the proton decoupler being gated off during the spin echo and therefore attenuating the nOe enhancement. Secondly, and more importantly, the JMOD involves the use of a 90º ^{13}C excitation pulse, increasing the demands on the relaxation delay used for magnetization recovery between pulses. The normal ^{13}C{^{1}H} sequence we use on the 500 uses a 30º excitation pulse. In other words, if you are having a hard time seeing C_q carbons in your ^{13}C spectrum, it will be even harder in the JMOD! As an example, the figure on the left shows the ^{13}C spectrum (bottom) and the JMOD spectrum (top) for a 19 mM solution of the steroid cortisone in DMSO-d6 obtained on the 500 with the BBO probe. Each spectrum was obtained using 1000 scans and a total relaxation time (D1+AQ) of 1.1s. Increased sensitivity is apparent throughout the entire ^{13}C spectrum as compared to the JMOD.

A second popular experiment is DEPT, which achieves editing in a completely different manner from JMOD. DEPT involves a magnetization transfer from ^{1}H to ^{13}C and makes use of the fact that the phase and amount of magnetization transferred in the experiment depends upon the multiplicity of the carbon signals involved. By setting the final ^{1}H pulse to 135º, we have DEPT-135 where CH_2 signals are antiphase to CH_3 and CH carbons. A significant advantage over JMOD is that DEPT allows faster data accumulation as the experiment can be recycled based upon the faster (normally) ^{1}H relaxation times rather than ^{13}C. However, a significant disadvantage is that any C_q carbons in the sample are absent in a DEPT experiment, meaning time/sensitivity advantages in DEPT are offset by the need to run a ^{13}C{^{1}H} experiment to obtain C_q carbon chemical shift information.

Burger and Bigler have published an experiment known as DEPTQ, which has the same basic features of DEPT but *extends it to include signals from C_q carbons* (*J. Magn. Reson.* **1998**, 135, 529-534). The DEPTQ-135 experiment gives signal phases identical to the JMOD experiment, but can be recycled based on the ^{1}H relaxation times, making this a strong candidate to replace the single pulse ^{13}C{^{1}H} experiment. If multiplicity ambiguities remain after a DEPTQ-135, a DEPTQ-90 can be run which yields CH and C_q carbon signals only that are antiphase to one another. **Both of these experiments are now available to users of the AV-500 at ARMRC.** The figure on the right shows the DEPTQ-135 experiment run on the 19 mM cortisone sample, using 1000 scans and three different recycle delays (D1); 1s (bottom), 100 ms (middle) and 1 µs (top). The 1s spectrum shows very good sensitivity across the entire spectral range, including the quaternary signals. An interesting feature of this experiment is that as the recycle delay is decreased, all proton-bearing carbon signals become saturated but quaternary signal intensity remains strong. This is clearly evident in the upper 1 µs spectrum where the only signals that remain with any appreciable intensity (excluding solvent) are the 7 quaternary carbon signals of cortisone. This behaviour is contrary to our normal experience where the longer T_1's of quaternary signals often make them the hardest to see, frequently necessitating a lengthening of the recycle delay to measure their chemical shifts. This anomalous DEPTQ behaviour is probably due to the fact that proton nuclei are becoming saturated as the recycle delay shortens, decreasing the magnetization available for dept transfer. Quaternary carbons, on the other hand, are not subject to such a transfer of proton magnetization and will gain some intensity due to long range nOe enhancements during the acquisition time, when ^{1}H decoupling is applied. Subsequently, during D1, this enhancement will be attenuated since the decoupler is gated off, with longer recycle delays meaning greater intensity loss. In summary, **lengthening your acquisition time** (to increase nOe build-up for Cq carbons) and **shortening D1** (to both saturate proton magnetization *and* minimize nOe losses for Cq carbon nuclei) in a DEPTQ experiment appears to be a viable alternative for obtaining quaternary-only carbon sub-spectra.