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Accordion-optimized DEPT experiments

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In this contribution, a pulse sequence is described for recording accordion-optimized DEPT experiments. The proposed ACCORDEPT experiment detects a wide range of one-bond coupling constants using accordion optimization. As a proof of concept, this strategy has been applied to a mesogen containing a large range of one-bond $^1J_{CH}$ coupling constants associated with the various structural elements. The ACCORDEPT experiment afforded significant enhancements for the resonances with the larger $^1J_{CH}$ couplings, similar SNR for aliphatic resonances, but reduced SNR for aliphatic resonances as compared with the standard DEPT experiment. In addition, the ACCORDEPT is straightforward to implement, does not require any supplementary calibration procedures and can be used under automated conditions without difficulty by inexperienced users.

Supporting information may be found in the online version of this article.

Keywords: NMR; ^{13}C; DEPT; accordion optimization; ACCORDEPT; spectral editing

Introduction

With the development of 2D methods, NMR spectroscopy has clearly become a very powerful tool, in particular in chemistry, as evidenced by the high proportion of compound structure elucidation papers in this and other journals that make use of NMR. Nevertheless, multiplicity-dependent 1D ^{13}C-NMR experiments such as INEPT,[1] DEPT[2] or APT[3] still belong to the most common experiments for assigning ^{13}C signals and for the elucidation of molecular structures on a routine level. However, many scientists often overlook the fact that the effectiveness of polarization transfer is directly influenced by the magnitude of the $^1J_{CH}$ coupling constant value during the precession periods. For a given molecular system containing different chemical functionalities with different $^1J_{CH}$ coupling constants, the proper choice of the duration of a fixed delay, Δ, which is inversely related to $^1J_{CH}$, may be challenging if the range of $^1J_{CH}$ is large. Incomplete polarization transfer cannot be avoided in such cases, and maximum polarization transfer cannot be achieved for all CH fragments simultaneously because Δ is in general chosen using a single, average, $^1J_{CH}$ coupling value, typically 145 Hz.

Nowadays, since DEPT experiments are mostly run under automated conditions, this tendency 'set it and forget it' may cause serious sensitivity problems for chemical compounds that possess chemical functionalities like furanyl, aldehydes and alkynes groups. Indeed, these groups have unusually large $^1J_{CH}$ coupling constants, which are far from the average value set for the coherence transfer period. Therefore, in statically optimized DEPT experiments, these specific chemical functionalities will exhibit resonances with extremely reduced intensities, while those having coupling constant values close to the average value set for recording the spectra will exhibit resonances with maximal intensities. Consequently, recording experiments for obtaining an acceptable SNR for all resonances takes a substantial amount of spectrometer time.

There have been several recent efforts to improve the basic DEPT pulse sequence, but the problem of weak responses has not been addressed so far. For instance, the DEPTQ experiment[4,5] allows the detection of quaternary carbons, and the QDEPT experiment provides quantitative data under specific experimental conditions.[6,7] Actually, the QDEPT experiment has been precisely designed to cancel the signal intensity dependence on $^1J_{CH}$ in polarization transfers between ^1H and ^{13}C and could therefore be applied to address the problem of weak responses. However, the possibility of establishing carbon multiplicity is lost, and hence the QDEPT scheme does not appear very useful for routine measurements excepted when quantitative data are needed.

By using the parallel between RF pulses and spin–spin coupling, several groups have devised J-compensated sequences that are less sensitive to variation in $^1J_{CH}$.[8–13] However, it turns out that the major drawback of these J-compensated experiments is that they incorporate more RF pulses and delays than the conventional experiments. This makes them longer in duration and therefore more susceptible to signal loss through relaxation.

Herein, we propose a modified DEPT experiment, ACCORDEPT, which intends to overcome this deficiency of the standard DEPT experiment. During the last two decades, accordion optimization[14] has emerged as an elegant solution to sample a wide range of coupling constants in one experiment. The methodology has been successfully applied to make uniform the response intensity for all proton–carbon pairs in direct and long-range 2D correlation experiments[15–20] and ACCORD–ADEQUATE experiments,[21] and also in experiments that measure spin-lattice relaxation rates,[22] chemical exchange,[23] variable TOCSY spin-lock length,[24] diffusion coefficients,[25] coupling constants[26] and more recently in protein NMR spectroscopy.[27] To the best of our knowledge, however, the application of the accordion

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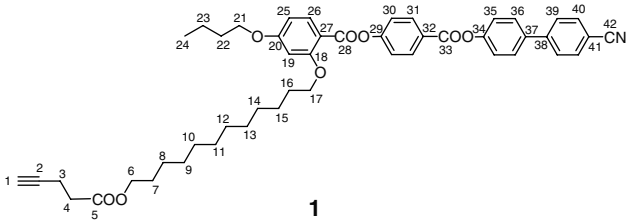

Figure 1. Molecular structure of mesogen **1** and carbon atom numbering.

method to uniform the response intensity over wide ranges of coupling constants has not been exploited for 1D INEPT or DEPT experiments. As proof of the concept, ACCORDEPT experiments of compound **1** (mesogen **1,** Fig. 1) have been recorded and compared to standard optimized DEPT experiments. Compound **1**, which can be used for the design of various mesomorphic materials, was selected as the first sample, since it possesses different chemical functionalities and exhibits a large range of one-bond coupling constants.

