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Dynamic nuclear polarization solid-state NMR spectroscopy as a tool to rapidly determine degree of modification in dialcohol cellulose

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Abstract Dialcohol cellulose can be prepared by periodate-mediated oxidation of cellulose followed by reduction with borohydride. The two-step reaction creates a modified cellulose polymer which is ring-opened between the C2 and C3 carbons in the glucose unit. This material has attracted both scientific and commercial interest, due to its potential role in the transition towards a fossil-fuel-free society. In order to become a reliable component in the materials of tomorrow, chemical properties such as degree of modification must be accurately quantified. In this work we describe how solid-state

NMR spectroscopy, enhanced by dynamic nuclear polarization (DNP), can be used for this purpose. Our results illustrate that it is possible to obtain high sensitivity enhancements in dialcohol cellulose with the DNP enhanced solid-state NMR technique. Enhancements above a factor of fifty, on a 400 MHz/263 GHz DNP system in the presence of 12 mM AMUPol radical were achieved. This allows us to quantify the degree of modification in dialcohol cellulose samples in time spans as short as 20 min using DNP enhanced multiple-contact cross polarization experiments. We also exemplify how DNP enhanced, ^{13}C - ^{13}C dipolar recoupling experiments can be used for the same purpose and for studying chemical shift correlations in dialcohol cellulose.

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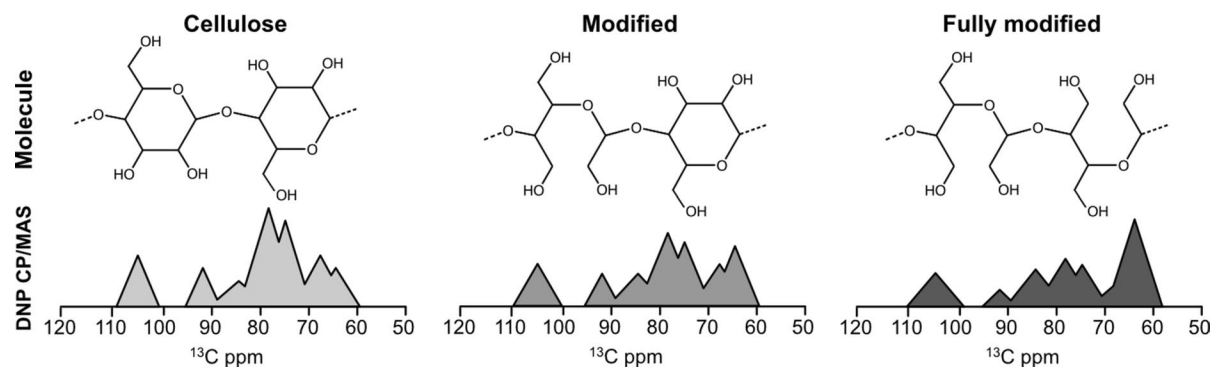
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Graphical abstract



Keywords Dialcohol cellulose · Dynamic nuclear polarization · Solid-state NMR · Degree of modification · Renewable thermoplastics

Introduction

The accumulation of non-degradable plastic materials in nature is an increasing environmental problem and poses a great threat to sensitive ecosystems and maritime wildlife (Rhodes 2018; MacLeod et al. 2021). This problem constitutes a major motivator for the development of more environmentally friendly and bio-degradable materials (Kaplan 1998; Klemm et al. 2005). In many applications, these materials need to be thermoplastic, i.e., heat-processable into three-dimensional, functional shapes. They should in addition have other properties that make them suitable for replacing petroleum-based plastics, such as sufficient mechanical strength, resistance to moisture and ability to create oxygen-barriers (Comyn 1985; Börjesson et al. 2019). Dialcohol cellulose i.e. a partly or fully modified cellulose where the C2–C3 bonds in the glucose units have been broken and the secondary hydroxy groups have been transformed to primary ones, is one such material that has shown promise for forming a basis for renewable thermoplastics (Head 1955; Casu et al. 1982; Simon et al. 2024). Chemically, this modification is achieved by a two-step reaction where the cellulose first is oxidized with periodate to dialdehyde cellulose and subsequently reduced to dialcohol cellulose using a reducing agent, typically sodium borohydride (Fig. 1).

The oxidation may be performed in a heterogeneous reaction in which the solid cellulose starting material is suspended in water, together with dissolved periodate (Zeronian 1963; Maekawa 1991). It has been suggested that the mechanism is to slowly oxidize the cellulose from the surface, on a nanofibrillar level inside the fiber wall. Dialcohol cellulose samples prepared via heterogeneous reactions, have shown promising mechanical properties and it has been possible to process such modified fibers into three-dimensional shapes resembling functional products (Larsson et al. 2014; Larsson and Wågberg 2016; Lo Re et al. 2023). An alternative to a heterogeneous reaction is to first dissolve the cellulose, for example by the formation of a more water-soluble derivative such as methylol cellulose and then proceed to oxidation. This leads to a more homogenous and rapid oxidation (Morooka et al. 1989). Just as dialcohol cellulose prepared via the heterogeneous reaction can be further processed into useful objects, such as being cast into thin, transparent films, dialcohol cellulose prepared via homogeneous oxidation possesses similar potential (Kasai et al. 2014).

The continued development of renewable thermoplastic materials from dialcohol cellulose relies on being able to characterize chemical properties of the modified molecule and correlate them with the macroscopic properties of the final product. One such chemical property is the fraction of ring opening and subsequent conversion to primary hydroxyl groups in the cellulose, from here on referred to as degree of *modification*. The fraction of ring opening,

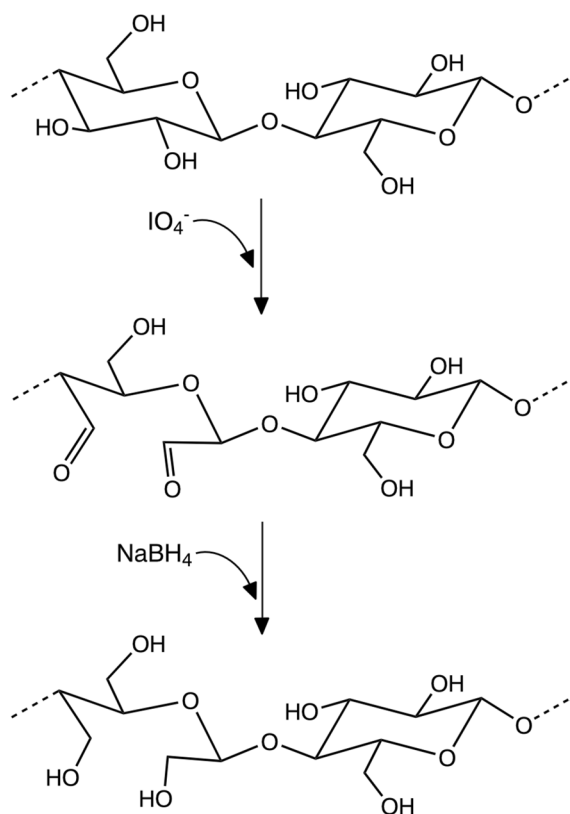


Fig. 1 Reaction scheme for the synthesis of dialcohol cellulose. Cellulose is first site-specifically oxidized at the 2,3 position to form ring-opened dialdehyde cellulose. The aldehyde is then reduced to form dialcohol cellulose

and amount of aldehyde groups formed, i.e. in the first reaction step is referred to as degree of *oxidation*. In the text we will use these two definitions and refer to “degree of oxidation” for values from methods measuring the first reaction step, and “degree of modification” for values from methods measuring the final dialcohol cellulose. Degree of oxidation can be determined using acid base titration after reacting the aldehyde groups in the dialdehyde cellulose with hydroxylamine hydrochloride (Zhao and Heindel 1991). An alternative method for quantifying degree of oxidation is to measure the consumption of the periodate during the reaction, typically by monitoring the UV absorbance (Maekawa 1991; Morooka et al. 1989). Both of these methods measure the progress of the first step of the reaction, the oxidation step. Since, complete conversion to primary hydroxyl groups is not guaranteed for all positions, the degree of modification may differ from the degree of oxidation, even

