Selective Measurement of Heteronuclear $^{1}H-^{13}C$ Dipolar Couplings in Motionally Heterogeneous Semicrystalline Polymer Systems

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A pulse sequence for the selective recoupling of heteronuclear dipolar interactions in mobile amorphous phase of powdered semicrystalline polymers is described. $^{1}H-^{13}C$ dipolar interactions are selectively measured by PISEMA-type sequence. Selection of ^{13}C magnetization originating from amorphous phase is achieved by a train of saturation pulses followed by a short delay and a direct excitation pulse on ^{13}C spins. The development of undesired net ^{13}C magnetization during the recoupling sequence is prevented by the efficient "reverse" $^{13}C \rightarrow {}^{1}H$ cross-polarization. The efficacy of the 2D method to measure $^{1}H-^{13}C$ dipolar couplings selectively for mobile components is demonstrated on powdered crystalline L-alanine, semicrystalline polyethylene, and nanocomposite polyamide-6/montmorillonite.

Introduction

Up to now various techniques have been developed to measure the strength of heteronuclear dipolar couplings. For static or quasi-static samples the polarization inversion spin exchange at the magic angle (PISEMA)[1] is widely used to probe structure of membrane-bound proteins. This sequence combining frequency- and phase-switched Lee−Goldburg (LG) homonuclear decoupling[2] provides significantly better resolution of dipolar spectra compared with traditional two-dimensional (2D) separated-local-field (SLF) NMR techniques.[3] However, for powdered samples the dipolar spectra of individual sites must be measured at high-speed MAS to suppress large chemical shift anisotropy (CSA). Unfortunately, the standard PISEMA does not work because the fast MAS attenuates the dipolar couplings. That is why the recoupling techniques must be employed. Rotor synchronized experiments such as DIPSHIFT,[4] however, do not yield detailed information because the time (t_1) data produce only a broad envelope of the dipolar-coupling pattern. Much better results are provided by the rotor-asynchronous sideband version of Lee−Goldburg cross-polarization (LG-CP).[5] High-quality dipolar data were obtained by sensitivity-enhanced modification known as PILGRIM (phase inverted LG recoupling under MAS).[6] Other improvement is provided by the sideband version of PISEMA technique with well-defined amplitude modulation of one of the two radio frequency fields.[7] In contrast to the conventional PISEMA, heteronuclear recoupling is retained by an alternation of the Hartmann−Hahn matching condition between $n = +1$ and $n = -1$ sidebands synchronized with the phase switching. The produced well-resolved dipolar spectra have been recently analyzed with respect to internuclear distances, angles between interatomic vectors in multiple spin-systems[8,9] as well as to investigate amplitude and geometry of local segmental motion.[6]

These techniques thus provide exciting possibilities to obtain highly resolved site-specific dipolar profiles probing structure and dynamics of powdered materials. At present, synthetic polymer systems such as nanocomposites exhibiting unexpected motional behavior attract high scientific interest due to an apparent lack of simple relations between their material properties and composition. In some cases, large interfacial area produced by nanoparticles leads to apparently counterintuitive changes in thermo-mechanical properties.[10] That is why the investigation of the fine relations between segmental dynamics and modified material properties affecting applicability of the polymer systems became a subject of high relevance.

Unfortunately, the polymer nanocomposites are often semicrystalline systems producing complex and overlapped NMR spectra with a poor dispersion of NMR signals in isotropic dimension. Only in rare cases (e.g., polyethylene, PE) are the signals reflecting amorphous and crystalline components well separated. As segmental dynamics of the polymer chains in crystallites and in amorphous phase significantly differ, the limited spectral resolution leads to the detection of dipolar profiles reflecting wide range of motionally averaged dipolar couplings. From such series of overlapped Pake's doublets,[11] individual components can be hardly separated. Consequently, the accurate analysis of segmental dynamics requires measurement of the dipolar spectra for each polymer segment separately in amorphous and crystalline phases. This requirement becomes more clear especially when we realize that changes in molecular dynamics affecting thermomechanical properties occur in amorphous phase rather than in crystallites.