Experimental Procedures

Materials

Compound **1** (Fig. 1) was obtained in 80% yield by reacting 4-pentynoic acid with the corresponding alcohol precursor (see structure **4** in Ref. [28]) under standard esterification conditions. MS (ESI, positive mode): (771.38) 794.70 $[M + Na]^+$. Calculated values for $C_{48}H_{53}NO_8$ (771.38): C, 74.68; H, 6.92; N, 1.81; O, 16.58. Experimental values: C, 74.67; H, 6.91; N, 1.89; O, 18.42.

NMR experiments

For all experiments, either a sample of *ca* 15 mg of **1** (Fig. 1) dissolved in 600 µl CD_2Cl_2 or the standard 100 mg/ml $CDCl_3$ cholesteryl acetate sample was used. All NMR experiments were performed on a Bruker Avance II spectrometer operating at a nominal proton resonance frequency of 400.13 MHz, equipped with a 5-mm broadband direct probehead (BBFOplus) with an additional *z*-gradient accessory. For all experiments, the 90° pulse lengths were 9.4 µs for ^1H and 9.3 µs for ^{13}C. The 180° ^{13}C hard pulses were replaced by a pair of BIP 720-100-10 pulses.[29] Uniform inversion over ±10.6 kHz was obtained with a pulse length of 192 µs, which adequately covers the standard ^{13}C spectral widths at 9.4 T. This pulse offers B_1 inhomogeneity compensation of $(B_1/B_1{}^0 = 1 \pm 0.1)$. The power level was adjusted according to a 90° rectangular pulse of 24 µs. All experiments were conducted at 298 K using 32 K points and a relaxation delay of 2 s. The spectral width was set to 18 116 Hz, leading to an acquisition time of 0.9 s. The data sets were zero-filled to 64K points and multiplied by a 1-Hz line broadening factor before Fourier transform. The $^1J_{CH}$ coupling constant values for **1** have been measured using a CLIP–HSQC experiment.[30]

Results and Discussion

As an illustrative example, DEPT135 spectra of **1** are shown in Fig. 2. The experiments have been optimized for a $^1J_{CH}$ coupling constant of 145 Hz (A), 190 Hz (B) and 250 Hz (C). A first inspection of the DEPT (A) optimized for $^1J_{CH} = 145$ Hz reveals that the terminal acetylenic carbon C-1 is hardly visible. This signal remains difficult to detect since the $^1J_{CH}$ coupling constant for this terminal acetylenic group is 251 Hz, while the DEPT experiment has been optimized for a $^1J_{CH}$ coupling constant of 145 Hz. The DEPT135 spectrum (A) of molecule **1** shown in Fig. 2 is a good demonstration that chemical functionalities having unusual $^1J_{CH}$ coupling constants values that are far from the average value set in the DEPT experiment can remain barely discernible, even if the experiment is recorded with a large number of transients. (B) and (C) are the DEPT spectra when the delay Δ is optimized for coupling constant values of 190 and 250 Hz, respectively. A rapid inspection of these spectra reveals that these optimizations are far from ideal. While the aromatic and acetylenic resonances emerge with an acceptable SNR, the intensity of the aliphatic resonances remains overall weak, and some of the resonances are even missing. This is not surprising, since in terms of coupling evolution, the aliphatic resonances experience a 180° phase shift.[31]

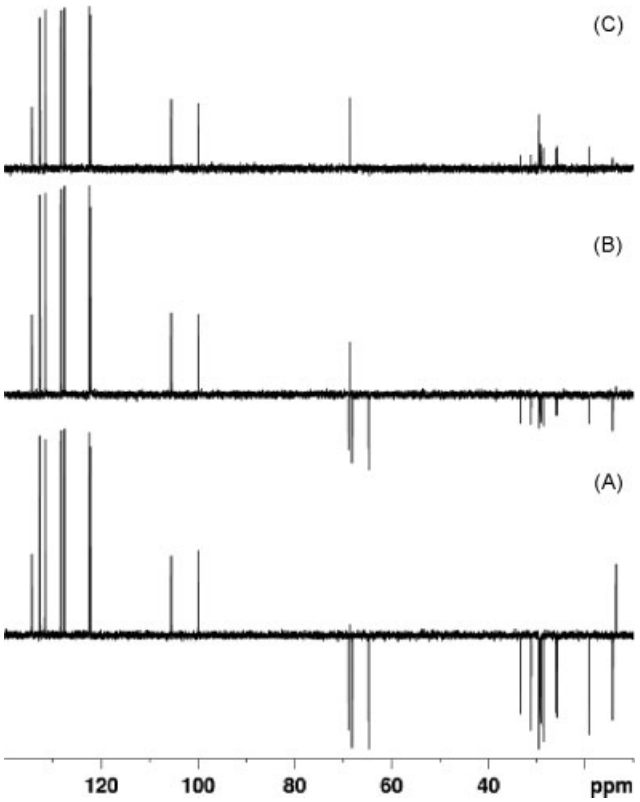

Figure 2. DEPT135 spectra of compound **1** dissolved in CD_2Cl_2: (A) optimized for a $^1J_{CH}$ coupling constant of 145 Hz; (B) optimized for a $^1J_{CH}$ coupling constant of 190 Hz; (C) optimized for a $^1J_{CH}$ coupling constant of 250 Hz. Other experimental parameters are given in the experimental part.