if one commonly assumes that they are the same. The second borohydride reduction step might be incomplete or lead to unwanted β -elimination side reactions (Simon et al. 2023). Therefore, to ascertain the quantity of the ultimate reaction product, dialcohol cellulose, with a specific degree of modification, it is preferable to utilize techniques that analyze the final dialcohol cellulose molecule instead. Solution-state NMR spectroscopy is one such technique that has been used to characterize dialcohol cellulose (Maekawa 1991; Siller et al. 2015). But, since the solubility of dialcohol cellulose is modification-dependent, solution-state NMR experiments can be challenging, although ionic liquid-based methods for cellulosic materials exist (King et al. 2018; Fliri et al. 2023). The challenges of studying these cellulose derivatives with solution-state NMR has spurred cellulose researchers to use solid-state NMR to characterize oxidized cellulose (Kim et al. 2000; Lindh et al. 2014) and recently solid-state NMR-based methods for determination of degree of oxidation have been described in literature (Leguy et al. 2019). A drawback of studying cellulose materials with conventional solid-state NMR is that one typically needs to rely on the low abundance ^{13}C isotope for the experiments. This tends to make experiments lengthy and usually data collection over several hours with thousands of acquisitions is necessary to ensure sufficient spectral quality and signal-to-noise ratio. A possible way to speed up solid-state NMR experiments is to use dynamic nuclear polarization (DNP). DNP is a signal enhancement technique, and the much stronger NMR signal obtained leads to significantly shorter experimental times compared to conventional solid-state NMR methods. DNP is based on transfer of spin polarization from unpaired electrons (free radicals) in the sample to the atomic nuclei in the presence of continuous wave microwave irradiation (Ni et al. 2013; Thankamony 2017; Corzilius 2020). DNP experiments are typically performed at low temperatures (~ 100 K) and the radicals need to be added to the sample prior to the experiments. Stable organic radical molecules designed for DNP usage, such as AMUPol (Sauvée et al. 2013) are popular and development of new DNP radicals is a highly active research area (Menzildjian et al. 2023). The radicals are dissolved in a cryoprotective solvent system, that prevents crystallization upon freezing and instead forms a glass upon solidification. A commonly used

mixture is glycerol-d8/D₂O/H₂O in volume proportions 6:3:1 (Casano et al. 2020). Dimethyl sulfoxide (DMSO) is also a common alternative additive to prevent crystallization upon freezing (Michaelis et al. 2014). In contrast to glycerol, DMSO is also favorable in the cellulose context because of its ¹³C chemical shift properties as the CH₃ signals show up around 39.5 ppm (Gottlieb et al. 1997) in the carbon spectrum, which does not overlap with any of the carbon signals from the cellulose. If glycerol is favored as cryoprotectant, the problem of overlapping with the cellulose signals can be solved by using glycerol depleted of ¹³C. Another approach for applying the radical that also resolves the issue of spectral overlap with solvent peaks is instead to use the so-called *matrix-free* method introduced by Takahashi and coworkers (Takahashi et al. 2012). In this method the radical is dissolved in a solvent and then applied to the solid, after which the solvent is evaporated away in a desiccator. The matrix-free method has been frequently used for DNP studies of cellulosic materials (Takahashi et al. 2012; Kirui et al. 2019; Deligey et al. 2022). The signal enhancement and reduction of experimental times associated with the DNP-NMR technique makes it much more feasible to perform solid-state NMR experiments on cellulose samples at natural abundance levels of ¹³C. In addition, to perform normal cross-polarization (CP) experiments (Pines et al. 1972) it also becomes achievable to record experiments where the CP transfer step is repeated, so-called multiple-contact CP experiments (Zhang et al. 1990; Johnson and Schmidt-Rohr 2014). By repeating the CP transfer step in a loop one can compensate for differences in heteronuclear dipolar coupling and obtain more quantitative spectra. This type of experiment has recently been proven feasible under DNP conditions (Brownbill et al. 2018; Bertarello et al. 2021; Kumar et al. 2023). Here, we take full advantage of the possibility to perform fast, DNP-enhanced multiple-contact CP experiments in order to quantify degree of modification in dialcohol cellulose. We show how the multiple-contact CP experiment improves the agreement of the normalized integrals in our spectra with the corresponding number of carbon atoms in the molecule. This is further discussed in relation to possible changes in heteronuclear dipolar coupling properties under DNP conditions, in the presence of radical solution and at low temperature. In addition to the usage of

multiple-contact CP experiments for quantification of degree of modification, we also explore the possibility to use homonuclear dipolar recoupling (Ji et al. 2021; Nielsen et al. 2012) techniques for the same purpose and also to obtain ¹³C chemical shift correlations and assignments. ¹³C-¹³C correlations of DNP-enhanced, natural abundance cellulose samples have been extensively exemplified in literature before. Both as dipolar or *J*-coupling based single/double-quantum correlation spectra (Takahashi et al. 2012; Perras et al. 2017; Kirui et al. 2019; Berruyer et al. 2021; Deligey et al. 2022) and via techniques involving the ¹H network (Kobayashi et al. 2017; Zhao et al. 2021; Deligey et al. 2022). But here we primarily focus on the usage of a 1D selective experiment using the SPC5 pulse sequence element (Hohwy et al. 1999) and the correlation between C1 and C2 in dialcohol cellulose. We perform these experiments on two sample categories: dialcohol cellulose with lower degrees of modification and originating from fibers, and dialcohol cellulose originating from microcrystalline cellulose (MCC) with higher degrees of modification.

Experimental section

Dialcohol cellulose sheets from beaten softwood kraft fibers

Four different paper sheets made from dialcohol cellulose fibers were studied as representations of “low” degree of modification. These were the same sheets as those used in a study by Larsson and Wågberg (Larsson and Wågberg 2016) and had degree of oxidation of 0%, 13%, 24% and 40% (i.e. fraction of aldehyde groups on C2 and C3 positions, if all C2 and C3 positions are oxidized to aldehydes, degree of oxidation would be 100%), assessed by hydroxylamine titration after the initial periodate oxidation step, these samples are here referred to as *Fiber 1–4* in the text.

Dialcohol cellulose from microcrystalline cellulose MCC

Sodium metaperiodate was dissolved in 1.5 L of deionized water in a 2 L Erlenmeyer flask. Isopropanol to a concentration of 6.3 vol% was added to the flask, followed by adjustment of pH to the interval 3.6–4.0 with acetic acid. After this, 30 g of

oven-dried (50 °C) MCC was added to the flask. The molar ratio of glucose units relative to sodium metaperiodate was 1.1:1. The flask was wrapped with aluminum foil to protect the reaction solution from light and stirred at 200 rpm. The degree of oxidation was monitored by measuring the UV absorbance at 290 nm (Maekawa and Koshijima 1984). Reactions were stopped when absorbance values corresponded to 55%, 75% and 100% degree of oxidation. The reactions were stopped by filtering off the oxidized cellulose and washing it with deionized water until no UV absorbance from the periodate was detected. It was then stored in refrigerator, at 4 °C until further processing. The oxidized cellulose was reduced to dialcohol cellulose, by resuspending the product in 200 mL deionized water in a 2 L Erlenmeyer flask, and adding monobasic phosphate to a final concentration of 0.02 M. After this, a solution of 15 g sodium borohydride in 100 mL deionized water was added dropwise to the larger Erlenmeyer flask with the suspension of the oxidized cellulose. The reduction reaction was then stirred at 200 rpm for four hours at room temperature, after which the reaction solution was dialyzed for one week. After dialysis, the dialcohol cellulose was oven dried at 40 °C. These three samples are referred to as MCC 1–3 in the text.