In this contribution, we present a PISEMA-based pulse sequence modified to achieve selective measurement of dipolar couplings in amorphous phase. The efficiency of coherence selection and recoupling of heteronuclear dipolar couplings is documented on several systems. Crystalline alanine with a rigid CH group and rapidly rotating methyl group provides the simplest model. Further, the pulse sequence was tested on semicrystalline polyethylene (PE) and semicrystalline nanocomposite polyamide-6/montmorillonite. Reliability of the proposed concept is confirmed by a simple technique modified to selectively detect dipolar spectra of crystalline (rigid) components. According to our best knowledge, such an experi-

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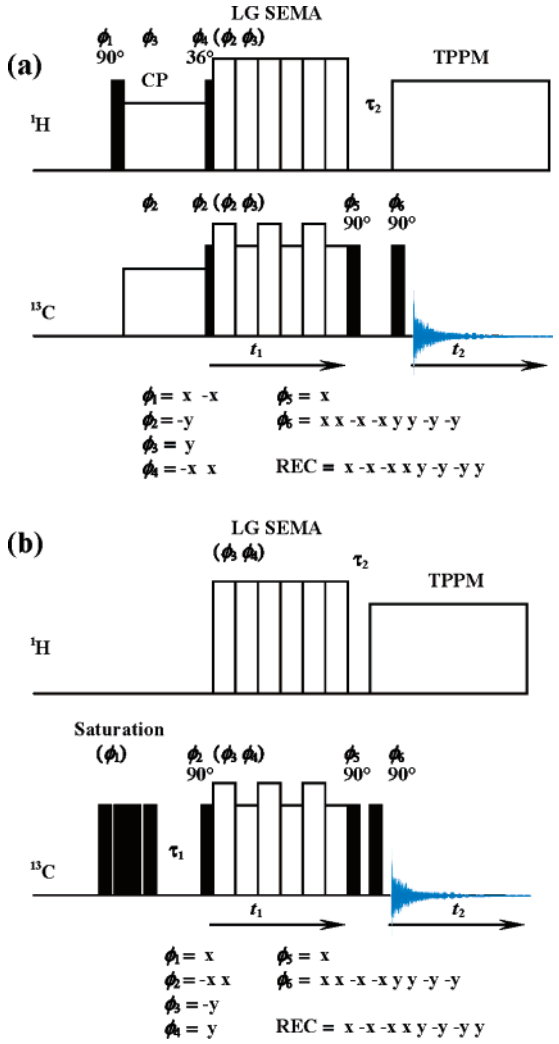
SCHEME 1: Schematic Representation of the Applied Pulse Sequences: (a) Basic 2D PISEMA Experiment[7] Modified by Flip-Back and Read-Out Pulses To Detect Magnetization of Crystalline Phase Only; (b) 2D PISEMA Experiment Modified by a Train of Saturation Pulses and Reverse $^{13}C \rightarrow {}^{1}H$ Cross-Polarization To Detect Magnetization of Amorphous Phase (Direct Polarization SEMA with Presaturation; DP−SEMA)



mental procedure has not been presented yet and provides an efficient probe of segmental dynamics in complex polymer systems.

Pulse Sequence

Borrowing basic ideas for the selection of carbon magnetization of either rigid or mobile component we examined the amplitude-modulated PISEMA[7] experiment. Selective detection of $^{13}C-\{^{1}H\}$ dipolar spectra reflecting heteronuclear dipolar couplings in rigid (crystalline) fraction of the sample is easily achieved by Torchia's approach.[12] Using a sufficiently long delay (τ_2) between flip-back and read-out 90°(^{13}C) pulses, typically 1−3 s, we get ^{13}C magnetization of the amorphous phase completely relaxed (Scheme 1a) so that net crystalline phase is reflected by the dipolar spectra.