The DEPT pulse sequence we propose for equalizing signal intensities over wide ranges of coupling constants is shown in Fig. 3. This accordion DEPT, for which we gave the acronym ACCORDEPT, is a straightforward modification of the original DEPT pulse sequence. Instead of statically optimized Δ delays, the ACCORDEPT experiments contain delays *vd* which can be decremented from Δ_{max} to Δ_{min}, incremented from Δ_{min} to Δ_{max} or set as a random list. The rectangular ^{13}C inversion pulse is replaced by a pair of BIP pulses,[29] which are much shorter than adiabatic inversion pulses and possess superior inversion performance and high tolerance to B_1 field inhomogeneity.[29]

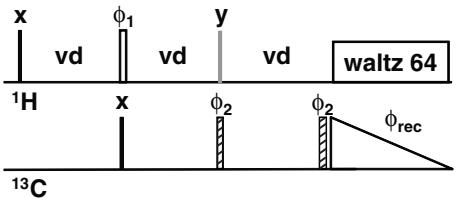

Figure 3. Pulse sequence for the ACCORDEPT experiment incorporating the accordion delays, labeled *vd*, derived from a standard DEPT pulse sequence.[4] These delays can be decremented or incremented from a value of $^1J_{min}$ (inverse of twice the smallest $^1J_{CH}$ coupling) to $^1J_{max}$ (inverse of twice the largest $^1J_{CH}$ coupling), or set as a random list. A pair of 192 µs BIP 720-100-10 pulses[20] for ^{13}C refocusing has been used instead of the classical 180° rectangular pulse.

When used for refocusing transverse magnetization, the BIPs must be applied in pairs, like adiabatic pulses.[32] The phase error induced by the first BIP is then exactly compensated by that of the second, leading to optimal performances.[33–35] The rest of the sequence is identical to the original DEPT pulse sequence.[2]

For a CH group and a standard DEPT135 experiment, the signal intensity J dependence of polarization transfers between ^1H and ^{13}C nuclei, I^{DEPT}, can be described by Eqns (1–3),[31,36,37]:

$$I_{CH}^{DEPT} = (\sqrt{2}/2)\sin^2(\pi\,\Delta J_{CH}) \quad (1)$$

$$I_{CH_2}^{DEPT} = (\sqrt{2}/4)\sin^2(2\pi\,\Delta J_{CH}) - \sin^4(\pi\,\Delta J_{CH}) \quad (2)$$

$$I_{CH_3}^{DEPT} = \frac{3}{4}\Big[\sqrt{2}\sin^6(\pi\,\Delta J_{CH}) + (\sqrt{2}/2)\sin^2(2\pi\,\Delta J_{CH})\cos^2(\pi\,\Delta J_{CH}) - \sin^2(2\pi\,\Delta J_{CH})\sin^2(\pi\,\Delta J_{CH})\Big] \quad (3)$$

where Δ is the evolution delay. Protons are assumed equivalent and the homonuclear couplings as well as relaxation effects are ignored.

Similarly, Eqns (4–6) govern the average intensities $I^{ACCORDEPT}$ of the signal assuming an ACCORDEPT experiment:

$$I_{CH}^{ACCORDEPT} = \frac{1}{n}\sum_{i=1}^{n}(\sqrt{2}/2)\sin^2(\pi\,\Delta_i J_{CH}) \quad (4)$$

$$I_{CH_2}^{ACCORDEPT} = \frac{1}{n}\sum_{i=1}^{n}(\sqrt{2}/4)\sin^2(2\pi\,\Delta_i J_{CH}) - \sin^4(\pi\,\Delta_i J_{CH}) \quad (5)$$

$$I_{CH_3}^{ACCORDEPT} = \frac{1}{n}\sum_{i=1}^{n}\frac{3}{4}\Big[\sqrt{2}\sin^6(\pi\,\Delta_i J_{CH}) + (\sqrt{2}/2)\sin^2(2\pi\,\Delta_i J_{CH})\cos^2(\pi\,\Delta_i J_{CH}) - \sin^2(2\pi\,\Delta_i J_{CH})\sin^2(\pi\,\Delta_i J_{CH})\Big] \quad (6)$$

where n is the number of values used for sampling the chosen coupling constant range and Δ_i the evolution delay for the corresponding ith value of that coupling range.