Solid-state NMR spectroscopy

Room temperature NMR experiments on the Fiber 1–4 samples were performed on a Varian Inova 600 MHz spectrometer equipped with a 3.2 mm magic angle spinning (MAS) probe. CP/MAS experiments were recorded with 8192 scans, a relaxation delay of 5 s and at 15 kHz MAS rate. DNP NMR experiments were performed on a 400 MHz Bruker Ascend DNP instrument with a 3.2 mm LTMAS probe and a 263 GHz gyrotron for microwave generation. MAS rates of 5.5, 8 and 10 kHz were used. AMUPol (CortecNet) at a concentration of 12 mM was used as DNP radical, dissolved in either D₂O/H₂O in volumetric proportions 9:1 or DMSO-d₆/D₂O/H₂O in volumetric proportions 6:3:1, depending on sample. Typically, a radical solution without DMSO was used for samples where the degree of modification was known to be high (i.e. samples where MCC was the starting material), but for the MCC 3 sample, radical solutions both with and without DMSO was tested for

comparison. The usage of an AMUPol concentration of 12 mM was based on our previous optimizations performed on cellulose ethers. During these earlier experiments different radical systems were tested (protonated tetrachloroethane with 16 mM TEKPol, 12 mM AMUPol in D₂O/H₂O, with varying D₂O proportions) and performance of the radical solutions was measured as the DNP sensitivity enhancement calculated from spectra recorded with the microwaves turned on and off. The samples were wetted with the radical solution, packed into 3.2 mm sapphire rotors, and stored in freezer at –20 °C. The exposure time to the radical solution at room temperature conditions during packing procedures was around 10–15 min. CP experiments (Pines et al. 1972) and multiple-contact CP experiments, based on (Johnson and Schmidt-Rohr 2014; Bertarello et al. 2021), were performed. ¹³C chemical shifts were calibrated relative to the DMSO CD₃ signal, which was assumed to have a chemical shift of 39.5 ppm (Gottlieb et al. 1997) also under cryogenic, DNP conditions. This put the C1 chemical shift of dialcohol cellulose at 106 ppm and it was assumed to have this chemical shift also in the room temperature CP/MAS spectra. For multiple-contact CP experiments the recovery delays between repetitions of the CP step were set to $1.3 \times {}^1\text{H } T_{\text{DNP}}$ (¹H T_{DNP} refers to longitudinal build-up time of ¹H magnetization under DNP conditions, i.e. in the presence of radical solution and microwave irradiation). Experiments with an increasing number of CP repetitions were performed to ensure that a plateau in peak intensity was reached, and that the maximum amount of magnetization had been transferred to all carbons. Longitudinal relaxation times were measured with standard saturation and inversion recovery experiments. Proton T_1 relaxation times were measured with the Bruker satrect1 pulse sequence. For the Fiber 3 sample, ¹³C detected ¹H saturation recovery data was acquired with the microwaves turned on and off respectively and the DNP enhancement as a function of relaxation delay was calculated from the ¹³C spectra. For these experiments 576 scans were used for the experiments with microwaves turned off and 32 scans for the experiments with microwaves turned on. The spectra from the measurements with microwaves turned on were then scaled $\times 18$ prior to DNP enhancement calculation, which was calculated as the ratio of the signal

integral from the spectra with microwaves turned on and off. ^{13}C T_1 relaxation times were measured for MCC 1 using the Bruker cppt1 pulse sequence. Selective, 1D ^{13}C - ^{13}C dipolar recoupling experiments were performed using (after ^1H - ^{13}C CP transfer) a 1 ms Gaussian shaped pulse with 10% truncation level for selective C1 flip-back followed by a z-filter and the SPC5 dipolar recoupling block. These experiments were carried out at a MAS rate of 8 kHz with a SPC5 RF-field strength of 40 kHz and 500 μs excitation, and 500 μs reconversion time as described by (Hohwy et al. 1999) and 2-4 k scans. Continuous wave (CW) ^1H decoupling with a field-strength > 100 kHz was used during the SPC5 transfer block.

Spectral analysis and calculation of degree of modification

NMR-data for DNP-enhancement calculations and from relaxation measurements, were Fourier transformed, phase corrected and subjected to fifth order polynomial, automatic baseline correction in Topspin 4.1.1. The data was then exported and analyzed using Python 3.8 scripts. Relaxation data was fitted to exponential or stretched exponential functions (Narayanan et al 1995).

Quantification of degree of modification from CP/MAS and multiple-contact CP/MAS experiments were done in MestreNova 14.2.0, in order to be able to treat Varian and Bruker data in the same way. The data was phased, and baseline corrected using third order Bernstein polynomial. The integral of the spectral region belonging to C1 (96.1–112.9 ppm) was normalized to one. Integrals for two other spectral regions were then calculated: non-modified C2, C3, C4, C5, modified C4, and modified C5 (69.4–96.1 ppm) as well as C6, modified C2, and modified C3 (56.4–69.4 ppm). Modified (ring opened with primary hydroxyl groups) C2 and C3 atoms are from here on referred to as $\text{C}_{2\text{mod}}$ and $\text{C}_{3\text{mod}}$. It was assumed that any normalized integral area in the C6, $\text{C}_{2\text{mod}}$ and $\text{C}_{3\text{mod}}$ region (56.4–69.4 ppm) exceeding the value of one originated from signals of $\text{C}_{2\text{mod}}$ and $\text{C}_{3\text{mod}}$ carbons. This excess area was thus divided in two, to immediately give the fraction of open rings, or degree of modification. Degree of modification was also calculated from the *reduction* in integrals in the

69.4–96.1 ppm region due to loss of non-modified C2 and C3 signals.

In addition to these two calculation methods, degree of modification was also calculated from the selective SPC5 experiment for the MCC 2 and MCC3 samples. This was done by Fourier transforming, phase- and baseline correcting the 1D spectrum in Topspin 4.1.1 after which the data was exported and analyzed using Python 3.8. Degree of modification was directly calculated by comparing the ratio of the integrals belonging to the signal regions of $\text{C}_{2\text{mod}}$ (56.3–68.0 ppm) and non-modified C2 signal (68.0–78.2 ppm) for the MCC2 sample and for the MCC3 sample, the regions $\text{C}_{2\text{mod}}$ (58.6–69.7 ppm) and C2 (69.7–77.6 ppm) were used.

Results and discussion

DNP enhancements and chemical shifts of dialcohol cellulose

Depending upon degree of modification, the water solubility of dialcohol cellulose tends to increase with increasing degree of modification. Whereas almost fully modified samples dissolve in water to some extent, samples with low degree of modification more resemble intact cellulose which is not soluble in water (Wohlert et al. 2022). When performing DNP-NMR experiments, the sample material is wetted or soaked in the radical solution, and naturally, the wetting and the solubilizing properties of the radical solution on the solid sample material will greatly affect the outcome of subsequent experiments. This is a major difference compared to conventional solid-state NMR; the addition of the radical solution to the solid and its potential interaction with the solid. If the radical solution does not dissolve the solid material and large particles remain intact, spin polarization from the ^1H bulk in the radical solution needs to spin diffuse longer distances into the solid material in order to enhance the signal. If on the other hand the radical solution partly dissolves or swells the sample material, protons with high spin polarization from the radical solution are in closer proximity to a larger proportion of the sample molecules and polarization does not need to spin diffuse particularly long in order to give high signal for the sample molecules.

For all of the dialcohol cellulose samples we have tested in this work, we either used a radical solution consisting of 12 mM AMUPol in D₂O/H₂O, with volume proportions 9:1 or DMSO-d₆/D₂O/H₂O, with volume proportions of 6:3:1. For samples originating from MCC and with higher expected degree of modification, radical solutions without DMSO-d₆ were used, but for the MCC 3 sample, solutions both with and without DMSO was tested. The reason for not using DMSO in the radical solution for the MCC samples was since it was known from experience that more hydrophilic cellulose derivatives typically swell in water based radical solutions and give high DNP enhancement. DMSO-d₆ was used in the radical solution for samples originating from fibers, where the degree of modification was known to be lower, since swelling and glass formation in water were not anticipated for those samples. The comparison of spectra for the MCC 3 sample (Fig. S1) acquired both with and without DMSO-d₆ containing radical solution shows a difference in signal-to-noise ratio but a negligible impact on relative peak integrals, because of this D₂O/H₂O was used as radical solvent for the experiments with the MCC samples. For all samples, addition of the radical solution either led to a visually

obvious swelling/dissolution or at least a proper wetting of the dialcohol cellulose samples tested, and it was easy to obtain a high DNP enhancement. This is illustrated by the DNP enhanced ¹³C CP/MAS spectra (Fig. 2a) for the Fiber 4 sample. This sample had a degree of oxidation of 40% (measured with the titration-based method, see Experimental section) and it was wetted/swelled with the DMSO based radical solution. The DNP enhancement = 53, was calculated as the ratio of the integrated signal in the 53–115 ppm region of the spectra with microwaves turned on and off, respectively. We stick to this way of reporting the DNP enhancement in this work, even if a considerable depolarization (Mentink-Vigier et al. 2015) of the sample associated with the AMUPol usage is likely to occur during the microwave off experiments. The CP/MAS spectra of dialcohol cellulose changes with increasing degree of modification (Fig. 2b) and the ¹³C signals of the ring opened C2 and C3 carbons (C_{2mod}/C_{3mod}) are upfield shifted and typically overlap with the signal from C6 in the 50–70 ppm region. It is this upfield chemical shift change we take advantage of in the next sections to calculate the degree of modification. The spectra (Fig. 2b) have been normalized such that the C1 regions have been set equal