In contrast, much more preconditions must be fulfilled to selectively detect $^{13}C-\{^{1}H\}$ dipolar spectra of the mobile (amorphous) segments. First, magnetization originating from the rigid component must be suppressed retaining only ^{13}C magnetization of the amorphous phase before the reintroduction of dipolar interactions. Second, no additional ^{13}C magnetization must be created by the cross-polarization (CP) during the recoupling period. In the opposite case, the cross-polarization from ^{1}H to ^{13}C nuclei occurring during the recoupling sequence (LGSEMA) creates net ^{13}C magnetization of the crystalline component and simultaneously recoupled dipolar oscillation distorts the required selective spectra.

To suppress ^{13}C magnetization of the crystalline phase, the initial cross-polarization block was replaced by a train of 90°-(^{13}C) pulses separated by short delays (0.05−0.1 s). This train effectively saturates transitions of ^{13}C spins in both crystalline and amorphous phases (Scheme 1b). According to the T_1 relaxation time, the following delay τ_1 (0.4−2 s) brings ^{13}C magnetization of the amorphous phase back to the z direction while the magnetization of crystalline components remains suppressed. Next the 90°(^{13}C) pulse excites magnetization of amorphous phase only. During the subsequent recoupling sequence $^{13}C-^{1}H$ dipolar interactions are reintroduced back by amplitude modulation of $B_1(^{13}C)$ field, which is synchronized with the phase switching as proposed by Dvinskikh et al.[7] The Hartmann−Hahn matching condition is fulfilled for ±1 sidebands ($\omega_{eff,H} - \omega_{1,C} = n\omega_r$, $n = \pm 1$).

In contrast to the standard PISEMA experiment[1] and the amplitude-modulated version,[7] the transverse ^{1}H magnetization is not created and aligned at the magic angle with respect to the static magnetic field at the beginning of the recoupling period. As a consequence, natural ^{1}H magnetization is not spin-locked by off-resonance flip-flop LG irradiation and $^{1}H \rightarrow {}^{13}C$ cross-polarization cannot occur. Oppositely, spin-locking of ^{13}C natural magnetization during the recoupling sequence (LG-SEMA) produces reverse cross-polarization transfer from ^{13}C nuclei to ^{1}H atoms. That is why no additional net ^{13}C magnetization is created by HH contact. Finally, the heteronuclear dipolar couplings are monitored through the oscillation of the initially selected ^{13}C magnetization due to the coherent polarization transfer between ^{13}C and ^{1}H nuclei during the t_1 period. This procedure results in the detection of dipolar spectra of amorphous phase only. As the direct excitation usually causes significant baseline distortion, we inserted a pair of 90°(^{13}C) flip-back and read-out pulses separated by a short delay τ_2 (1−10 ms) between the recoupling sequence and the data acquisition. This way, the baseline distortion is minimized and high-quality dipolar spectra are obtained.

The high efficiency of the reintroduction of heteronuclear dipolar couplings is demonstrated on the experimentally determined ^{13}C signal intensities of CH_3 and CH units of alanine, which were obtained by the proposed pulse sequence DP−SEMA (Scheme 1b) without the train of presaturation pulses. The observed frequency of the dipolar oscillation of corresponding signals nicely fit the dipolar oscillation detected by the standard amplitude modulated PISEMA technique (Figure 1a,c, dependences I and II). This indicates that heteronuclear couplings are recoupled back without additional scaling. The decrease in amplitude of the oscillation follows from the fact that in our case the initial ^{13}C magnetization is created by direct polarization pulse, whereas in the standard PISEMA or PIL-GRIM experiment the phase-inverted on-resonance cross-polarization is applied. The principles of the sensitivity enhancement by polarization inversion accompanied by the spin diffusion have been described previously by Hong et al.[6]