The aim of the ACCORDEPT experiment is to afford data with improvement of the SNR for responses exhibiting significantly different couplings from the optimization of the static experiment. For this purpose, we first focus on the sampling technique of the accordion delay.

In Fig. 4, theoretical curves for a DEPT135 experiment and variable delay Δ are shown. Figure. 4(A), (C) and (E) has been obtained by varying the delay Δ on the basis of equal steps in time and Fig. 4(B), (D) and (F) by varying the delay Δ on the basis of equal steps in frequency (hertz). The nonlinear sampling of the desired coupling range is clearly apparent in Fig. 4(A), (C) and (E), where 50% of the values cover the range 120–164 Hz, and 50% the range 164–260 Hz, as a result of equal *time* between decrementation steps, but not equal in frequency. In the following, for clarity, the decrementation technique on the basis of equal steps in hertz will be denoted by DCC and the decrementation technique on the basis of equal steps in time by DCT.

Figure 4(A) and (B) illustrates the theoretical curves for a CH_3 group ($^1J_{CH} = 120$ Hz), Fig. 4(C) and (D) for a CH_2 group ($^1J_{CH} = 140$ Hz) and Fig. 4(E) and (F) for a CH group ($^1J_{CH} = 250$ Hz). From these curves, it can be seen that the intensity of a CH_3 group drops rapidly as the mismatch between the actual coupling constant value and the value of Δ grows [Fig. 4(A) and (B)]. The intensity reaches only 0.3 when Δ is matched for $^1J_{CH} = 164$ Hz and remains below this value for larger values of Δ. The intensity of a CH_2 group also drops as the mismatch between the actual coupling constant value and the value of Δ grows. Interestingly, when Δ is optimized for large coupling constants (>240 Hz), the curves predict that the intensity of a CH_2 group becomes positive, thus indicating a wrong multiplicity in a DEPT135 spectrum. Therefore, an ACCORDEPT experiment optimized for the range 115–260 Hz will exhibit CH_2 groups with a considerably decreased SNR, since the experiments optimized for larger couplings destroy the magnetization previously built up during the experiments optimized for smaller couplings. Finally, Fig. 4(E) and (F) reveals that the intensity of a CH group with a coupling constant of 250 Hz increases as a monotonic function with the value of the delay Δ. Interestingly, the curves predict a 'plateau' starting from 230 Hz. According to these curves, it should thus be appropriate to use the range 120–230 Hz for sampling ACCORDEPT experiments.

It can be seen from the different curves that the difference between the two decrementation techniques can be significant. The average signal intensities can be calculated from Eqns (4–6) and the average values are indicated in the respective figure parts by horizontal dashed lines. Figure 4(A) and (B) reveals that the average intensity is only 0.27 for a CH_3 group (assuming a one-bond coupling $^1J_{CH}$ of 120 Hz) and considering the DCC method. When the delays Δ are decremented using the DCT method, this average intensity increases to 0.43. Likewise, Fig. 4(C) and (D) shows that the average intensity is −0.52 for a CH_2 group (assuming a one-bond coupling $^1J_{CH}$ of 140 Hz) and considering the DCC method. When the DCT method is used, the average intensity increases to −0.66. Finally, Fig. 4(E) and (F) shows that the average intensity is 0.46 for a CH group (assuming a one-bond coupling $^1J_{CH}$ of 250 Hz) and considering the DCC method. When the DCT method is used, this average intensity decreases to 0.34. On the other hand, if the range 120–230 Hz is used for sampling ACCORDEPT experiments, the values become 0.30 (CH, $^1J_{CH} = 250$ Hz), −0.74 (CH_2, $^1J_{CH} = 140$ Hz) and 0.47 (CH_3, $^1J_{CH} = 120$ Hz) for the DCT method, and 0.39 (CH, $^1J_{CH} = 250$ Hz), −0.65 (CH_2, $^1J_{CH} = 140$ Hz) and 0.34 (CH_3, $^1J_{CH} = 120$ Hz) for the DCC method. From these simple calculations, and in contrast to accordion-optimized HSQC experiments,[19] it is not obvious to define a decrementation technique, DCT or DCC, which uniforms at best the polarization transfer efficiencies.

However, it should be stated that the DCC method is somewhat laborious to use. Indeed, for obtaining equal steps in hertz, delays Δ that are decremented from Δ_{max} to Δ_{min} in steps of $\{0.5/[n_i.(^1J_{CHmax} - {}^1J_{CHmin})]\}$ must be used, which imply that the