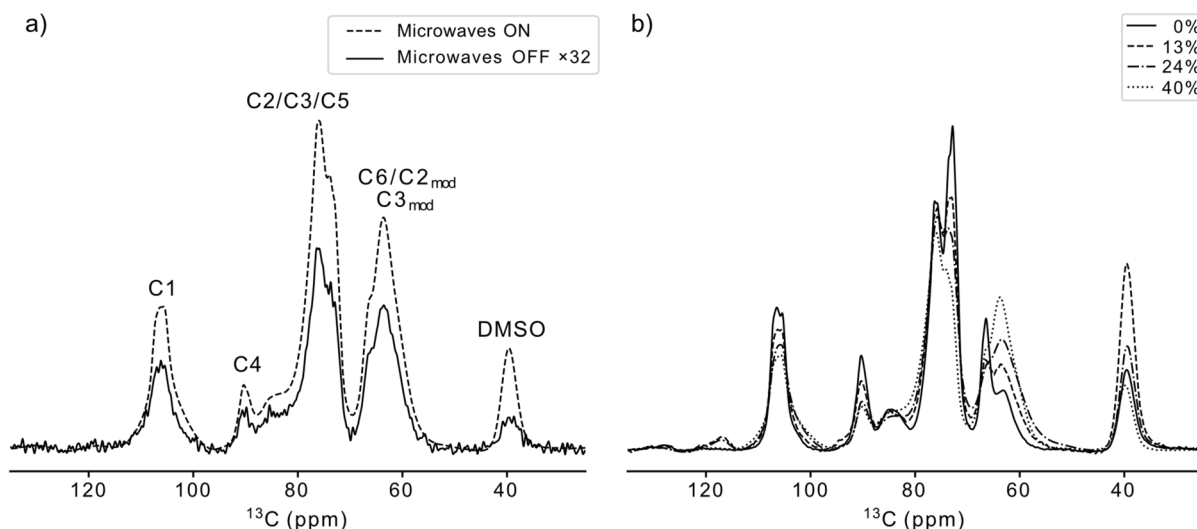


Fig. 2 DNP enhanced ¹³C CP/MAS spectra of dialcohol cellulose originating from fibers, T ≈ 105 K, MAS = 5.5 kHz. **a** Comparison of spectra recorded with the microwaves turned on and off (microwave off spectrum scaled 32 times) for the Fiber 4 sample, DNP enhancement = 53, using a recycle delay of 10 s. **b** Spectra for the Fiber 1–4 samples (degree of oxida-

tion values from titration-based method are shown), normalized such that the integral for the C1 region for all spectra are equal. The spectra illustrate changes upon increasing modification, notice especially the increase in signal in the 50–70 ppm region, due to C_{2mod}/C_{3mod} signals from opened rings in the cellulose

to one. In addition to the increase in signal in the 50–70 ppm region due to $C2_{\text{mod}}/C3_{\text{mod}}$, also notice the broadening of C1 (106.5 ppm) and reduction of signals corresponding to crystalline C4 (90.3 ppm) and C6 (66.5 ppm) associated with increasing degree of modification. We primarily attribute the broadening of the NMR signals, associated with increased degree of modification, to an increased conformational heterogeneity associated with the ring opening.

Regarding the DNP enhancement, it is important to consider to what extent the dialcohol cellulose fibers (or to be precise, the cellulose nanofibrils inside the fiber wall), swell or even dissolve in the radical

solution. In order to investigate this further, we performed ^1H saturation recovery experiments on the Fiber 3 sample (Fig. 3).

This data shows that the ^1H T_1 of the dry material is long (95.8 s) which is favorable in order to get high sensitivity enhancements (Rossini et al. 2012, 2014; Pinon et al. 2017). But the question is whether or not there is a homogenous distribution of ^1H polarization in our dialcohol cellulose samples under the DNP conditions used. Rossini et al. (Rossini et al. 2014) exemplifies how this can be assessed by plotting the DNP enhancement as a function of relaxation delay (Fig. 3d). If such a curve is flat and the enhancement

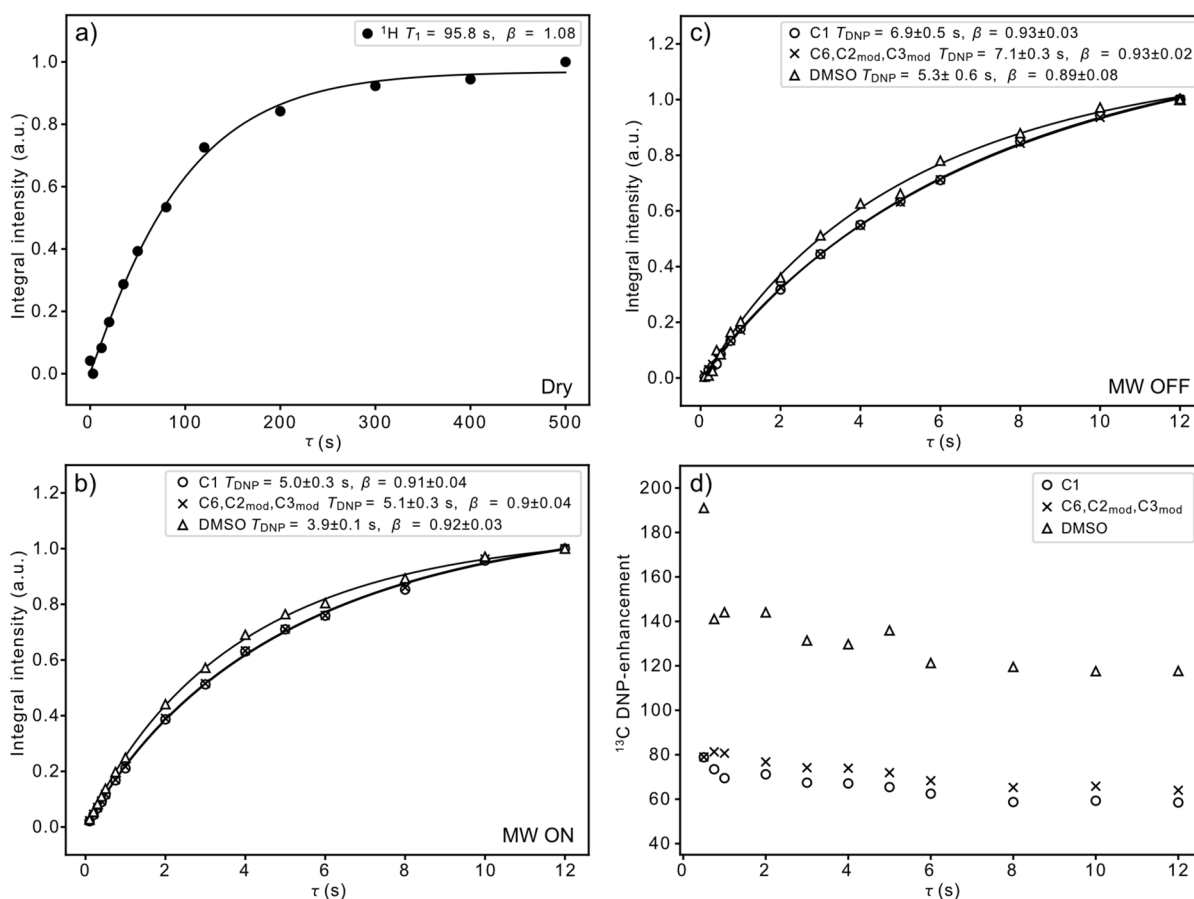


Fig. 3 **a** ^1H detected ^1H saturation recovery data for the dry (no radical solution) Fiber 3 sample recorded, without microwaves at $T=97$ K and $\text{MAS}=5.5$ kHz and fitted to a stretched exponential. The curve illustrates the long ^1H T_1 relaxation time for the dry intact material. **b** ^{13}C detected ^1H saturation recovery data for the same material but under DNP conditions (microwaves on, 12 mM AMUPol DMSO- d_6 /D $_2$ O/H $_2$ O (6:3:1), $\text{MAS}=5.5$ kHz, $T=105$ K) showing the ^1H T_{DNP}