The employed train of presaturation pulses and the short τ_1 delay selectively suppress the ^{13}C signal of the CH group due to the significant differences in $T_1(^{13}C)$ relaxation (Figure 1a, dependence III), whereas the intensity and evolution of mag-

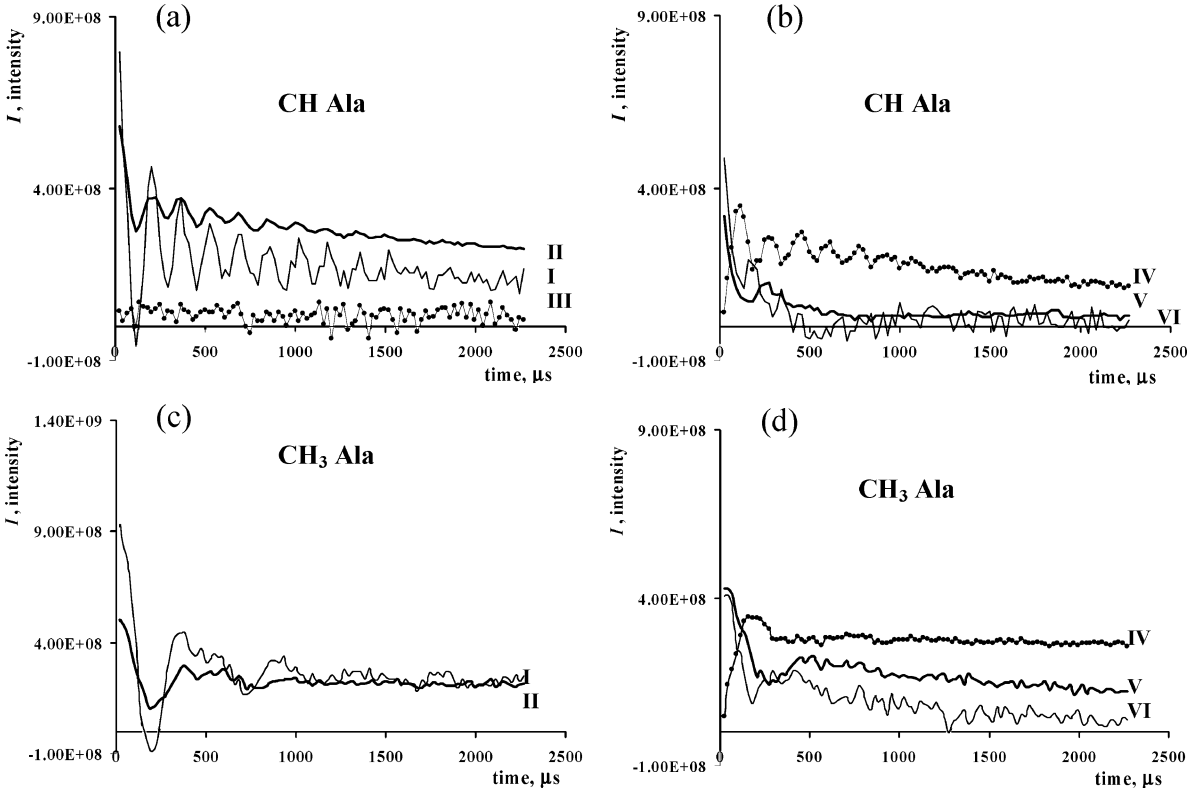


Figure 1. Experimental ^{13}C signal intensity of CH (graphs a and b) and CH_3 units of alanine (graphs c and d) versus the recoupling time (t_1) for amplitude-modulated PISEMA[7] (dependences I), direct-polarization SEMA (dependences II), DP-SEMA with presaturation pulses (dependence III), DP-SEMA with presaturation and initial excitation of ^1H transitions (dependences IV), DP-SEMA with misadjusted ^1H offset (dependences V), and DP-SEMA with mismatched HH condition (dependences VI).