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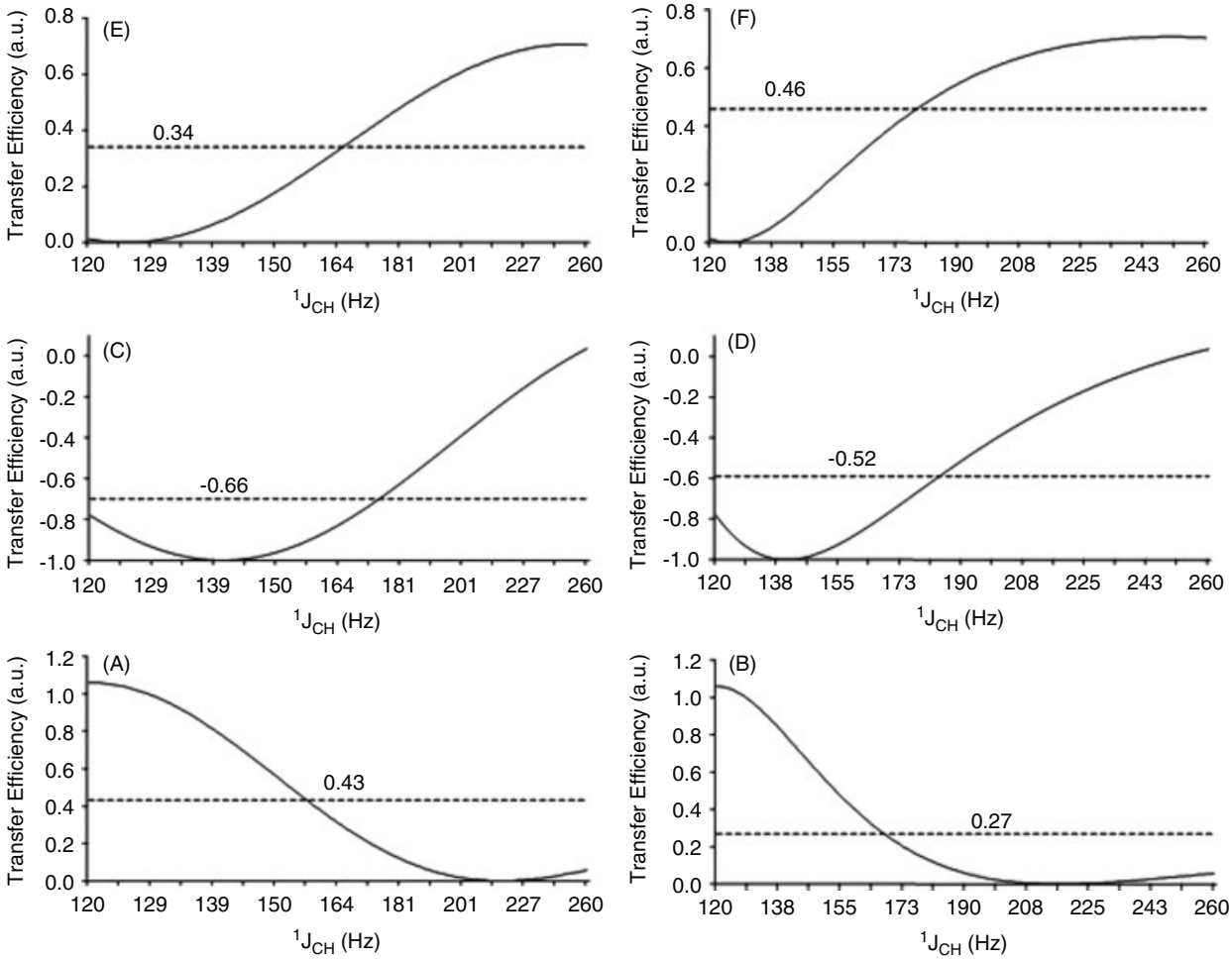

Figure 4. Theoretical curves for ACCORDEPT135 and variable delay Δ for (A), (B) a CH_3 group ($^1J_{CH}$ = 120 Hz); (C), (D) a CH_2 group ($^1J_{CH}$ = 140 Hz); (E), (F) a CH group ($^1J_{CH}$ = 250 Hz). For clarity, the *x*-scales have been labeled according to the corresponding heteronuclear coupling $^1J_{CH}$ (Δ is the inverse of twice the heteronuclear coupling $^1J_{CH}$). Equations (1–3) have been used for plotting the curves.[28] All curves have been obtained using the range 120–260 Hz. In (A), (C) and (E), the theoretical curves were obtained using the DCT method. In (B), (D) and (F), the theoretical curves were obtained using the DCC method. The dashed horizontal lines materialize the average polarization transfer efficiency, calculated using Eqns (4–6).

division in time decreases successively. Such a method cannot be easily implemented in pulse programs and the user is compelled to use external and specific lists of delays Δ, which have to be generated using external programs. Since the only experimental parameter that must be manually inputted before launching the DCT method is the $^1J_{CH}$ coupling range of interest, the ACCORDEPT using the DCT method can be used without difficulty by inexperienced users and under automated conditions.