observed on the different carbons. **c** ^{13}C detected ^1H saturation recovery data for the same sample as in **b**) but with the microwaves turned off. **d** Plot of the DNP enhancement (integrals from the ^{13}C spectra with microwaves turned on divided by the corresponding integrals from the microwave off spectra) as a function of the relaxation delay (τ). The data points for $\tau < 0.5$ s, were associated with high noise and excluded from the plot

is independent of the τ delay, polarization is likely to be evenly distributed. As can be seen (Fig. 3d) our DNP enhancement plot is not flat and there is a slight reduction in DNP enhancement with increasing τ delay, thus we cannot rule out an uneven polarization distribution and spin diffusion effects, especially not in the $\tau \approx 2\text{--}5$ s range where the majority of our recycle delays are set. This could be discouraging for the prospect of using the DNP enhanced multiple-contact CP experiment for the quantification of the degree of modification in dialcohol cellulose. However, as will be shown in the next section we perform both DNP enhanced multiple-contact CP experiments as well as conventional, room temperature CP/MAS experiments for the Fiber 1–4 samples and we do obtain very similar degree of modification values from these different experiments. So even if an uneven enhancement might occur to some extent in our dialcohol cellulose DNP samples, the parts that are enhanced are likely representative for the whole samples. Thus, we dare to trust the degree of modification values obtained from the DNP enhanced multiple contact CP experiments.

It should also be highlighted that there are slight differences in the T_{DNP} times observed on the DMSO CD_3 carbon in the saturation recovery data obtained with microwaves on and off (Fig. 3b, c). It is our understanding that this would also contribute to a reduction of the DNP enhancement with increasing relaxation delay. The slight reduction in DNP enhancement observed (Fig. 3d) could thus both originate from inhomogeneous wetting as well inhomogeneous radical distribution in the sample. To disentangle the two possible origins of the decreasing DNP enhancement, more rigorous spin diffusion simulations on the dialcohol cellulose system would be necessary, but we consider this to be outside scope of this study. For now we settle for the observation that we obtain similar degree of modification values from the multiple contact CP experiments and the room temperature CP/MAS experiments, which will be shown in the next section.

Quantification of the degree of modification using multiple-contact CP experiments

The classic solid-state NMR CP transfer experiment (Pines et al. 1972) in which spin polarization is transferred from a high gamma nucleus to a low gamma

nucleus to enhance the NMR signal thereof has become a cornerstone of modern solid-state NMR. A drawback of this experiment is that it relies on a dipolar-mediated, through-space transfer of magnetization and since the heteronuclear dipolar coupling constants may vary throughout the molecule this might lead to an uneven amount of transferred signal to the heteronuclei, which make studies of the resulting peak integrals unreliable. Suggestions such as ramping the amplitude of the contact pulses (Metz et al. 1996) or inclusion of homonuclear spin diffusion steps (Hou et al. 2006) have been proposed over the years to counteract this problem, but here we choose to use the multiple-contact CP approach (Zhang et al. 1990; Johnson and Schmidt-Rohr 2014) in order to get more quantitative ^{13}C CP/MAS spectra of our dialcohol cellulose samples. In the multiple-contact CP experiment (Fig. 4a), the CP step is repeated in a loop with flip-back pulses, recovery delay and flip-down pulses in it. When the loop is repeated, hyperpolarized proton magnetization will continuously build up during the recovery delay in the loop while the heteronuclear magnetization is saved as slowly relaxing longitudinal magnetization. The repeated CP step in the multiple-contact experiment ensures that magnetization is transferred to the heteronuclear spins in such a way that the maximum possible amount of magnetization is transferred to all heteronuclei, regardless of variations in magnitudes of the heteronuclear dipole coupling constants, thus making peak integrals more representative of the actual corresponding carbon quantities. The repetition of the CP step makes the multiple-contact CP experiment time-demanding, and if, for instance, five CP repetitions are made the experiment will take five times as long compared to the normal CP experiment. This makes the multiple-contact CP experiment unattractive to run on normal solid-state NMR spectrometers with natural abundance levels of ^{13}C or other low abundant heteronuclear isotopes. But here, on our dialcohol cellulose samples with strong DNP enhancement we can perform multiple-contact CP experiments with five CP repetitions and signal to noise ratios above 700 ($S/N > 700$) in less than 30 min acquisition time.

It is important to ensure that the CP step has been repeated a sufficient number of times to reach a plateau of the ^{13}C signal intensities. Bertarello et al. (2021) reports that typically five repetitions of the CP step are enough to get quantitative results,

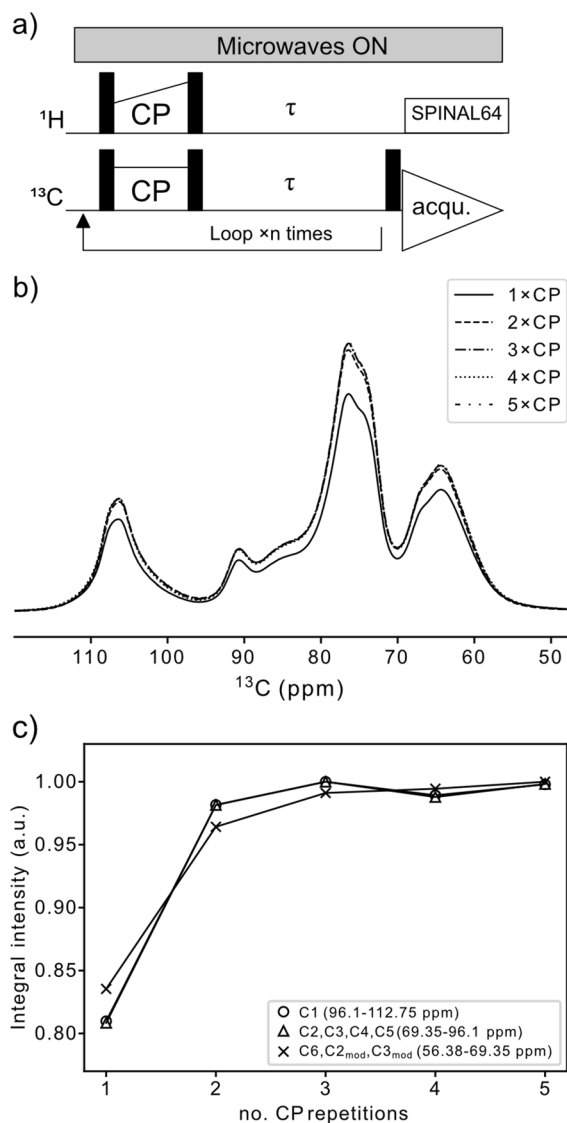


Fig. 4 **a** The multiple-contact CP pulse sequence. **b** Multiple-contact CP/MAS spectra with increasing number of CP repetitions for the MCC 1 sample (degree of oxidation = 55%, according to UV method). Notice the build-up of signal with increasing number of CP repetitions. **c** Normalized integrals for different spectral regions for the spectra in **b**, notice with increasing number of CP repetitions how the difference between the C1 region (empty circle) and the region belonging to C6, C_{2mod} and C_{3mod} (black cross) is reduced

Brownbill et al. (2018) reports six to ten. This is in good agreement with what we observed (Fig. 4b, c) on the dialcohol cellulose samples studied here and typically between four and eight CP repetitions

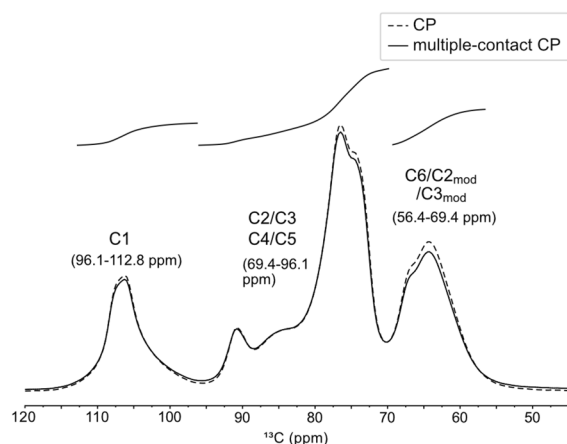


Fig. 5 Comparison of DNP enhanced ¹³C CP/MAS and multiple-contact CP/MAS spectra for the MCC 1 sample. Multiple-contact CP/MAS spectrum with five CP repetitions (solid line), superimposed on CP/MAS spectrum (dashed line). The spectra are normalized such that their C1 integrals are equal, notice especially the reduction in signal for the C6/C_{2mod}/C_{3mod} region

were used to reach a plateau in ¹³C intensities. A comparison between spectra from the normal DNP enhanced CP/MAS experiment and the DNP enhanced multiple-contact CP/MAS experiment with five CP repetitions can be made (Fig. 5).