netization of CH_3 is almost unchanged. This clearly confirms prevented polarization transfer from ^1H reservoir to ^{13}C nuclei. If a 54.7° ^1H pulse is applied before the recoupling period, then additional ^{13}C magnetization is created (Figure 1b,d, dependence IV) as the result of HH contact with the spin-locked ^1H polarization. As shown, the required selectivity is completely lost. Finally, the high efficiency of the reintroduction of heteronuclear dipolar couplings is achieved only after relatively careful set up of LG off-resonance and HH matching conditions for ± 1 sidebands. Relatively small mismatch ± 2 kHz (expressed in the intensity of $B_1(^1H)$ field) causes the dipolar oscillation to rapidly attenuate and carbon magnetization to lose its coherence (Figure 1b,d, dependences V and VI).

Experimental Section

The following samples were used to test the proposed pulse sequences: L-alanine, high-density polyethylene (HDPE, Liten MB62), and nanocomposite polyamide-6/montmorilonite[13] (PA6/MMT, Ultramid B5 and Cloisite 30B). All NMR spectra were measured using Bruker Avance 500 WB/US NMR spectrometer (Karlsruhe, Germany, 2003) equipped with 4 mm double-resonance probehead. The magic angle spinning (MAS) frequency was $\omega_r/2\pi = 12$ kHz. The nutation frequency of the $B_1(^1H)$ field for on-resonance cross-polarization and LG irradiation in SEMA recoupling sequence was $\omega_1/2\pi = 76$ kHz with resonance offset ± 54 kHz. This results in cycle time $\tau_c = 2 \times 10.75$ μs. The ^{13}C field strength for presaturation and direct excitation was 84 kHz. During SEMA sequence the ^{13}C field strength was amplitude modulated to fulfill HH matching conditions at $+1$ and -1 spinning sideband. The difference in amplitude was equal to $2(\omega_r/2\pi)$. Heteronuclear decoupling during the signal detection was achieved by 90 kHz ^1H TPPM irradiation.[14] For all experiments the repetition delay was 2 s and the number of t_1 increments (21.5 μs) was 100 covering thus 46 kHz spectral width with 460 Hz FID resolution. The number of scans (NS) varied from 16 to 512 (exact values are introduced in the captions of figures).

Results and Discussion

The high selectivity and efficiency of reintroduction of heteronuclear dipolar couplings achieved by the modified recoupling sequence (DP-SEMA) is clearly demonstrated on the following 2D spectra separating ^{13}C isotropic chemical shifts versus $^{13}C-\{^1H\}$ dipolar profiles. At first, the proposed experimental technique was tested on crystalline alanine, providing high sensitivity of the 2D experiment with direct excitation even at natural isotopic abundance (Figure 2). In addition, CH and CH_3 groups provide simulation of behavior of rigid (crystalline) and highly mobile (amorphous) components due to their significant differences in $T_1(^{13}C)$ relaxation times.

That is why the applied Torchia's T_1 filter inserted into the original sequence (Scheme 1a) and the train of presaturation pulses (DP-SEMA, Scheme 1b) clearly suppress the undesired coherences of mobile and rigid components, respectively. Consequently, the corresponding dipolar spectra are completely erased. Exactly the same shapes of the dipolar spectra of CH_3 group obtained either by the basic amplitude-modulated PISEMA sequence[7] or by DP−SEMA confirm unchanged evolution of heteronuclear interactions upon the applied sequence (Figure 2).