Obviously, homonuclear coupling evolution during the pulse sequence as well as relaxation effects affect the polarization transfer to the desired coherence and therefore affect the final SNR. Homonuclear coupling evolution is active in the DEPT experiment during all three evolution periods Δ. However, it was shown for a 2D-INEPT experiment that if J_{HH} does not exceed 10% of J_{CH}, which clearly exceed typical values of $^3J_{HH}$ versus $^1J_{CH}$, the intensity loss remains negligible.[38] Conversely, if the transverse relaxation T_2 is very efficient, which is always the case for large molecules, the magnetization can significantly attenuate between the first pulse and the beginning of the acquisition. For a rigorous treatment of the dynamics of the spin system during an ACCORDEPT experiment, an elaborate mathematical treatment should be used. However, the attenuation of magnetization $I_{x,y}$ during the DEPT sequence can be reasonably approximated using the Bloch equations as follows:

$$I_{x,y} = \exp(-t_{ps}/T_2) \quad (7)$$

where t_{ps} is the time of magnetization on the transverse plane, during which T_2 relaxation takes place (roughly the time between the first pulse and the beginning of the acquisition time). The curves presented in Figs. S1 and S2 show that the attenuation can be significant. However, the profiles of the curves are preserved, and therefore the general conclusions drawn above when relaxation phenomenons are neglected still apply when relaxation is considered.

In accordion-optimized 2D sequences,[15–20] the number of increments used for sampling the selected range of $^1J_{CH}$ coupling constants depends on the number of t_1 increments used for sampling the indirect dimension. In contrast, for 1D ACCORDEPT experiments, the number of increments can be freely selected, since a 1D experiment can be repeated at will and the accumulated data automatically summed. We have thus investigated the influence of the number of increments for sampling the selected $^1J_{CH}$ coupling constant range on the

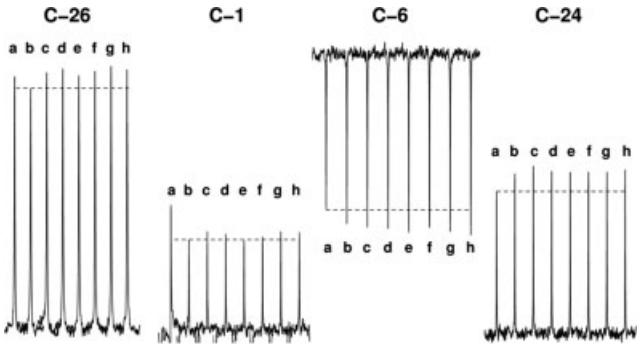

Figure 5. Signal intensities obtained from ACCORDEPT135 spectra for C-26 (CH, $^1J_{CH}$ = 166 Hz), C-1 (CH, $^1J_{CH}$ = 251 Hz), C-6 (CH_2, $^1J_{CH}$ = 133 Hz) and C-24 (CH_3, $^1J_{CH}$ = 124 Hz) of mesogen **1** dissolved in CD_2Cl_2. For all experiments, the range of $^1J_{CH}$ coupling constants sampled is 120–230 Hz. All spectra have been recorded using 512 scans, and using respectively 4 (a), 8 (b), 16 (c), 32 (d), 64 (e), 128 (f), 256 (g), and 512 values (h) for sampling the coupling range. All other experimental parameters are given in the experimental part. The ^{13}C chemical shifts of **1** are given in the Supporting Information.

signal intensity. To this end, ACCORDEPT spectra of **1** have been recorded using the DCT method and using 4, 8, 16, 32, 64, 128, 256 and 512 values – Figure 5(a)–(h), respectively – for sampling the coupling constant range. The signal intensities obtained for C-26 (CH, $^1J_{CH}$ = 166 Hz), C-1 (CH, $^1J_{CH}$ = 251 Hz), C-6 (CH_2, $^1J_{CH}$ = 133 Hz) and C-24 (CH_3, $^1J_{CH}$ = 124 Hz) are shown, in order to encompass most of the functionalities that can be found in chemical compounds. It can be seen from these spectra that the intensity difference remains marginal and that virtually any number of values for sampling the selected coupling range can be envisaged. However, phase distortions, particularly evident for the C-1 resonance, occur when the experiment is recorded using four values. Therefore, we recommend using at least eight values for sampling the selected $^1J_{CH}$ coupling range.

To confirm the ability of accordion optimization to provide resonances with more equalized intensities, ACCORDEPT135 spectra of **1** using different conditions have been recorded and compared. For comparison, a DEPT135 spectrum optimized for $^1J_{CH}$ = 145 Hz is also shown in spectrum (A) of Fig. 6. In spectrum (B), the ACCORDEPT spectrum has been obtained using the DCT method while spectrum (C) has been obtained using the DCC method. A short examination of these three spectra demonstrates that the accordion technique is able to provide all carbons resonances with a satisfactory SNR. In agreement with the theoretical curves shown in Fig. 4, the CH_2 and CH_3 groups are more intense when the DCT method is used, while the acetylenic C-1 resonance is more intense when the DCC method is used.