In the spectra (Fig. 5), one can see a clear difference and that the signal intensity in the 56.4–69.4 ppm region where the C6, C_{2mod} and C_{3mod} signals reside is reduced in the multiple-contact experiment. This difference between the spectra must be due to a slightly higher ¹H to ¹³C transfer efficiency for the C6, C_{2mod} and C_{3mod} region under DNP conditions, that in the multiple-contact experiment is compensated for when a sufficient number of CP repetitions are made. The plot (Fig. 4c) also illustrates this, where it can be seen that after only one CP repetition the normalized integral for the C6, C_{2mod}, C_{3mod} spectral region (56.4–69.4 ppm) is slightly larger relative to the normalized integrals of the two other regions, whereas differences are reduced when the number of CP repetitions are increased.

In order for the multiple-contact CP experiment to work correctly, it is important that the *T*₁ relaxation times of the ¹³C nuclei are longer than the ¹H *T*_{DNP} build-up times and the τ delay in the pulse sequence (Fig. 4a). Otherwise, the ¹³C magnetization will decay down to a lower steady state value during the

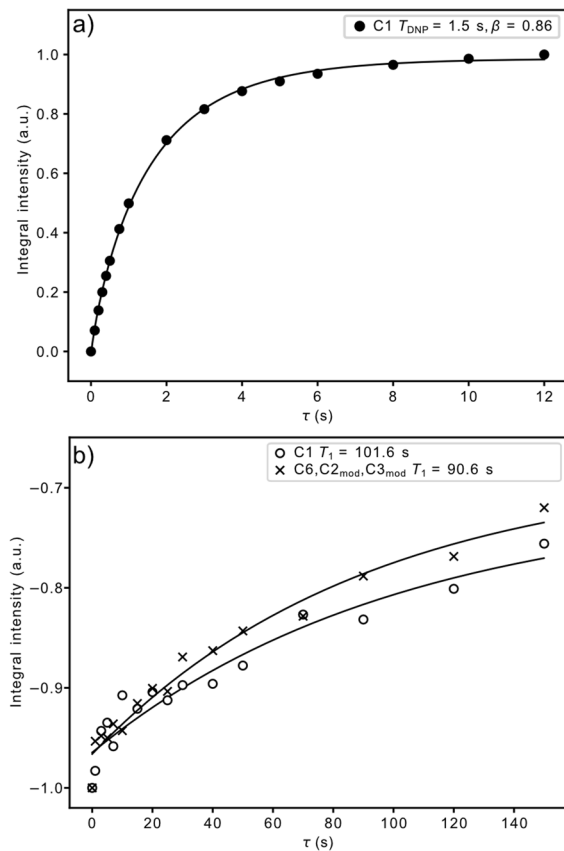


Fig. 6 Relaxation data for the MCC 1 sample, recorded under DNP conditions. **a** Proton saturation recovery data, illustrating the short T_{DNP} , fitted to stretched exponential. **b** Initial data points of build-up curve from ^{13}C CP/MAS inversion recovery experiment. Fitted to the standard equation for inversion recovery, indicating a slightly shorter longitudinal relaxation time for the C6/C2_{mod}/C3_{mod} region relative to C1, still ^{13}C $T_1 >> {}^1\text{H}$ T_{DNP}

CP repetitions (Fig. S2 in Supplementary Material). The ^{13}C T_1 relaxation times of cellulosic materials in the solid-state are typically long (Sparman et al. 2019) and that is also the case for the dialcohol cellulose samples studied here under DNP conditions, where ^{13}C T_1 relaxation times are typically longer than ${}^1\text{H}$ T_{DNP} build-up times (Fig. 6). However, it is not always the case that the ^{13}C T_1 relaxation times are long under DNP conditions (low temperature, ~ 100 K). For example, CH_3 groups tends to still rotate rapidly at solid-state NMR DNP conditions (Berruyer et al. 2021; Zagdoun et al 2013), therefore giving fast longitudinal ^{13}C relaxation. Under such circumstances, one needs to pay attention when using

the multiple-contact CP experiment as the peak integrals of the CH_3 signal might get underestimated. It is thus important to have an idea of the ^{13}C T_1 relaxation times, their relation to ${}^1\text{H}$ T_{DNP} build-up times, and also potential variations in ^{13}C T_1 relaxation times for different signals. Variations in ^{13}C T_1 relaxation time might lead to relative differences in the signal integral intensities due to different relaxation decay during the last repetition of the loop in the multiple-contact CP pulse sequence (Fig. 4a). This can be counteracted by adding a final, additional CP step outside the loop in the pulse sequence, as exemplified by (Björgvinsdóttir et al. 2018). For dialcohol cellulose, there is an indication of different ^{13}C relaxation times for the C1 and C6, C2_{mod}, C3_{mod} regions of the MCC 1 sample (Fig. 6b). However, the variations in ^{13}C T_1 relaxation times are in this case small ($\sim 10\%$) and both carbons have long T_1 values (101.6 and 90.6 s), the τ delay used in the CP loop was in this case 1.9 s, so this would lead to signal difference of around 0.2% after the last repetition of the CP loop and should have negligible impact on integral calculations. If for instance the ^{13}C T_1 values would have been 100 and 70 s and the τ delay slightly longer, for instance 5 s, the difference in signal intensity would be around 2.1%. For such scenarios and slightly more extreme ones (larger ^{13}C T_1 differences and longer τ delay) one needs to start to pay attention and a good control of the longitudinal relaxation parameters becomes important. The ^{13}C inversion recovery data (Fig. 6b), shows a poor fitting using either the standard mono-exponential T_1 fitting equation or a stretched exponential function. It is most likely that the fit would improve drastically and give more reliable values of ^{13}C T_1 relaxation times if the data was sampled with a longer variable relaxation delays. Yet, the data should be satisfactory to show that the carbon relaxation times are sufficiently long and that effects of different relaxation times are negligible.

We used the DNP-enhanced multiple-contact CP/MAS experiment to quantify the degree of modification in two categories of dialcohol samples: one originating from beaten wood pulp (samples: Fiber 1–4) and the other from MCC (samples: MCC 1–3). The results from these experiments were compared with conventional CP/MAS experiments performed at room temperature as well as conventional DNP enhanced CP/MAS experiments (Table 1).