By the same way, the dipolar spectrum of the CH unit measured employing the DP-SEMA sequence without presaturation pulses (not shown here) is quite comparable with Pake's pattern expected for the isolated spin pair (CH). As a conse-

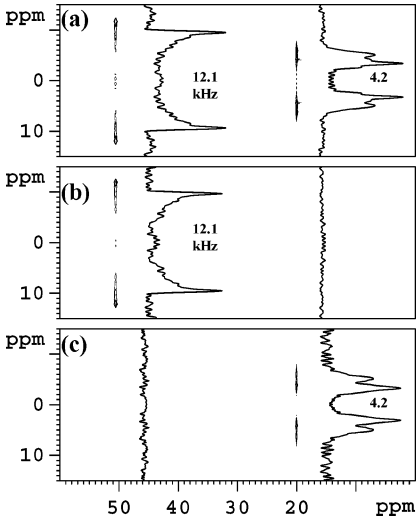


Figure 2. 2D spectra separating ^{13}C isotropic chemical shifts versus $^{13}C-\{^1H\}$ dipolar profiles obtained for alanine by (a) amplitude-modulated PISEMA[7] (AM-PISEMA, number of scans per increment NS = 16), (b) AM-PISEMA with T_1-filter (sequence 1a, NS = 16), and (c) DP−SEMA (sequence 1b, NS = 16). Slices reflecting $^{13}C-\{^1H\}$ dipolar profiles and the obtained splitting in kHz ($\Delta\nu$) are shown within the 2D spectra.

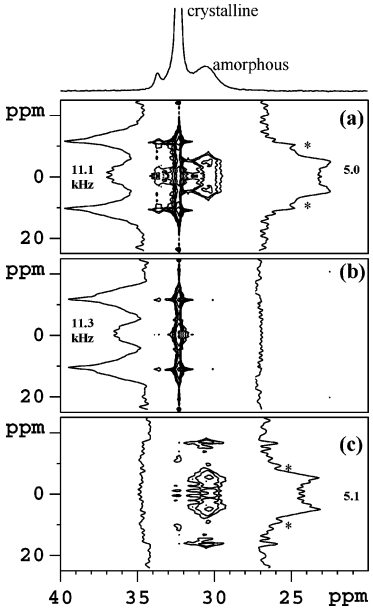


Figure 3. 2D spectra separating ^{13}C isotropic chemical shift versus $^{13}C-\{^1H\}$ dipolar profiles obtained for semicrystalline polyethylene by (a) AM-PISEMA, NS = 32, (b) AM-PISEMA with T_1-filter (sequence 1a, NS = 32), and (c) DP-SEMA (sequence 1b, NS = 128). Slices at ca. 32 and 26 ppm reflecting $^{13}C-\{^1H\}$ dipolar profiles of crystalline and amorphous phase are shown within the 2D spectra.

quence, the splitting between the singularities ($\Delta\nu = 12.1$ kHz) can be directly related to the dipolar coupling constant according to the well-known relation:[5−8] $\Delta\nu = (D_{CH}/\sqrt{2})\sin\theta_m$, where θ_m is magic angle.

Due to the significant differences in conformation of polymer chains in crystallites (all-trans) and amorphous phase (trans-gauche),^{13}C NMR spectra of semicrystalline polyethylene (Figure 3, upper projection) provide excellent spectral resolution of both components. The resulting direct comparison of the dipolar spectra of amorphous phase detected by both experimental techniques definitely confirms the ability of the proposed sequence to selectively measure heteronuclear dipolar couplings in amorphous phase of semicrystalline polymers. Motionally