Table 1 summarizes the results of integrating four selected signals of molecule **1** for a DEPT135 optimized for $^1J_{CH}$ = 145 Hz, a DEPT135 optimized for $^1J_{CH}$ = 190 Hz, a QDEPT experiment and for four ACCORDEPT135 experiments, optimized for the range 120–230 and 120–260 Hz, respectively, and using the DCT and DCC methods, respectively. Compared to a standard DEPT experiment, the results corroborate that ACCORDEPT experiments allow substantial saving in measurement time for molecules possessing large ranges of one-bond coupling constants. For instance, the SNR of the acetylenic carbon C-1 resonance remains very weak (SNR of 4.0) for the standard DEPT experiment optimized for $^1J_{CH}$ = 145 Hz, increases to 15.0 for the DEPT experiment optimized for $^1J_{CH}$ = 190 Hz and, depending on the coupling range selected, reaches fairly good values between 9.1 and 11.2 for

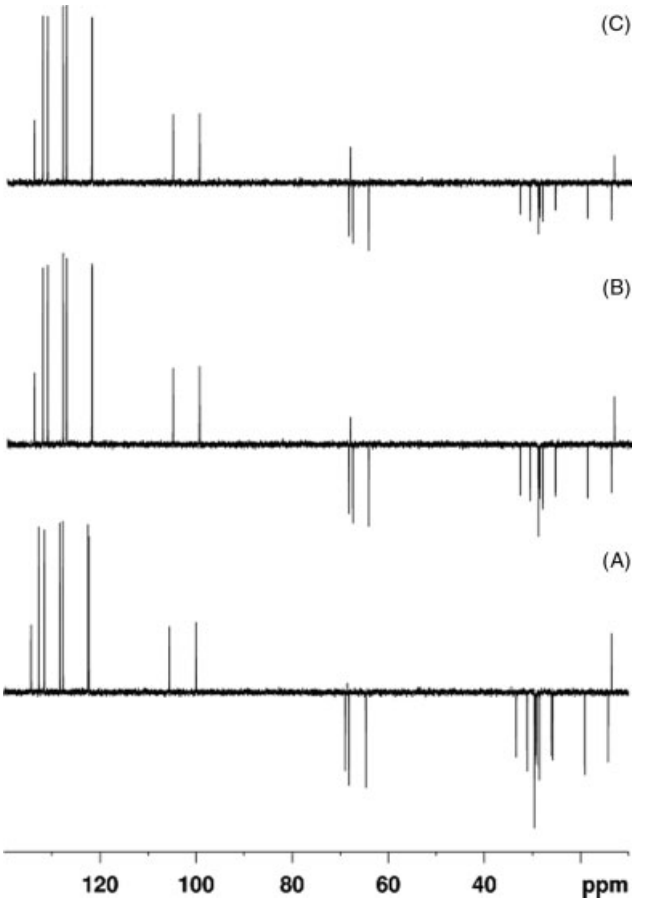

Figure 6. DEPT135 and ACCORDEPT135 spectra of molecule **1** dissolved in CD_2Cl_2. (A) DEPT135 optimized for a $^1J_{CH}$ coupling constant of 145 Hz. (B) ACCORDEPT135 recorded using the DCT method. (C) ACCORDEPT135 recorded using the DCC method. For both ACCORDEPT, the range of $^1J_{CH}$ coupling constants sampled is 120–260 Hz. It has been sampled using 32 values equally spaced in time or frequency. Other experimental parameters are given in the experimental part.

both accordion optimizations. Table 1 also provides the minimal number of acquisitions required for obtaining a SNR of at least 10 for the four selected resonances. To fulfill this condition, a DEPT experiment optimized for $^1J_{CH}$ = 145 Hz would require 900 transients, which represents a total experimental time of about 45 min. The ACCORDEPT experiment recorded using the DCT method allows reducing the number of scans to 131 and 174, and the DCC method to 174 and 115, respectively. This represents a total experimental time around 7 and 9 min. In comparison, the QDEPT scheme[6] would require 133 scans (about 7 min) to obtain SNR of at least 10 for the four selected resonances. However, the possibility of establishing carbon multiplicity is lost when using the QDEPT scheme, which is in essence the main attribute of DEPT experiments.

Finally, it is worth noting that the proposed ACCORDEPT experiment can also be applied to molecules containing chemical functionalities that exhibit narrower $^1J_{CH}$ ranges. In Fig. 7, a standard DEPT experiment optimized for $^1J_{CH}$ = 145 Hz [spectrum (A)] recorded on a sample of cholesteryl acetate dissolved in $CDCl_3$ is compared to an ACCORDEPT optimized for the range 120–160 Hz [spectrum (B)]. It can be seen from these spectra that the overall intensity difference remains insignificant. In (C), an ACCORDEPT optimized for the range 120–230 Hz has

Table 1. SNR calculated for a DEPT135 (optimized for $^1J_{CH}$ = 145 Hz) spectrum, a QDEPT spectrum, and for ACCORDEPT135 spectra for four selected resonances. The SNR values have been obtained using the macro 'sino' provided with the program Topspin (Bruker). The same spectral region (150–160 ppm) was used for estimating the noise. All experiments were acquired and processed with identical parameters described in the experimental section. The $^1J_{CH}$ values are obtained from a CLIP–HSQC experiment.[30] The number of scans required for obtaining an SNR of at least 10 for all resonances is also indicated, given that the SNR values shown have been obtained using 144 scans (the QDEPT scheme must be recorded using multiples of 48 scans).[6] ACCORDEPT spectra have been sampled using 16 values equally spaced in time or frequency