Table 1 Normalized integral intensities for room-temperature CP/MAS experiments, DNP-enhanced CP/MAS experiments, and DNP-enhanced multiple-contact CP/MAS experiments. The first column shows type of experiment and sample. Columns two to four show normalized integrals for different spec-

tral regions. Column five shows their sum and average sum for each experiment/sample group. Columns six and seven show degree of modification determined from the NMR experiments, and columns eight and nine degree of oxidation from other methods

Experiment/sample	Spectrum regions and normalized integrals				Degree of modification (%)		Degree of oxidation (%)	
	96.1–112.9 ppm	69.4–96.1 ppm	56.4–69.4 ppm	56.4–112.9 ppm	NMR (56.4–69.4 ppm)	NMR (69.4–96.1 ppm)	UV	titration
<i>CP/MAS (room temp.)</i>								
Fiber 1	1.00	4.03	1.00	6.03	0	–2	–	0
Fiber 2	1.00	3.74	1.24	5.98	12	13	–	13
Fiber 3	1.00	3.49	1.49	5.98	25	26	–	24
Fiber 4	1.00	3.24	1.76	6.00	38	38	–	40
Average				6.00				
<i>DNP CP/MAS</i>								
Fiber 1	1.00	3.99	1.06	6.05	3	0	–	0
Fiber 2	1.00	3.79	1.29	6.08	15	11	–	13
Fiber 3	1.00	3.61	1.58	6.19	29	20	–	24
Fiber 4	1.00	3.24	1.90	6.14	45	38	–	40
Average				6.12				
MCC 1	1.00	3.47	1.63	6.10	32	27	55	–
MCC 2	1.00	3.01	2.27	6.28	64	50	75	–
MCC 3	1.00	2.40	2.76	6.16	88	80	100	–
Average				6.18				
<i>DNP multiple-contact CP/MAS</i>								
Fiber 1	1.0	3.99	1.01	6.0	1	0	–	0
Fiber 2	1.0	3.80	1.23	6.03	12	10	–	13
Fiber 3	1.0	3.49	1.44	5.93	22	26	–	24
Fiber 4	1.0	3.24	1.83	6.07	42	38	–	40
Average				6.01				
MCC 1	1.0	3.41	1.53	5.94	27	30	55	–
MCC 2	1.0	2.98	1.93	5.91	47	51	75	–
MCC 3	1.0	2.34	2.66	6.0	83	83	100	–
Average				5.95				

Table 1 shows, for each type of experiment and group of samples, normalized integrals and degree of modification values calculated from our NMR experiments as well as degree of oxidation values from alternative methods (UV absorbance and titration-based method) where this is available. The spectrum integrals from our NMR experiments (Fig. S3-S20 in Supplementary Material) are shown in columns two to four. They have been normalized such that the C1 region in the 96.1–112.9 ppm range is set to one. The

values in the third column represent the normalized integral for the 69.4–96.1 ppm region, containing the signals from the C2, C3, C4 and C5 carbons in non-modified cellulose rings, as well as C4 and C5 from modified ones. The fourth column represents the integrals for the 56.4–69.4 ppm region where the signals from C6, C2_{mod} and C3_{mod} can be found, i.e. signals from carbon atoms with primary hydroxyl groups. Column five shows the sum of the integrals for these three regions, i.e. the sum of integrals for

all carbon atoms, C1, C2, C2_{mod}, C3, C3_{mod}, C4, C5 and C6. This is the sum that should take the value of six, to represent the six carbon atoms in the molecule, since the integrals have been normalized to the well-resolved C1 signal that originates from one carbon atom. The degree of modification values calculated from our NMR experiments, presented in columns six and seven have been calculated from the additional area in the 56.4–69.4 ppm region due C2_{mod} and C3_{mod} (column six) and from the loss of area in the 69.4–96.1 ppm region, due loss of signals from non-modified C2 and C3 (column seven).

If CP-transfer behavior is equal for all carbons in the molecule, the degree of modification values calculated from these two different spectral regions should be the same. If on the other hand, there are differences in ¹H-¹³C dipolar coupling between the different sites in the molecule, or other differences such as variations in T_{1ρ} relaxation, they might differ. The quality of the data (Table 1) and CP-transfer behavior can thus be assessed by examining the magnitude of the integrals from these spectral regions, how well they sum up to six and which degree of modification values that can be calculated from them.

For the first group of experimental data, that is the CP/MAS room temperature data collected at 600 MHz field for the Fiber 1–4 samples, the integrals sum up very close to six. The degree of modification values calculated from the 56.4–69.4 ppm region (column six) agrees well with the corresponding values calculated from the 69.4–96.1 ppm region. In addition, these degree of modification values from our NMR experiments also agree well with the degree of oxidation values from the titration-based method, found in the rightmost column. We interpret this as, that under room temperature CP/MAS conditions, proton to carbon transfer conditions must be equivalent between the different sites in the cellulose molecule and that this yields ¹³C CP/MAS spectra that can be used to reliably quantify degree of modification. The drawback of these conventional CP/MAS experiments are that they take several hours to record (the spectra underlying the room temperature data for the Fiber 1–4 samples in Table 1, were recorded in slightly more than eleven hours).

For the second and the third groups of experimental data in Table 1, that is the DNP CP/MAS data for the Fiber 1–4 and MCC 1–3 samples, the situation is different. Here, in general the sums of integrals

exceed six and there is a larger discrepancy between the degree of modification values calculated from the 56.4–69.4 and 69.4–96.1 ppm regions (column six and seven). The degree of modification values calculated from 69.4–96.1 ppm region i.e., from the reduction of the C2 and C3 signals are lower, and for the Fiber 1–4 samples tend to agree better with the room temperature and titration data. The values calculated from the 56.4–69.4 ppm region, from the additional signal of the C2_{mod} and C3_{mod} carbons are on the other hand higher and deviate more for the Fiber 1–4 samples when compared to room temperature NMR data or titration data. For the MCC 1–3 samples the degree of modification values seem to agree better with the degree of oxidation values from the UV-method when calculated from the addition of signal in the 56.4–69.4 ppm region. Our explanation and interpretation of these observations are, that under DNP conditions (low temperature and cellulose swelled in radical solution), the carbons with primary hydroxyl groups are in better contact with their respective dipolar coupled protons. This leads to a slightly higher CP-transfer efficiency for the C6, C2_{mod} and C3_{mod} positions in the molecule under DNP conditions that in the CP/MAS spectra manifests itself as larger signals in the 56.4–69.4 ppm region and subsequent overestimation of the degree of modification. This was also exemplified previously (Fig. 5) where it could be seen that especially the signal in the 56.4–69.4 ppm was larger in the CP/MAS spectrum compared to the multiple-contact CP/MAS spectrum.

In the lower part of Table 1, the data for the DNP enhanced multiple-contact CP/MAS experiments of the Fiber 1–4 and MCC 1–3 samples can be found. The sums of integrals (column five) for these samples are now again closer to six. One can also see that integrals for the 56.4–69.4 ppm region in column four are reduced for all samples. We interpret this as, that the multiple-contact CP experiment under DNP conditions works and does what it is supposed to do. That is, making sure that the ¹³C signals of all carbon nuclei in the sample reach their maximum (or at least very close to) possible values. This removes the uncertainties or skewness in peak integrals associated with no or too few CP-repetitions and due to different CP-transfer characteristics and give spectra that are more accurate for quantification.

Based on the observations of the behavior of our dialcohol cellulose samples during conventional, and

DNP enhanced CP/MAS and multiple-contact CP/MAS experiments, and the associated data (Table 1), one can summarize like this: The degree of modification values from the room temperature CP/MAS spectra for the Fiber 1–4 samples are likely accurate, but each experiment required about eleven hours of acquisition time. So realistically, including optimization of CP transfer conditions, relaxation delays etc. one can perhaps analyze one dialcohol cellulose sample per day using conventional CP/MAS NMR. If DNP enhanced CP/MAS spectra or too few CP repetitions in multiple-contact experiments are used for quantification, experimental times will be short (minutes instead of ~half an hour), but degree of modification values are likely to be inaccurate, due to differences in CP-transfer characteristics. Usage of the multiple-contact CP experiment under DNP conditions with sufficient number of CP-repetitions seems to improve the results and almost reestablishes the accuracy in the integrals to those observed for the room temperature experiments. Under such circumstances the degree of modification can be quantified, quickly and accurately, and one can analyze a handful of dialcohol samples in a day.

The results from our NMR experiments and the degree of modification values calculated thereof (Table 1) are highly sensitive to spectral processing procedures. Small differences in baseline correction settings or phasing options that are almost not visible to the eye, will affect peak integrals such that the degree of modification values can vary with several percent for different settings. This is important to know, and that exactly the same processing options might not be possible in different NMR software, such as Topspin and MestreNova and this will affect the values. We used MestreNova for our processing, mainly because it is capable of handling both Varian and Bruker data and offers automatic baseline correction options. But this sensitivity to processing parameters and that it opens up for some user-dependent subjectivity, is good to be aware of.