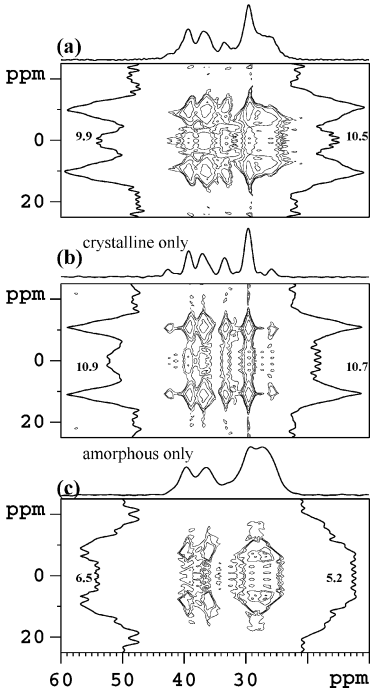


Figure 4. 2D spectra separating ^{13}C isotropic chemical shift versus $^{13}C-\{^1H\}$ dipolar profiles obtained for nanocomposite PA6/MMT by (a) AM-PISEMA, NS = 256, (b) AM-PISEMA with T_1-filter (sequence 1a, NS = 256), and (c) DP-SEMA (sequence 1b, NS = 512). Slices at ca. 39 and 29 ppm reflecting $^{13}C-\{^1H\}$ dipolar profiles and the obtained splitting ($\Delta\nu$) are shown within the 2D spectra.

averaged dipolar couplings of the most abundant fraction of noncrystalline polymer segments are reflected by the reduced splitting between the main maxima (approximately 5.0 kHz). The observed slight difference in the shapes of the both dipolar spectra at higher frequency (marked by asterisks) reflects the decreased signal-to-noise ratio following from the direct excitation of ^{13}C coherence. The presented selective 2D spectrum was recorded almost 13 h to achieve acceptable signal-to-noise ratio (Figure 3c). It is a more than 4 times longer experimental time compared with that of the standard amplitude-modulated PISEMA experiment (Figure 3a).

In general, the ratio of the motionally reduced coupling strength (D_{CH}) and the rigid-limit dipolar coupling constant ($D_{CH,rig}$) defines the bond order parameter,[15] S_{CH}. Assuming segmental motion to be axially symmetric and small in amplitude, the bond order parameter can be converted to the root-mean-square (rms) angle of the motion according to the following relation:[15] $S_{CH} = 1 - (3/2)\langle\theta^2\rangle$. Therefore the proposed DP-SEMA sequence provides the efficient tool to investigate amplitudes of segmental motion in amorphous phase of complex systems.

The great advantage of this technique is demonstrated on the semicrystalline nanocomposite polyamide-6/montmorillonite (PA6/MMT). As the ^{13}C NMR chemical shift is sensitive to the conformation of polyamide chains, ^{13}C CP/MAS NMR spectra of methylene units (50−20 ppm) are very complex and individual signals reflecting all three polymorphs (α- and γ-form, and amorphous phase) strongly overlap (Figure 4, upper projection). Consequently, the slices through dipolar dimension (Figure 4a) reflect several overlapped Pake's doublets, corresponding to a wide range of motionally averaged dipolar couplings. Although the crystallinity of the system is relatively low (ca. 25%), the resulting dipolar profiles are dominated by the dipolar couplings typical for crystalline phase. The detected splitting between the maxima is, however, slightly reduced due

to the interference with dipolar spectrum of amorphous component, indicating that the analysis of these dipolar spectra is very difficult.

Selectively detected dipolar interactions in crystallites are more then 1 kHz stronger, reflecting amplitudes of reorientation of CH_2 units of polymer chains with rms angle ca. 11−16° (calculated for all CH_2 groups within one monomer unit). On the other hand, substantially released reorientation of CH_2 units as reflected by their high amplitudes (rms 23−35°) is detected for polymer chains in amorphous phase (Figure 4c). In this particular case, relatively low resolution indicates that these dipolar spectra must be considered as a superposition of various dipolar doublets. The broad maxima reflect the presence of multiple CH_2 units in various environments exhibiting different amplitudes of segmental reorientation in amorphous phase of polyamide matrix.

Conclusion

The accurate detection of dipolar couplings in the amorphous phase of semicrystalline polymer systems requires application of a selective experimental technique that efficiently suppresses undesired coherences. These requirements are achieved by the proposed amplitude-modulated PISEMA-type pulse sequence. The ability of this 2D technique (DP-SEMA) to measure $^1H-^{13}C$ dipolar couplings selectively for mobile components was at first demonstrated on semicrystalline polyethylene and subsequently confirmed on complex nanocomposite system polyamide-6/montmorillonite.

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