		Signal-to-noise ratios						
Atom number	$^1J_{CH}$ (Hz)	DEPT (145 Hz)	DEPT (190 Hz)	QDEPT	ACCORDEPT (120–230 Hz) DCT	ACCORDEPT (120–230 Hz) DCC	ACCORDEPT (120–260 Hz) DCT	ACCORDEPT (120–260 Hz) DCC
C-1 68.6 ppm	251	4.0	15.0	10.4	10.5	10.2	9.1	11.2
C-26 134.3 ppm	162	34.0	36.4	21.4	33.7	34.0	29.4	29.3
C-6 33.4 ppm	133	34.1	14.0	27.7	22.7	20.1	16.5	14.3
C-24 13.6 ppm	124	29.1	2.7	31.9	18.3	13.3	15.3	11.9
Number of scans[a]		900	1975	133	131	138	174	115

[a] Number of scans required for obtaining a SNR of at least 10 for the four resonances.

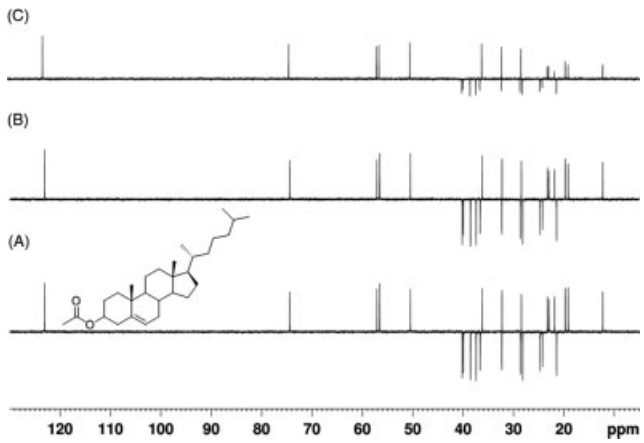

Figure 7. DEPT135 and ACCORDEPT135 spectra of cholesteryl acetate dissolved in $CDCl_3$. (A) DEPT135 optimized for a $^1J_{CH}$ coupling constant of 145 Hz. (B) 120–160 Hz ACCORDEPT135 using the DCT method. (C) 120–230 Hz ACCORDEPT135 using the DCT method. All spectra have been recorded using 32 scans. Other experimental parameters are given in the experimental part.

been added for comparison purposes. This spectrum exhibits aliphatic resonances that are significantly attenuated, as a result of the non-optimal accordion optimization. This would represent an experiment that could be used by default all the time, without modification. However, except for the cases in which the compound under study is completely unknown, we believe that the use of a 'standard' ACCORDEPT experiment is not the best strategy. Indeed, these days, the synthesized or extracted compounds are known, at least partly, and it is therefore fairly unproblematic to estimate the range of $^1J_{CH}$ couplings that should be sampled. Since the sampling of the coupling constant range can be very easily modified, either manually or in automation mode, we strongly advice against the use of a single, standard ACCORDEPT experiment.

Conclusions

Standard DEPT experiments have the drawback of being optimized for only a single $^1J_{CH}$ coupling constant. For molecules possessing small ranges of $^1J_{CH}$ coupling constants, this limitation can be ignored, but when chemical functionalities are present with a broad range of $^1J_{CH}$ coupling constants, the responses at the extremes of the coupling range will suffer in intensity or may not be observed at all. Here, we have proposed the ACCORDEPT experiment to overcome the problem of weakly observed resonances in standard optimized DEPT experiments. The usefulness of the proposed ACCORDEPT experiment has been verified for mesogen 1 that contains different chemical functionalities with a broad range of $^1J_{CH}$ coupling constants. While for standard DEPT experiments, the responses at the extremes of the coupling range suffer in intensity or cannot be detected, ACCORDEPT experiments provide spectra with improved SNR for the responses at the extremes of the $^1J_{CH}$ coupling constant range. However, as accordion optimization always provide better and worse response intensity over standard optimization, the SNR for some responses will correspondingly decrease compared to a standard optimized DEPT experiment. We have also shown that the standard DEPT experiment can be replaced by the ACCORDEPT experiment for routine situations, provided that the $^1J_{CH}$ coupling range is suitably selected. Finally, the ACCORDEPT experiment is straightforward to implement, does not require any supplementary calibration procedures and can be used under automated conditions without difficulty by inexperienced users. In the light of the results presented in this study, we believe that the ACCORDEPT experiment represents a valuable alternative to standard DEPT experiments, and that it could become a useful tool in NMR spectroscopy for analyzing molecular compounds.

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Supporting information

Supporting information may be found in the online version of this article.

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