In addition to being sensitive to the processing parameters, it should also be addressed that for the samples in which DMSO-d₆ were used in the radical solution and spectra were acquired at MAS 5.5 kHz, the first sideband of DMSO ends up close to 95 ppm in the spectrum. This is just in between C1 signal and the C4 signal and can in theory affect the computed integrals and resulting degree of modification values

to some extent. The signals from the dialcohol cellulose were however typically ~300 times larger than the DMSO sidebands, it was thus considered to have negligible impact on the calculated values and of no concern. The reason for using this somewhat suboptimal MAS rate was temporary rotor spinning stability issues, and it was then used for all Fiber samples for consistency reasons. Another important aspect of our data is that the room temperature data for the Fiber 1–4 samples was collected on a 600 MHz Varian instrument whereas the DNP data was acquired on newer 400 MHz Bruker Ascend DNP-system. The spectral regions used for calculating the degree of modification in Table 1 were to a high extent chosen based on the 600 MHz data for the Fiber 1–4 samples. The broadening associated with the low temperatures and radicals used during the DNP experiments, combined with the slightly lower resolution of the 400 MHz spectrometer for the DNP experiments makes the spectral regions used, slightly suboptimal for the DNP data. As may be seen (Fig. S3–S20 in Supplementary data) for some of the DNP spectra the up-field and down-field edges of the signals are cropped of by these more narrow integration regions, this however has negligible impact on the results and for the work presented here we have kept these integration regions.

Selective dipolar recoupling experiments in dialcohol cellulose

In the previous section we showed how the DNP-enhanced multiple-contact CP experiment could be used to quantify degree of modification in dialcohol cellulose. Another possible way of investigating the degree of modification and to characterize chemical shift correlations, is to use dipolar recoupling experiments (Ji et al. 2021; Nielsen et al 2012). Here we have tested use of a selective, 1D dipolar recoupling experiment (Fig. 7a) using the SPC5 (Hohwy et al. 1999) dipolar recoupling pulse sequence element to recouple the C1 and the C2 carbons in dialcohol cellulose. This experiment, investigated on the MCC 2 and MCC 3 samples, yields NMR spectra (Fig. 7b, c) that show correlation signals between the C1 and the two C2 and C_{2,mod} carbons. The spectra (Fig. 7b, c) could be used for quantifying the degree of modification by comparing the integrals belonging to the two C2 correlation peaks, as exemplified in the figure.

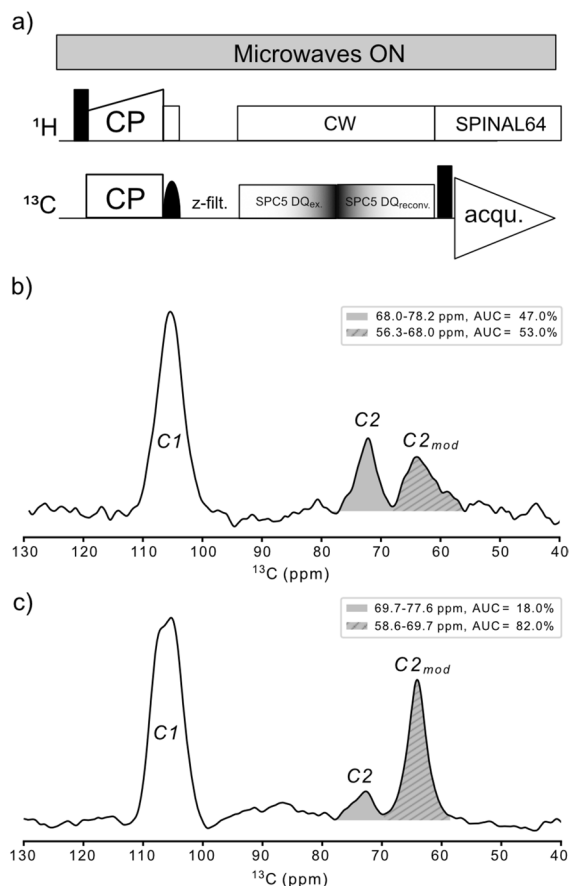


Fig. 7 **a** Selective dipolar recoupling pulse sequence, after CP transfer C1 signal is selected with a selective flip-back pulse and a z-filter, transfer from C1 to C2 and C2_{mod} follows using the SPC5 dipolar recoupling block, typically in the presence of > 100 kHz ¹H CW decoupling. **b** The resulting NMR spectrum using the pulse sequence in a) on the MCC 2 sample, showing the correlation signals of C2 and C2_{mod}. **c** The corresponding spectrum for the MCC 3 sample

The degree of modification values calculated from the spectra (47 and 82%) agree very well with the values calculated from the multiple-contact CP experiment, previously shown in Table 1. Another important aspect of this spectrum is that it can be used to confirm the chemical shifts of C2 (73.4 ppm) and C2_{mod} (65.2 ppm), which are suffering from signal overlap in the ¹³C, 1D spectra previously shown (Figs. 2, 4, 5). In other words, these results clearly show that, in the solid-state, the signal from C2_{mod} overlaps with the chemical shift associated with C6 position in cellulose, just above 60 ppm (Gast et al. 1980; Idström et al. 2016). This type of chemical shift information in

the solid-state is convenient, complementary information to solution-state chemical shift data in HSQC type spectra of dialcohol cellulose, recently published by Simon et al. (Simon et al. 2023).

Although a DNP enhanced, selective dipolar recoupling experiment (Fig. 7) can be used to quantify the degree of modification and obtain chemical shift information, it is a considerably more elaborate experiment than the multiple-contact CP experiment. It also takes longer time to record, the spectrum (Fig. 7b) was recorded with 2 k scans and an acquisition time of around two hours, the spectrum (Fig. 7c) was recorded with 4 k scans in about 5.5 h. We also tested the selective SPC5 experiment for the Fiber 1–4 samples, we then observed a systematically lower degree of modification (data not shown). We have hypothesized if for instance small differences in the proton carbon dipolar coupling environments between the C2 and C2_{mod} positions could play a role in this, or other factors such as differences in relaxation or chemical shift anisotropies due to different cellulosic origins. The selective SPC5 experiment for the Fiber 1–4 samples were also acquired at a lower MAS-rates (5.5 kHz instead of 8 kHz as for the MCC 2 sample, exemplified in Fig. 7). Since the symmetry based SPC5 pulse sequence element uses an RF-field strength proportional to the MAS-rate, we have considered if RF-field inhomogeneities associated with lower field strengths could play a role, but that we would expect to affect the C2 and C2_{mod} signals equally.

Based on this, we believe that the dipolar recoupling experiment show great advantage for qualitative studies of dialcohol cellulose in the solid-state, but also high potential for quantitative determination, once factors affecting ¹³C-¹³C transfer efficiencies in dialcohol cellulose under DNP conditions are better understood. Considering the current uncertainties regarding transfer efficiency, the multiple-contact CP experiment is at this stage the faster, more robust, and user-friendly alternative for quantification of degree of modification in dialcohol cellulose samples.

Conclusion

In this work we have shown that DNP-enhanced solid-state NMR is a suitable technique for analyzing dialcohol cellulose (fibrous and non-fibrous).

Dialcohol cellulose is a material that easily swells in H₂O/D₂O- or DMSO-containing DNP radical solutions and observed NMR signal enhancements are high, allowing for short acquisition times. We have illustrated how the multiple-contact CP experiment can *and is* recommended to be used in order to quantify degree of modification under DNP-enhanced solid-state NMR conditions. We have also exemplified how DNP enhanced, dipolar recoupling experiments can be used for both chemical shift studies and quantification purposes in dialcohol cellulose. Overall, we believe that DNP-enhanced multiple-contact CP experiments should constitute an attractive option for researchers and industry interested in quantifying degree of modification in dialcohol cellulose samples and related cellulosic materials in a fast manner.

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Author contributions HK, LE, PL, AB, AL, and LS contributed to the idea and progress of the project. HK and LS performed the NMR experiments. PC and RN synthesized the samples originating from MCC, PL synthesized the samples originating from Fibers. LE, AP, SS and LK have contributed with continuous support and advice on NMR related matters and cellulose chemistry. HK has been managing the writing process of the manuscript.

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Data availability NMR data is available on <https://doi.org/10.5281/zenodo.13341685>.

Declarations

Conflict of interest The authors declare no competing interests.

Ethics approval and consent to participate Not applicable.

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